Amphiphilic Approach for Preparing Homopolyrotaxanes of Poly(ethylene oxide)

Coleen Pugh,*,† Jin-Young Bae,† Jill R. Scott,‡ and Charles L. Wilkins‡

Department of Chemistry, Macromolecular Science and Engineering Center, University of Michigan, Ann Arbor, Michigan 48109-1055, and Department of Chemistry, University of California, Riverside, California 92521

Received August 11, 1997; Revised Manuscript Received October 20, 1997[⊗]

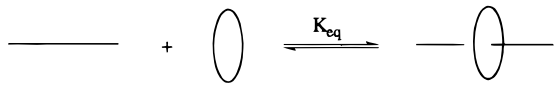
ABSTRACT: A new concept was introduced for preparing homopolyrotaxanes that lack an enthalpic driving force for threading, and which are therefore truly a physical entrapment of non-interacting cyclic molecules and linear polymer chains. It is based on the ability of amphiphiles to form lamellae and columnar micelles in solvents that selectively solvate one of its components. An amphiphilic macrocrown ether (MC-12) with an average ring size of 42 atoms attached to a hydrophobic 12-carbon tail was prepared by formation of the macrocycle in the presence of multiple potassium template ions under pseudo-high dilution conditions. MC-12 is almost insoluble in aliphatic hydrocarbon solvents, but forms micelles in benzene and toluene in the presence of water. The micellar solutions of MC-12 macroscopically phase separate if water-soluble threads are added, but remain intact and solubilize less-water-soluble poly(ethylene glycol) (PEG) derivatives. PEG3350−bis(amine) threads do not react, or react incompletely, with *tert*-butyltrityl end-capping agents functionalized with benzaldehyde and acid chloride groups, respectively. The optimum end-capping reaction for PEG−bis(amine) threads in the water-induced micellar solutions of MC-12 is the addition reaction with 2-*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxymethyl-4,4-dimethylazlactone.

Introduction

A rotaxane is composed of a single cyclic compound threaded with a linear molecule,[1] whereas polyrotaxanes are composed of multiple cycles threaded with a linear polymer.[2] The impetus for the synthesis of polyrotaxanes is their predicted unusual physical and chemical properties resulting from the lack of covalent bonds between the cyclic and linear components. Rotaxanes and polyrotaxanes are most successfully synthesized when the cycle and chain form specific interactions, which are exothermic, and therefore result in a high equilibrium constant for threading (Scheme 1). In addition to direct complexation of the macrocycle and the linear components,[3] threading may be directed by metal complexation with either the monomer/polymer, macrocycle, or both.[4] Threading of cyclodextrins using hydrophobic interactions, which involve both an enthalpic and entropic component, is also high or even quantitative.[5] If the macrocyclic and linear components interact only weakly or not at all ($\Delta H = 0$), the entropic term in Scheme 1 determines the equilibrium, and threading is therefore statistical. In this case, the only current method for driving the equilibrium toward the threaded structure is to use a high concentration of the macrocycle, preferably as solvent, according to Le Chatelier's principle. Nevertheless, the low threading efficiency is rarely satisfactory for polyrotaxane synthesis. Consequently, although polyrotaxanes are intriguing because of the noncovalent interaction between the cyclic and linear components, only heteropolyrotaxanes with extremely strong interactions between the two components are readily accessible.

Using homopolyrotaxanes of poly(ethylene oxide) (PEO) as the prototype, we are developing an amphiphilic route for synthesizing homopolyrotaxanes that

Scheme 1. Equilibrium between the Threaded Structure and Linear and Cyclic Components of Rotaxanes



$$\ln K_{eq} = \frac{\Delta S^\circ}{R} - \frac{\Delta H^\circ}{RT}$$

are truly a physical entrapment of non-interacting cyclic molecules and linear polymer chains. As outlined in Scheme 2, an amphiphilic macrocycle should form lamellae and columnar micelles in solvents that selectively solvate its linear tail. For example, if a hydrophobic alkyl tail is attached to a hydrophilic macrocrown ether, it should aggregate into reverse micelles in hydrocarbon solvents due to selective solvation of the hydrophobic tails. If hydrophilic PEO, which has limited solubility in hydrocarbon solvents, is then added to the organized solution of the micelles, it will be forced into their interior, thereby threading the crown ethers. Threading will be driven by both the high concentration of macrocycles and their optimum alignment, as well as by the increase in entropy which results from dilution of the macrocycles with linear thread.

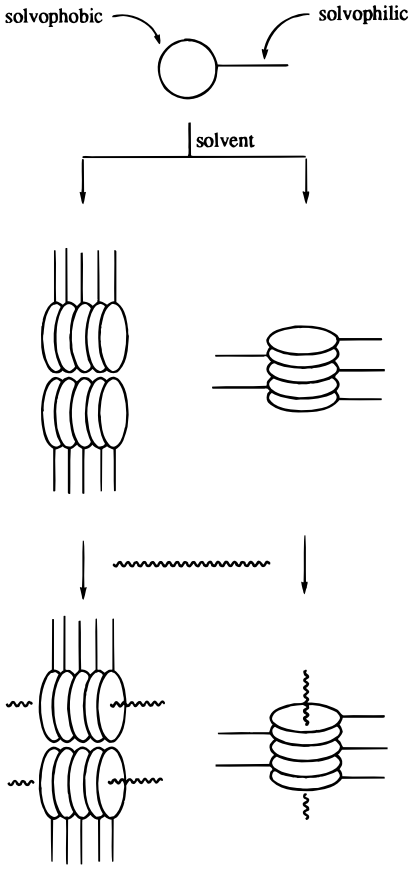
The extent of threading macrocrown ethers (15, 30, 44, or 58 atoms in the ring) with polyethylene glycols (PEG400, PEG600, PEG1000) under statistical conditions was previously determined by "freezing" the rotaxane by chain extension with naphthalene-1,5-diisocyanate and extracting out unthreaded macrocrown ether.[6] In this system, threading increased significantly with increasing ring size, especially when comparing the 30- and 44-membered rings. We have therefore initiated this study using "3,4-(42-crown-14)benzyl dodecyl ether," which is a hydrophilic macrocrown ether with an average ring size of 42 atoms functionalized with a hydrophobic 12-carbon tail.

"3,4-(42-Crown-14)benzyl dodecyl ether" (MC-12) is similar to classic linear oligo(oxyethylene) alkylphenyl

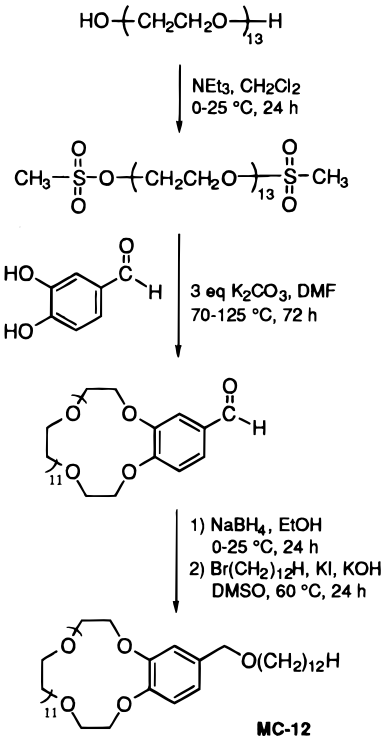
* To whom correspondence should be addressed.
† University of Michigan.
‡ University of California.
⊗ Abstract published in *Advance ACS Abstracts,* December 15, 1997.

S0024-9297(97)01215-1 CCC: $14.00

Scheme 2. Amphiphilic Approach for Preparing Homopolyrotaxanes



Scheme 3. Synthesis of 3,4-(42-Crown-14)benzyl Dodecyl Ether (MC-12)



ether surfactants,[7] such as Igepal-CO-720 (oligo[oxyethylene] nonylphenyl ether),[8] with a single oligo(oxyethylene) segment that contains a distribution of the number of oxyethylene units. MC-12 also contains a distribution of ring sizes corresponding to that of "monodisperse" poly(ethylene glycol) 600 (PEG600; 600 g/mol) from which it was synthesized (Scheme 3). However, it contains approximately 17% of double ring sizes formed in the cyclization step. Since any commercial application of either homopolyrotaxanes or this amphiphilic approach to their synthesis will preclude elaborate separations and purifications, this paper will demonstrate that MC-12, with its distribution of ring sizes, forms micelles. It also explores the stability of these micellar aggregates in the presence of PEG derivatives with different end groups, and the synthesis and reactions of three potential end-capping agents. This leads to a recommended procedure, which we are pursuing, for the synthesis of homopolyrotaxanes of MC-12 and poly(ethylene oxide) using our amphiphilic approach.

Results and Discussion

Synthesis and Solution Behavior of "3,4-(42-Crown-14)benzyl Dodecyl Ether" (MC-12). Scheme 3 outlines the synthesis of MC-12 starting from PEG600, which was first converted to the corresponding bis(methanesulfonate ester) according to a literature[9] procedure. The most difficult step in the synthesis is the cyclization step. Since ring-closure is a first-order condensation reaction and chain extension is second order, cyclization is favored over chain extension and/or formation of larger rings under high dilution conditions;[10] pseudo-high-dilution conditions can be maintained by adding at least one of the reactants to the reaction mixture at a slower rate than the rate of reaction. In addition, the yields of large crown ethers with 27−60 atoms are increased by using multiple potassium or sodium ions as template ions that hold the oligo(oxyethylene) in a more folded conformation.[11] We therefore synthesized the benzaldehyde precursor of MC-12 by reacting 3,4-dihydroxybenzaldehyde with PEG600−bis(mesylate) in the presence of multiple potassium template ions (K_2CO_3 as base) under pseudo-high-dilution conditions. PEG600 was functionalized with mesylate end groups in order to follow their displacement in the cyclization step using ^{1}H-NMR by loss of the $-CH_2OMs$ resonance at 4.35 ppm; displacement of tosylate end groups was difficult to quantify using ^{1}H-NMR since the $-CH_2OTs$ resonance of the reactant at 4.13 ppm overlaps the $-CH_2OAr$ resonance of the product at 4.20 ppm.

The aldehyde group was then reduced using sodium borohydride. The resulting benzyl alcohol was most successfully etherified with *n*-bromododecane in the presence of potassium iodide using potassium hydroxide as base in DMSO at room temperature;[12] the reaction mixture was subsequently heated at 60 °C for 24 h to ensure complete conversion. As is typical of nonionic surfactants,[13] MC-12 is very soluble in polar solvents such as alcohols, but it is either insoluble or much less soluble in solvents with low dielectric constants, such as hydrocarbons and chlorinated hydrocarbons. MC-12 can therefore be purified by introducing it to a silica gel column, flushing the column first with chloroform to remove unreacted starting materials and then isolating MC-12 with methanol.

Figure 1 shows the matrix-assisted desorption/ionization Fourier transform (MALDI−FT)[14] mass spectrum of the MC-12 used in this study. The peak at *m*/*z* 885.5 is the monoisotopic peak of the sodium attached [3,4-(42-crown-14)benzyl dodecyl ether]Na^+; the corresponding inset shows the isotopic distribution of this peak.

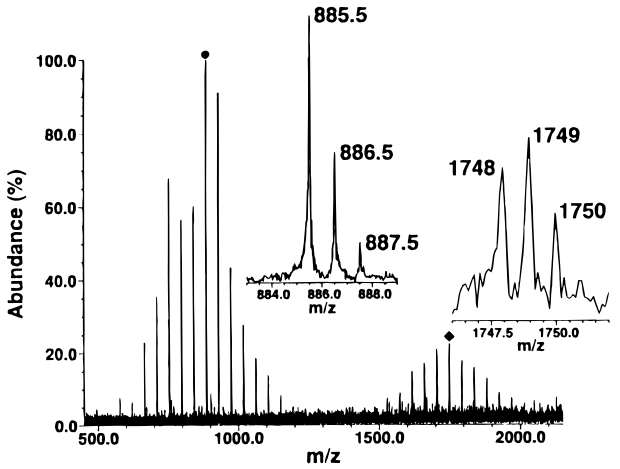


Figure 1. MALDI−FT mass spectrum of MC-12; insets show the isotopic distributions of the peaks labeled ● and ◆.

The molecular weight distribution of the peaks centered at *m*/*z* 885.5 corresponds to $M_n = 840$, $M_w = 855$, and polydispersity pdi $= M_w/M_n = 1.02$; the mass of 3,4-(42-crown-14)benzyl dodecyl ether with exactly 42 atoms in the ring is 863.09 g/mol. As expected,[15] the spectrum also shows that this sample of MC-12 is contaminated with a small amount of the dimeric dibenzocrown ether (Chart 1). The peak at *m*/*z* 1749 is the monoisotopic peak of the sodium-attached cyclic dimer; the corresponding inset shows the isotopic distribution of this peak. The molecular weight distribution of the peaks centered at *m*/*z* 1749 corresponds to $M_n = 1693$, $M_w = 1700$, and polydispersity pdi = 1.004; the mass of the dimer of 3,4-(42-crown-14)benzyl dodecyl ether with exactly 84 atoms in the ring is 1726.2 g/mol. Although MALDI-MS is not quantitative for broad polydispersities,[14,16] we can use the spectrum shown in Figure 1 to roughly estimate that the composition of MC-12 is 83% monomeric 3,4-(42-crown-14)benzyl dodecyl ether, and 17% cyclic dimer. Throughout the rest of this paper, however, all calculations will use the molecular weight of MC-12 corresponding to PEG600 with 13.2 repeat units (872.42 g/mol).

MC-12 is almost insoluble in hydrocarbon solvents such as hexane and heptane. However, it is soluble in benzene and toluene and forms homogeneous solutions. Nevertheless, micellar aggregation occurs by adding small amounts of water to these homogeneous solutions. This is evidently because water hydrogen bonds to the macrocrown ether, thereby decreasing its solubility in aromatic solvents. Similarly, oligo(oxyethylene) alkyl ether surfactants aggregate in benzene[7] if water is added but form only isotropic solutions in this solvent in the absence of water. The water-induced aggregation of MC-12 is obvious both visually and experimentally. For example, the transparent solutions of MC-12 in benzene and toluene become turbid when water is added. In addition, since light scattering is proportional to the molecular weight of the solute, the scattering intensity of MC-12 in toluene or benzene should remain constant until the apparent molecular weight increases due to aggregation. Table 1 summarizes the intensity of scattered light at three different angles (as measured by the voltage at each detector) of a 0.362 mM solution of MC-12 in toluene as a function of the concentration of added water. The voltage reading at each detector remains essentially constant up to 51 equiv of added water, but increases significantly once 102 equiv have been added. Therefore, the critical concentration of water for micelle formation is between 4 and 8 equiv of water per oxyethylene (EO) unit. The size of the aggregates increases as the water concentration increases; water is evidently solubilized in the interior of the reverse micelles in the form of water pools.[17]

Figure 2 presents the ^{1}H-NMR spectra of a 75.5 mM solution of MC-12 in toluene-d_8 (6.5 wt % MC-12, 93.5 wt % toluene-d_8) before and after adding D_2O.[18] When D_2O is added, a resonance appears at 4.5−5.0 ppm due to HOD of the solubilized water.[19] In addition, the oxyethylene resonances at 3.5−4.3 ppm broaden. This broadening may be due to the changing environment of the oxyethylene segments, such that the three partially resolved resonances become more chemically inequivalent in the presence of water. The solution with $[D_2O]:[EO] = 2.2$ (6.3 wt % MC-12, 89.5 wt % toluene-d_8) is very turbid and apparently saturated with water, with a minute amount of phase-separated D_2O. These changes are reversed when the micelles are destroyed by heating, and in some cases, are regenerated when the micelles reform upon cooling. For example, Figure 2 shows that if the solution with $[D_2O]:[EO] = 2.2$ is heated to 40 °C, the HOD resonance of the water pool almost disappears, although the solution appears to be more turbid; the oxyethylene resonances sharpen significantly. The water pool reappears and the oxyethylene resonances rebroaden when the solution is cooled back to room temperature. However, if this solution is heated at 70 °C for 24 h and then cooled back to room temperature, it remains macroscopically phase separated into two layers; the toluene layer is transparent. In this case, the HOD resonance at 5.0 ppm completely disappears, and the oxyethylene resonances at 3.5−4.3 ppm are as sharp as those without any added water. Although this solution remains macroscopically phase separated at room temperature, it becomes turbid and macroscopically homogeneous if it is subsequently agitated by stirring or shaking.

In contrast to water, neither lithium chloride, which remains insoluble, nor ethylene glycol induce micelle formation of MC-12 in toluene or benzene. When 1.6−5.4 equiv of ethylene glycol are added to a 96 mM solution of MC-12 in toluene-d_8 (8 wt % MC-12, 92 wt % toluene-d_8), it becomes slightly turbid, but then macroscopically phase separates. Ethylene glycol solutions of amphiphiles are also rarely lyotropic, in contrast to the corresponding aqueous solutions.[20] Similarly, addition of ethylene glycol to aqueous surfactant solutions increases their critical micelle concentrations.[21]

Synthesis of Functionalized Poly(ethylene glycol) Threads and Their Effect on the Solution Behavior of MC-12. Since MC-12 forms reverse micelles and/or reverse lyotropic mesophases in toluene and benzene if small amounts of water are added, these systems can be used for threading experiments. The PEG threads must be functionalized with end groups that can be reacted with bulky blocking groups to prevent dethreading. Scheme 4 outlines the synthesis of the derivatives we tested that are not commercially available. All of these derivatives were synthesized by reacting PEG-bis(mesylate) with a nitrogen- or oxygen-based nucleophile.

Table 2 summarizes the effect of adding various PEG derivatives to a 76 mM solution of MC-12 in toluene-d_8 in the presence of D_2O, and Figure 3 presents representative ^{1}H-NMR spectra of the three outcomes. If either PEG600 or PEG600−bis(mesylate) are added to this micellar solution, the solution macroscopically

Chart 1

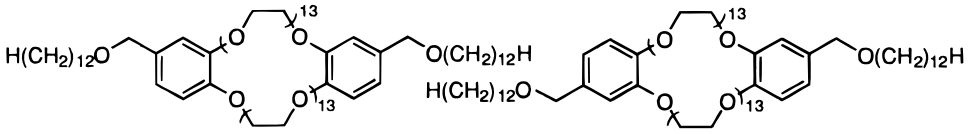


Table 1. Light (633 nm) Scattering Intensity of a Dilute Solution of MC-12 in Toluene[a]

H_2O added (μL)[a]	$[H_2O]$/ [MC-12]	$[H_2O]$/ [EO]	normalized average voltage (V) 48° detector	90° detector	132° detector
0	0	0	0.165	0.0573	0.0451
1	10.2	0.771	0.185	0.0589	0.0472
5	51.0	3.86	0.205	0.0704	0.0575
10	102	7.71	0.950	0.494	0.381
50	510	38.6	0.946	1.69	1.01

[a] Water added to 0.362 mM solution (15 mL) of MC-12 in toluene; 0.036 wt % MC-12.

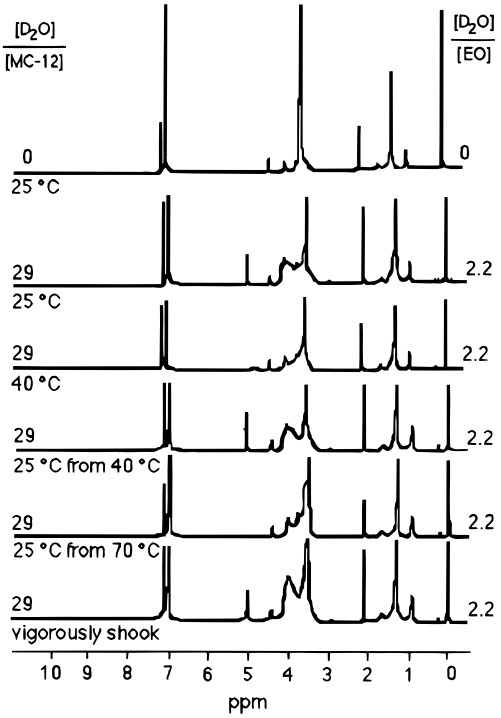


Figure 2. ^{1}H-NMR spectra of 76 mM solutions of MC-12 in toluene-d_8 (6.5 wt % MC-12/93.5 wt % toluene-d_8) as a function of the concentration of added D_2O.[18]

phase separates into a turbid upper layer and a very small isotropic layer. Figure 3a shows that the intensity of the HOD resonance at 4.5−5.0 ppm is very small. The $-OSO_2CH_3$ resonance of the thread at 2.6 ppm is also completely absent, even after adding 1.5 equiv (relative to MC-12). That is, the water-soluble PEG600 and PEG600−bis(mesylate) threads phase separate from the toluene solution of MC-12 into a distinct aqueous layer. Although the amount of D_2O in the toluene layer of MC-12 decreases with this phase separation, a small amount remains and the solution is still turbid. A corresponding isotropic solution of MC-12 and PEG600−bis(mesylate) also phase separates into a turbid upper layer and a very small transparent layer after adding one drop of D_2O.

MC-12 is also soluble in water, although much less so than either PEG600 or PEG600−bis(mesylate). If the PEG derivatives are less soluble in water, such as low molecular weight derivatives with hydrophobic end groups and higher molecular weight derivatives with hydrophilic end groups, they do not phase separate from the micellar solutions. However, PEG3400, which is water-soluble and toluene-insoluble, is solubilized by the micelles but slowly precipitates out of solution after 24 h. The turbidity of these MC-12 solutions either remains essentially constant or actually increases when the thread is added. Figure 3b shows the ^{1}H-NMR spectrum of a system in which the thread does not affect its turbidity; Figure 3c shows the ^{1}H-NMR spectrum of a system in which the thread increases its turbidity. There are two significant differences between these two type of spectra. In the spectrum with added *p*-H_2NPhO−PEG600−OPhNH_2, the internal oxyethylene resonances of the thread appear at 3.5 ppm, which is their normal position in toluene-d_8 and which overlaps those of the internal oxyethylene resonances of MC-12. In contrast, the internal oxyethylene resonances of PEG3350−bis(amine) (−O_2CNH$(CH_2)_2NH_2$ end groups) are shifted downfield from 3.5 to 4.1 ppm. We believe that this downfield shift indicates that the PEG thread is in the interior of the micelle and, if there is no shift, that the thread is in the surrounding toluene solvent. (1,13-Dichloro-3,6,8,11-tetraoxotridecane [CH_2(OCH_2-$CH_2OCH_2CH_2Cl)_2$] is apparently not solubilized in the interior of the micelles either since it behaves similarly to *p*-H_2NPhO−PEG600−OPhNH_2.)

In addition, the HOD resonance of the *p*-H_2NPhO−PEG600−OPhNH_2 system shifts slightly upfield and is only slightly broader than that in its absence, whereas that of the PEG3350−bis(amine) system also shifts upfield slightly but is much broader, and is apparently split into two broad peaks. The water proton(s) are apparently exchanging slowly on the ^{1}H-NMR time scale with the −N*H*− end groups of the threads; the spectrum with PEG−bis(glycidyl ether) shows only one broad peak at 4.8 ppm.

Synthesis and Reactions of End-Capping Agents. Trityl end groups are sufficiently large to prevent dethreading of rings with up to 29 atoms,[22] whereas *tert*-butyltrityl end groups block rings with up to 42 atoms.[23] Therefore, *tert*-butyltrityl end groups will prevent dethreading of 3,4-(42-crown-14)benzyl dodecyl ether, but not its cyclic dimer, and can therefore be used to remove the cyclic dimer from the polyrotaxanes of MC-12 threaded with poly(ethylene oxide). In this amphiphilic approach for preparing homopolyrotaxanes, any potential end-capping reaction involving *tert*-butyltrityl derivatives must tolerate the water that is used to induce micelle formation. Scheme 5 outlines the three end-capping reactions that we have investigated. All of the reactions use a PEG−bis(amine) thread reacted with either an aldehyde to form an imine bond, an acid chloride to form an amide bond, or an azlactone to also form an amide bond.

End-Capping PEG−Bis(amine)s with [*p*-[*p*-Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde. Table 3 summarizes the results of model reactions of benzaldehyde with aliphatic amines in toluene-d_8 or benzene-d_6 at room temperature; these experiments were performed in NMR tubes without stirring. As demonstrated by the first entry, benzaldehyde reacts quantitatively with a slight excess of

Scheme 4. Synthesis of Poly(ethylene oxide) Threads with Amine End Groups

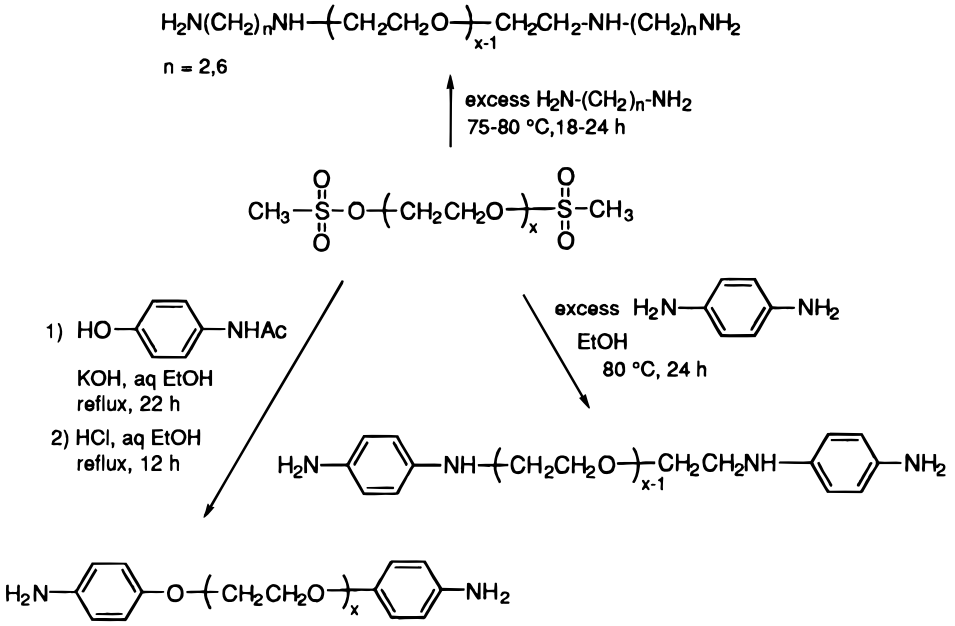


Table 2. Effect of Adding Derivatized PEG Threads to MC-12 Aggregates in Toluene-d_8[a]

PEG thread					
	theoretical[b]				
end groups	DP_n	M_n	turbidity	macroscopic homogeneity	^{1}H-NMR resonance of internal EO
−OH	13.2	600	decreased	2 phases	−[c]
−OMs	13.2	766	decreased	2 phases	−[c]
$-OC_6H_4NH_2$	13.2	782	no change	1 phase	3.5
$-NH(CH_2)_2NH_2$	13.2	684	increased	1 phase	4.1
$-NH(CH_2)_6NH_2$	13.2	796	increased	1 phase	4.1
−OH	76.8	3400	increased	1 phase	4.1
$-NH(CH_2)_6H$	75.6	3516	increased	1 phase	4.1
$-O_2C-NH(CH_2)_2NH_2$	75.6	3522	increased	1 phase	4.1
$-CH_2CH(O)CH_2$	75.6	3462	increased	1 phase	4.1

[a] PEG threads were added to a 76 mM solution of MC-12 in toluene in the presence of D_2O; 6.4 wt % MC-12/2.8 wt % D_2O/90.8 wt % toluene-d_8; $[D_2O]:[EO_{MC-12}] = 1.5$. [b] Based on synthesis from PEG600 ($DP_n = 13.2$) or PEG3350 ($DP_n = 75.6$). [c] An aqueous solution of the thread macroscopically phase separates from the toluene solution of MC-12.

n-hexylamine in the micellar solution of MC-12 in toluene-d_8. Similarly, it reacts almost quantitatively (72%) with PEG3350−bis(amine) ($-O_2CNH(CH_2)_2NH_2$ end groups) in toluene-d_8 in the absence of MC-12 (entry 2). However, there is essentially no reaction between benzaldehyde and PEG3350−bis(amine) in the micellar solution (entry 3). This indicates that although PEG3350−bis(amine) is solubilized by the micelle, its end groups are buried within the micelle and unable to react with benzaldehyde in the surrounding solvent. In contrast, the more hydrophobic end groups of $H_2N(CH_2)_6$-NH−PEG600−$NH(CH_2)_6NH_2$ are apparently accessible to benzaldehyde in the aromatic solvent, with 69% conversion after 10 h at room temperature (entry 4).

Since benzaldehyde reacts with PEG−bis(amine) in the micellar solution of MC-12 when the end groups are hydrophobic, we synthesized the corresponding bulky end-capping agent. As outlined in Scheme 6, *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzaldehyde was synthesized by reacting tris(*p*-*tert*-butylphenyl)(4-hydroxyphenyl)methane with 4-(bromomethyl)benzonitrile in the presence of NaOH, followed by reduction[24] of the cyano group of the resulting *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzonitrile with diisobutylaluminum hydride. Tris(*p*-*tert*-butylphenyl)(4-hydroxyphenyl)methane was synthesized according to a literature procedure.[25]

Similar to the model reactions with benzaldehyde, 52% of *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzaldehyde is converted to the imine in 18 h in benzene-d_6 at room temperature by reacting it with a stoichiometric amount of $H_2N(CH_2)_6NH$−PEG600−NH-$(CH_2)_6NH_2$ in the absence of MC-12 and water. However, the imine bond formed by reaction of $H_2N(CH_2)_6$-NH−PEG600−$NH(CH_2)_6NH_2$ with either benzaldehyde or *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde is easily hydrolyzed back to the starting materials, especially when purification is attempted by column chromatography on silica gel. Since Schiff-base imines, which are substituted with an aromatic group at nitrogen, are much more difficult to hydrolyze than imines with an alkyl substituent,[26] we synthesized *p*-NH_2PhNH−PEG600−$NHPhNH_2$ as shown in Scheme 3. In this case, the amine groups of *p*-NH_2PhNH−PEG600−$NHPhNH_2$ react quantitatively with 1.5 equiv of *p*-[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde in toluene within 24 h at room temperature. However, 31% of the imine bonds hydrolyze back to amine groups when purified by column chromatography (see Experimental Section) and are therefore more unstable than typical Schiff bases. For example, we found that under the same conditions, only a small amount of benzylideneaniline is hydrolyzed on a silica gel column using methanol or CH_2Cl_2 as eluant. In

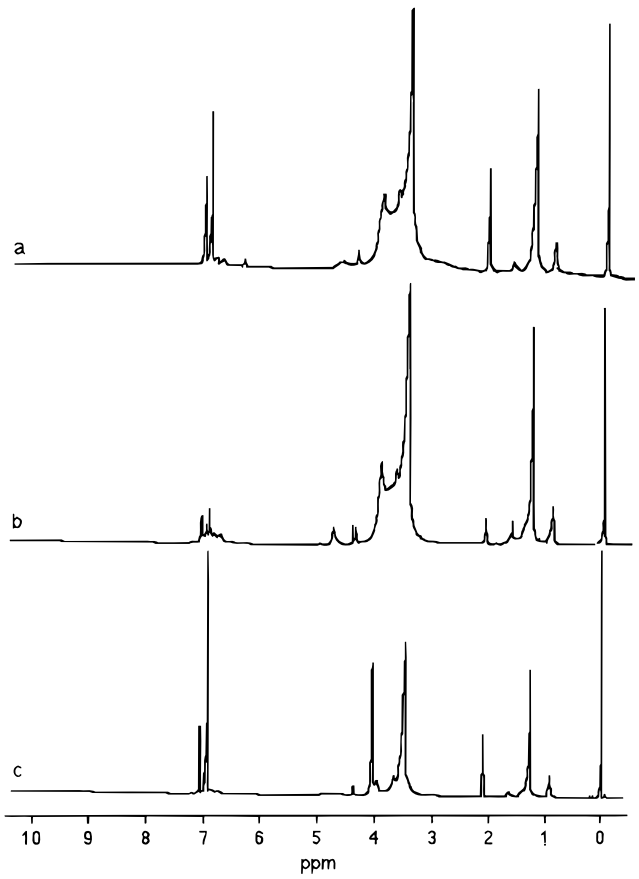


Figure 3. Representative ^{1}H-NMR spectra of a 76 mM solution of MC-12 in toluene-d_8 in the presence of D_2O ([D_2O]:[EO] = 1.5, 6.4 wt % MC-12/2.8 wt % D_2O/90.8 wt % toluene-d_8) after adding 0.051−1.5 equiv of derivatized PEG threads: (a) 1.5 equiv of PEG600−bis(mesylate); (b) 0.30 equiv of *p*-NH_2-PhO−PEG600−OPhNH_2; (c) 0.10 equiv of PEG3350−bis(amine) ($-O_2CNH(CH_2)_2NH_2$ end groups).

addition, hydrolysis is barely detectable when the silica gel is pretreated with 5% NH_4OH in methanol, and is nonexistent on basic activated alumina.

Although the imine bond of the end-capped thread is more hydrolytically unstable than that of typical Schiff bases, it is readily reduced to the stable amine using $NaBH_3CN$ (see Experimental Section).[27] However, we did not work with this system further because it has the disadvantage that the end-capping reaction generates H_2O. As discussed in the next paper in this series,[8] aggregation and organization of MC-12 in toluene is controlled by the concentration of both MC-12 and water. Therefore, this end-capping reaction in which PEG−bis(amine) threads are reacted with *p*-[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzaldehyde is not optimal because it increases the amount of water in the system, which may change how MC-12 is organized and preclude correlation of the mesophase and threading efficiency.

End-Capping PEG−Bis(amine)s with [*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl Chloride. Since amines react much faster than water and alcohols with acid chlorides, poly(amide)s can be synthesized by interfacial polymerizations in which the diacid chloride and diamine react at the interface of their organic and aqueous solvents, respectively.[28] It may therefore be possible to endcap the PEG−bis(amine) threads with an acid chloride in the water-induced micellar solutions of MC-12. [*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]-acetyl chloride was synthesized by a literature procedure,[25] except that we reacted tris(*p*-*tert*-butylphenyl)(4-hydroxyphenyl)methane with ethyl bromoacetate under phase transfer catalyzed conditions, which simultaneously hydrolyzed the resulting ester to [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid.

The position of the ArOCH_2- ^{1}H-NMR resonance of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride and its derivatives is very sensitive to the group bonded to carbonyl. The product mixture of model end-capping reactions can therefore be monitored by integration of the various ArOCH_2- resonances. Table 4 summarizes the results of model reactions of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride with trityl aniline and PEG−bis(amine)s; $H_2N(CH_2)_6NH$−PEG600−$NH(CH_2)_6NH_2$ threads can not be used because all of the nitrogens react with acid chlorides. In dilute solution, 85% of the amide is produced by stoichiometric reaction of tritylaniline and [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride in the presence of reagent grade triethylamine, and the remaining 15% of the acid chloride is hydrolyzed to the triethylammonium carboxylate (entry 1). Similarly, 51% of the theoretical yield of the amide is obtained from reaction of PEG3350−bis(amine) with excess acid chloride in toluene in the presence of triethylamine, and all of the unreacted acid chloride is hydrolyzed to the acid (entry 2). The yield of the PEG3350−bis(amide) (62%) is slightly higher when triethylamine is not used. In addition, only 42% of the unreacted acid chloride is hydrolyzed (entry 3); 50% of the unreacted acid chloride is hydrolyzed in the reaction of PEG600−bis(hexylamine) in the presence of triethylamine (entry 4). This indicates that triethylamine is not necessary to catalyze the reaction, and if it is, it should be stringently dried in order to prevent hydrolysis of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride.

These results also indicate that [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride is too hydrolytically unstable in these water-induced organized solutions of MC-12 to completely block PEG−bis(amine)s. Although 4-tritylaniline reacts quantitatively with the acid chloride even in the organized solution (entry 5), entries 6−8 confirm that hydrolysis prevents the amidation of the PEG−bis(amine)s from reaching 100% conversion in the organized solutions. Since 4-tritylaniline should be in the solvent surrounding the micelles, the lower yields of entries 6 and 8 support the idea that the PEG−bis(amine) threads are in the interior of the micelles, and that water can more effectively compete with their end groups for reaction with [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride. In contrast to the end-capping reaction with *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy)methyl]-benzaldehyde, the end groups of PEG3350−bis(amine) react with the acid chloride, evidently because [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride is more hydrophilic and penetrates the micelles to a certain extent. Comparison of entries 6 and 7 demonstrates that triethylamine, which forms a more reactive acyl ammonium intermediate, is preferable to 2,6-lutidine, which acts only as base to react with the HCl generated. The reaction in the presence of 2,6-lutidine produces a lower yield of the amide and therefore a higher yield of the hydrolyzed end-capping agent.

Therefore, the primary disadvantage of the end-capping reaction of PEG−bis(amine) threads with [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride is that the latter is hydrolytically unstable under the

Scheme 5. Potential End-Capping Reactions for Blocking PEG−Bis(amine) Threads with Bulky *tert*-Butyltrityl Groups

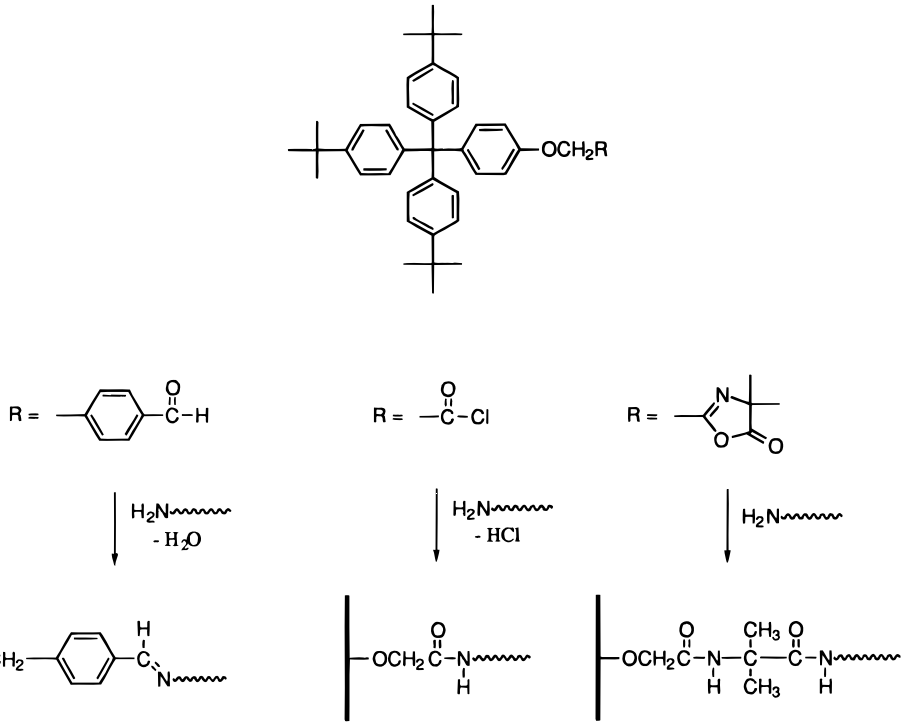
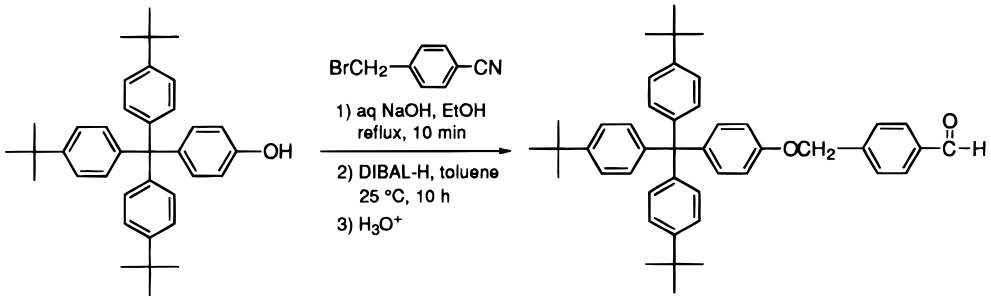


Table 3. Formation of Imines by Reaction of Benzaldehyde with Amines in Toluene-d_8 at Room Temperature

entry	amine	[MC-12] (mM)	[D_2O] (M)	[MC-12]/ [$-NH_2$]	[$-NH_2$]/ [PhCHO]	time (h)	conv of PhCHO (%)
1	$H(CH_2)_6NH_2$	76	1.5	0.53	1.2	22	∼99
2	PEG3350−bis(amine)[b]	0	0	...	0.95	24	72
3	PEG3350−bis(amine)[b]	124	1.1	29	0.96	24	∼1
4[c]	$PEG600[NH(CH_2)_6NH_2]_2$	114	2.2	3.0	1.0	10	69

[a] Reactions performed in NMR tubes without stirring. [b] Poly(ethylene glycol)3350−bis(amine); $-O_2CNH(CH_2)_2NH_2$ end groups made from PEG3350. [c] Reaction perfomed in benzene-d_6.

Scheme 6. Synthesis of *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzaldehyde



conditions that we have used to induce micelle formation. Although we could theoretically use a large enough excess of the acid chloride to completely react with the PEG−bis(amine) end groups, the carboxylic acid generated by hydrolysis is very difficult to remove from the reaction mixture in the presence of MC-12. In addition, this end-capping reaction is not optimal because it generates HCl as a byproduct, and a base must therefore be added to neutralize it. These two additional components may change how MC-12 is organized and preclude correlation of the mesophase with threading efficiency. For example, addition of acid, base, and electrolytes influences the solubilization region of water in any surfactant solution, including those of oligo(oxyethylene) alkylphenyl ethers.[7]

Again in analogy to the synthesis of poly(amide)s, another possibility is to preform an ammonium carboxylate salt, and then to dehydrate the salt at elevated temperature. The results of model dehydration reactions of the salts of PEG3350−bis(amine) and [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid are summarized in Table 5. When a stoichiometric amount of the carboxylic acid and amine end groups are mixed in chloroform in the presence of a protonic solvent, 55% of the carboxylate salt is converted to amide groups by dehydrating at 160 °C for 2 h. Similarly, 82% are converted to amide groups after 3.5 h when the salt is generated in a nonprotonic solvent; this is increased to 93% by a subsequent dehydration step at 180 °C for 3 h. However, when a stoichiometric amount of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid was added to PEG3350−bis(amine) in an organized solution of MC-12 and allowed to react for 5 h before dehydrating, only 22% of the amide groups were formed,

Table 4. Model Reactions of Amines with [*p*-[Tris(*p-tert*-butylphenyl)methyl]phenoxy]acetyl Chloride

entry	amine	solvent	[MC-12] (M)	[−NH_2] (mM)	[−COCl]/ [−NH_2]	[NEt_3]/ [−COCl]	time (h)	% conv of −NH_2 to amide	% unreacted −COCl hydrolyzed
1	4-tritylaniline	$CDCl_3$	0	14.6	0.99	1.0	10	85	100
2	PEG3350−bis(amine)[a]	toluene	0	60.2	1.6	1.0	24	51	100
3	PEG3350−bis(amine)[a]	$CDCl_3$	0	12.0	1.6	0	17	62	42
4	PEG600[$NH(CH_2)_6H]_2$	$CDCl_3$	0	11.4	0.99	1.0	15	61	50
5	4-tritylaniline	toluene, H_2O[b]	0.33	146	1.0	1.0	10	100	. . .
6	PEG3350−bis(amine)[a]	toluene, H_2O[c]	0.38	60.7	1.6	1.0	30	70	100
7	PEG3350−bis(amine)[a]	toluene, H_2O[d]	0.38	60.7	1.6	0.99[f]	24	37	100
8	PEG600[$NH(CH_2)_6H]_2$	toluene, H_2O[e]	0.38	168	1.5	1.0	24	60	100

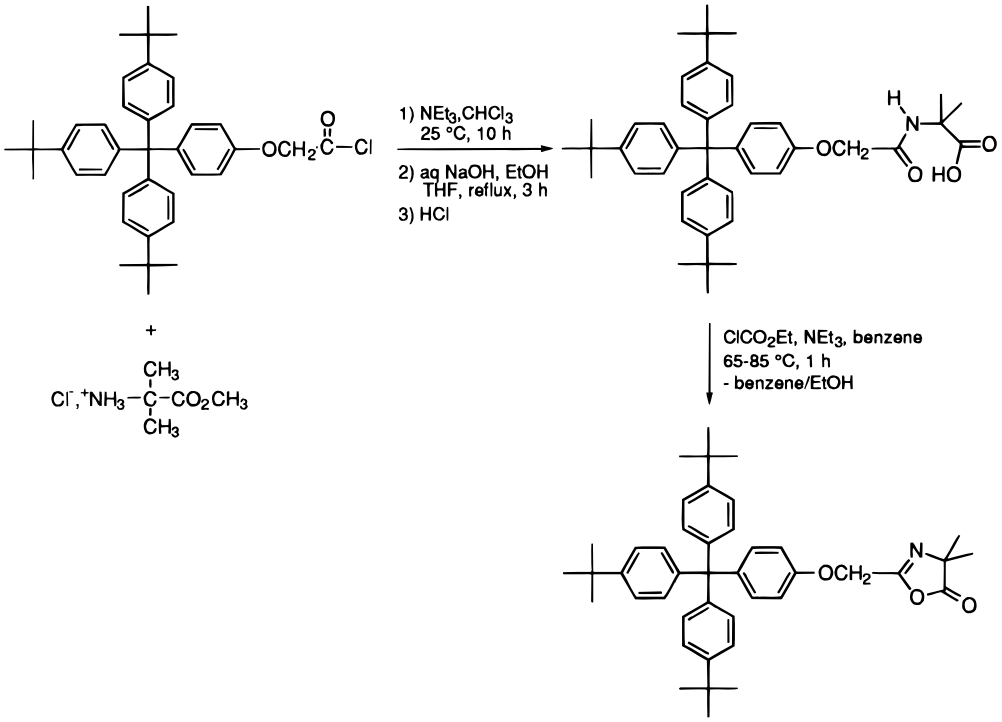
[a] Poly(ethylene glycol)3350−bis(amine); $-O_2CNH(CH_2)_2NH_2$ end groups made from from PEG3350. [b] $[H_2O]/[EO] = 0.73$. [c] $[H_2O]/[EO] = 0.73$. [d] $[H_2O]/[EO] = 9.7$. [e] $[H_2O]/[EO] = 0.73$. [f] 2,6-Lutidine as base instead of NEt_3.

Table 5. Model Reactions of PEG3350−Bis(amine)[a] with [*p*-[Tris(*p-tert*-butylphenyl)methyl]phenoxy]acetic Acid[b]

entry	solvent	[MC-12] (mM)	[−NH_2] (mM)	[acid]/ [−NH_2]	time of second stage reaction (h)	conv to −CONH− (%)
1	$CHCl_3$/EtOH (5:1)	0	4.7	1.1	2	55
2	THF	0	5.7	1.0	3.5	82[d]
3	toluene, H_2O[c]	0.38	60.6	1.1	3	22

[a] Poly(ethylene glycol)3350−bis(amine); $-O_2CNH(CH_2)_2NH_2$ end groups made from PEG3350. [b] After removing solvent(s) from preformed ammonium carboxylate salts, the salts were dehydrated at 160 °C under a nitrogen flow. [c] $[H_2O]/[EO_{MC-12}] = 0.73$. [d] The conversion to amide increases to 93% by heating again at 180 °C for 3 h.

Scheme 7. Synthesis of 2-[[*p*-[Tris(*p-tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone



and 78% of the carboxylic acid was recovered. As confirmed by ^{1}H-NMR, this is because the PEG−bis(amine) end groups were not completely converted to the ammonium carboxylate salt before the dehydration step.

End-Capping PEG−Bis(amine)s with 2-[[*p*-[tris(*p-tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone. In contrast to the condensation reactions of PEG−bis(amine)s with an acid chloride or benzaldehyde derivative, the corresponding reaction shown in Scheme 5 with an azlactone is an addition reaction, and does not generate a small molecule byproduct that could change the balance of components in organized solutions of MC-12. Although the azlactone ring is slowly hydrolyzed by water, it reacts rapidly with both primary and secondary amines at room temperature in the absence of a catalyst,[29] even in dilute aqueous solutions.[30] In contrast, ring-opening of the azlactone with alcohols requires acid or base catalysis or long reaction times.[29] As outlined in Scheme 7, 2-[[*p*-[tris(*p-tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone was synthesized by first N-acylating methyl 2-aminoisobutyrate with [*p*-[tris(*p-tert*-butylphenyl)methyl]phenoxy]acetyl chloride; the hydrochloride salt of methyl 2-aminoisobutyrate was synthesized by esterifying 2-methylalanine with methanol in thionyl chloride according to a literature[31] procedure. *N*-((Methylcarbonyl)isopropyl)[*p*-[tris(*p-tert*-butylphenyl)methyl]phenoxy]acetamide was then hydrolyzed and cyclodehydrated via the carboxylic−carbonic anhydride intermediate formed by reaction with ethyl chloroformate in the presence of triethylamine.

Table 6. Model Reactions of Water and Amines with 2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone at Room Temperature

entry	nucleophile	solvent	[nucleophile]/[azlactone]	time (h)	% ring-opened azlactone[a]
1	D_2O	$CDCl_3$	67	18	28
2	$H(CH_2)_6NH_2$	toluene-d_8	6.2	0.17	100
3	PEG3350−bis(amine)[b]	toluene-d_8	0.40	10	100

[a] Based on limiting reagent. [b] Poly(ethylene glycol) 3350−bis(amine); $-O_2CNH(CH_2)_2NH_2$ end groups made from PEG3350.

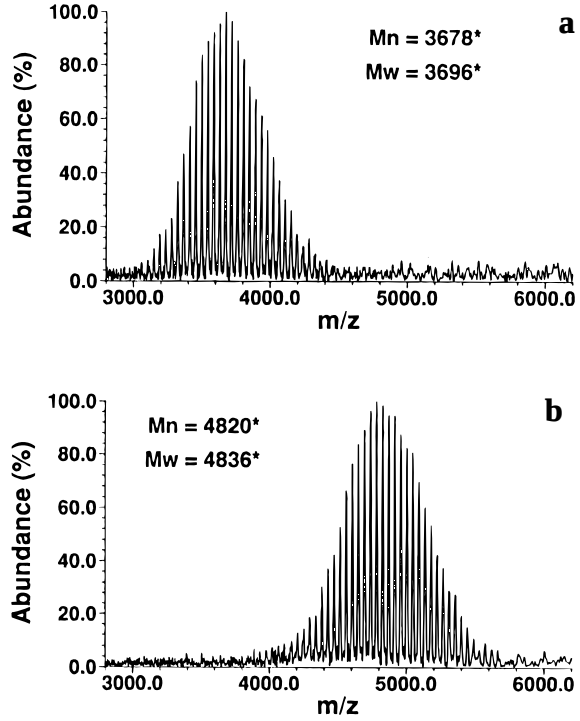
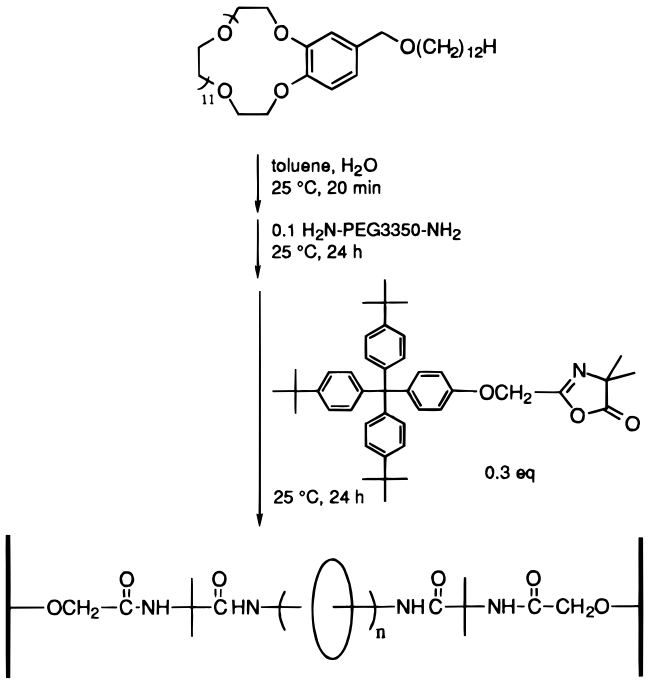


Figure 4. MALDI−FT mass spectra of PEG3350−bis(amine) (a) and of PEG3350−bis(amine) end-capped with 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone (b). M_n and M_w values were calculated from the mass corrected for the Na^+ attachment.

The data in Table 6 confirm that the azlactone ring of 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone is very reactive to amines, yet it is relatively stable to water. That is, only 28% of the ring is hydrolyzed in the presence of a large excess of water in $CDCl_3$ (entry 1). In contrast, a much smaller excess of hexylamine opens 100% of the ring in only 10 min (entry 2). Similarly, 100% of PEG3350−bis(amine) is end-capped with 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]methyl]-4,4-dimethylazlactone within 10 h at room temperature (entry 3). Parts a and b of Figure 4 present the MALDI−FT mass spectra of PEG3350−bis-(amine) before and after it was end-capped with 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone, respectively. The molecular weights (M_n = 3678 before end-capping, M_n = 4820 after end-capping) correspond fairly well to the expected molecular masses of 3522 and 4781 g/mol, respectively.

Reaction of PEG−bis(amine)s with 2-*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxymethyl-4,4-dimethylazlactone is therefore an ideal end-capping reaction for threading experiments of MC-12 in water-induced micellar solutions, both because the azlactone ring reacts much faster with amines than with water and because the reaction does not generate a small-molecule byproduct that could disrupt how MC-12 is organized. However, since MC-12 and the PEG−bis(amine) thread are both composed of oligo(oxyethylene) segments and one or two hydrophobic components, a major challenge will be to isolate their polyrotaxane from the reaction mixture. In addition, positively identifying the polyrotaxane will be difficult. Although the formation of a higher molecular weight polyrotaxane can generally be identified by gel permeation chromatography (GPC), MC-12 apparently aggregates in polystyrene-based GPC columns using either THF or CH_2Cl_2 as solvent.

Scheme 8. Recommended Procedure for Synthesizing Homopolyrotaxanes of MC-12 and Poly(ethylene oxide) Using an Amphiphilic Approach



We have performed a preliminary threading experiment at room temperature according to the procedure outlined in Scheme 8. As expected, GPC results are uninformative, although they demonstrate that the end-capped thread is present in both major fractions of the polyrotaxane and/or aggregated MC-12. However, reverse-phase HPLC on a C18-coated analytical column of silica gel using 70% methanol, 30% water as the eluant indicates that there is a higher molecular weight component in the water-soluble fraction. Our MALDI−FTMS results do not confirm this, probably because the molecular weight is too high for detection. For example, if 100% of the rings are threaded, the average homopolyrotaxane would contain 12 rings per thread and have a molecular weight of 1.5×10^4. Another difficulty with analysis by MALDI−FTMS is the distribution of rings per thread and, therefore, the distribution of molecular weights. However, a broad mass distribution can be detected by sequentially adding the time domain data from a range of gated deceleration times as described previously.[14]

We have therefore established the conditions necessary to identify lower molecular weight homopolyrotax-

anes of MC-12 threaded with PEG−bis(amine)s and end-capped with 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]methyl]-4,4-dimethylazlactone. The next paper in this series describes the lyotropic behavior of MC-12 in toluene in the presence of water in order to optimize the threading experiments.[8] We are currently optimizing a purification procedure for the homopolyrotaxanes, which will be combined with the optimized threading conditions and the conditions we have established for identifying the polyrotaxane using MALDI−FTMS and the integral method.

Conclusions

The amphiphilic macrocrown ether (MC-12) was synthesized in high yield by formation of the macrocycle in the presence of multiple potassium template ions under pseudo-high-dilution conditions. This produced not only the distribution of ring sizes corresponding to monodisperse PEG600 but also approximately 17% of the double ring-size distribution. MC-12 aggregates in benzene and toluene in the presence of small amounts of water. Aggregation is accompanied by both an increase in the intensity of scattered light and by a significant broadening of the oxyethylene ^{1}H-NMR resonances at 3.5−4.3 ppm. The appearance of an HOD resonance at 4.5−5.0 ppm with only small quantities of D_2O demonstrates that a water pool forms in the interior of reverse micelles and/or inverse lyotropic mesophases of MC-12 in toluene. Micellar aggregation is thermally reversible, although it may require agitation by stirring or shaking when recooled.

The micellar aggregates also macroscopically phase separate when water-soluble PEG derivatives are added, but are stable in the presence of less-water-soluble PEG derivatives, such as those with hydrophobic end groups and/or higher molecular weight. A downfield shift in the oxyethylene ^{1}H-NMR resonances of the thread, and decreased rates of reaction with model end-capping agents indicate that many of the PEG threads are solubilized in the interior of the micelles. The optimum end-capping reaction for PEG−bis(amine) threads in the water-induced micellar solutions of MC-12 is the addition reaction with 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]methyl]-4,4-dimethylazlactone, both because the azlactone ring reacts much faster with amines than with water and because the reaction does not generate a small-molecule byproduct that could disrupt how MC-12 is organized. In contrast, reaction of PEG−bis-(amine) threads with *p*-[[*p*-tris(*p*-*tert*-butylphenyl)-methyl]phenoxy]methyl]benzaldehyde increases the amount of water in the system, and the resulting imine bond is hydrolytically unstable. The primary disadvantage of the end-capping reaction of PEG−bis(amine) threads with [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]-acetyl chloride is that the latter is hydrolytically unstable in the water-induced micellar solution. It also generates HCl as a byproduct.

Our recommended amphiphilic procedure for synthesizing homopolyrotaxanes of MC-12 threaded with poly-(ethylene oxide) is therefore to preform an organized solution of MC-12, add the PEG3350−bis(amine) thread, allow the solution to equilibrate for several hours while stirring, and then end-cap the threads with 2-[[*p*-[tris-(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone. We are currently establishing a purification technique for these homopolyrotaxanes, although they can be identified in the presence of impurities if the molecular weight is not too high by MALDI−FT mass spectroscopy using a gated deceleration technique and sequentially adding the time domain data.

Experimental Section

Materials. Acetyl chloride (99%), benzaldehyde (>99%), 1-bromododecane (97%), 1,6-diaminohexamethylene (99%), diisobutylaluminum hydride, ethyl bromoacetate (98%), ethylenediamine (99%), 1,4-phenylenediamine (97%), poly(ethylene glycol) 3400 (PEG3400), sodium borohydride (98%), tetrabutylammonium hydrogen sulfate (TBAH, 97%) and thionyl chloride (99%) were used as received from Aldrich. Benzene-d_6 (99.6%D), deuterium oxide (99.9%D), and toluene-d_8 (99.6%D) were used as received from Cambridge Isotope Laboratories. 2-Aminoisobutyric acid (99%), *N*-bromosuccinimide (99%) and *p*-tolunitrile (98%) were used as received from Lancaster. 4-Acetamidophenol (99%), poly(ethylene glycol) 3350−bis(amine) ($-O_2CNH(CH_2)_2NH_2$ end groups made from PEG3350: 3506 g/mol), poly(ethylene glycol) 3350−bis(6-aminohexyl) (made from PEG3350: 3516 g/mol), and poly-(ethylene glycol) 3350−bis(glycidyl ether) (75%, made from PEG3350: 3462 g/mol) were used as received from Sigma. Poly(ethylene glycol) 600 (PEG600, Janssen, mol wt range 570−630), 2,6-lutidine (Lancaster, >98%), potassium iodide (Mallinckrodt) and sodium cyanoborohydride (Acros, >95%) were used as received. Distilled water was used for micelle formation. 2,2′-Azobisisobutyronitrile (AIBN, Johnson Mathey, 99%) was recrystallized from methanol below 40 °C. 3,4-Dihydroxybenzaldehyde (Janssen Chimica, 97%) was recrystallized from ethanol/toluene. Methanesulfonyl chloride (Lancaster, 98%) was distilled from P_2O_5 under N_2 before use. Triethylamine (Lancaster) was distilled from KOH under N_2. Tris(*p*-*tert*-butylphenyl)(4-hydroxyphenyl)methane and [*p*-[tris-(*p*-*tert*-butylphenyl)methyl]phenoxy]acetyl chloride were synthesized by literature procedures.[25] Dry CH_2Cl_2 was obtained by washing with 10% HNO_3 in H_2SO_4, storing over $CaCl_2$, and then distilling from CaH_2 under N_2. Toluene was dried by distillation from purple sodium benzophenone ketyl under N_2. Benzene was dried by distillation from CaH_2 under N_2. All other reagents and solvents were commercially available and used as received.

Techniques. All reactions were performed under a N_2 atmosphere using a Schlenk line unless noted otherwise. ^{1}H-NMR spectra (δ, ppm) were recorded on either a Bruker AC-200 (200 MHz) or a Bruker AM-300 (300 MHz) spectrometer. Unless noted otherwise, all spectra were recorded in $CDCl_3$ with TMS as an internal standard. Static light scattering experiments were performed on a Wyatt Technology miniDAWN instrument equipped with a He−Ne laser (633 nm). HPLC grade toluene (Fisher Scientific) and water (EM Science) were filtered through 0.02 and 0.45 μm PTFE filters, respectively. Gel permeation chromatography (GPC) was performed at 35 °C using THF as solvent (1.0 mL/min), a set of 50, 100, 500, 10^4 and linear ($50-10^4$) Å Styragel 5 μ columns, a Waters 486 tunable UV/vis detector set at 300 nm, and a Waters 410 differential refractometer.

All matrix-assisted laser desorption/ionization (MALDI) Fourier transform mass spectrometry (FTMS) analyses utilized 2,5-dihydroxybenzoic acid (DHB, Fluka) as the matrix. Samples were prepared by dissolving approximately 1 mg of polymer in methanol and adding an appropriate amount of DHB to provide an analyte-to-matrix molar ratio of 1:300 to 1:500. NaCl was added to form a saturated solution to enhance cationization by Na^+. Samples were aerosprayed onto a rotating stainless steel probe tip as previously described.[32,33] Experiments were performed using a Fourier transform mass spectrometer equipped for MALDI as previously described[34] with the exception that the 3 T superconducting magnet was upgraded to a 7 T superconducting magnet (Oxford Cryomagnetic Systems, Oxford, U.K.). Analyses were carried out with a background pressure between 10^{-7} to 10^{-8} torr. Gated trapping[35] was optimized for each sample using a deceleration voltage of 9.5 V for up to 290 μs. For PEG3350−bis(amine) and the azlactone-end-capped PEG3350−bis(amine) samples, ions were then trapped by applying potentials of 1.0 V to front and rear trapping plates on the source side of a 2-in. cubic

dual cell. Trapping potentials of 0.3 V were employed for the MC-12 sample. Typically, a delay up to 1 s was imposed prior to chirp excitation from 50 Hz to 100 kHz at 290 Hz/μs. For PEG3350−bis(amine) and azlactone-end-capped PEG3350−bis(amine), spectra were obtained after applying Hamming apodization to the 16 K data points acquired in direct mode. The transient for MC-12 was acquired using 64 K data points in direct mode; no apodization was applied to this data. Spectra shown were obtained by co-adding data from nine laser shots. Polymer distributions were calculated according to the method described by Pastor and Wilkins.[34] External calibration was performed using poly(ethylene glycol) 1000 prior to analysis.

Syntheses. PEG600−Bis(mesylate). The bis(mesylate) of poly(ethylene glycol) 600 (600 g/mol) was prepared in 81−93% yield as in the following example. Methanesulfonyl chloride (21 g, 0.18 mol) was added dropwise over 30 min to an ice-cooled solution of PEG-600 (36 g, 60 mmol) and triethylamine (30 g, 0.30 mol) in dry CH_2Cl_2 (300 mL). After being stirred at room temperature for 24 h, the reaction mixture was poured onto ice. The organic layer was separated, washed twice with ice-cold water (200 mL each), twice with 10% aqueous HCl (200 mL each), twice with saturated $NaHCO_3$ (200 mL each), and once with water (200 mL). After the organic mixture was dried over $MgSO_4$, the solution was filtered and CH_2Cl_2 was removed in vacuo to yield 39 g (85%) of PEG600−bis(mesylate) as a brown oil. ^{1}H-NMR: 3.03 (s, −C*H*$_3$, 6 H), 3.62 (m, −OC*H*$_2$−, 44.8 H), 3.73 (m, −C*H*$_2$CH$_2$-OMs, 4 H), 4.35 (m, −CH$_2$C*H*$_2$OMs, 4 H).

$NH_2(CH_2)_2NH-PEG600-NH(CH_2)_2NH_2$. PEG600−bis(mesylate) (2.0 g, 2.6 mmol) was reacted with ethylenediamine (3.2 g, 53 mmol) at 80 °C for 18 h in a capped Schlenk flask (closed system). Excess ethylene diamine was distilled off under a slightly reduced pressure, and the resulting crude oil was purified by column chromatography using silica gel as the stationary phase and $CHCl_3$/methanol (2:1) as the eluant to yield 0.79 g (44%) of $NH_2(CH_2)_2NH-PEG600-NH(CH_2)_2NH_2$ as a brown oil. ^{1}H-NMR: 1.85 (br s, −N*H*$_2$ and −N*H*−, 6 H), 2.65 (t, −OCH$_2$C*H*$_2$NH−, 4 H), 2.76 (m, −NHC*H*$_2$C*H*$_2$NH$_2$, 8 H), 3.55 (t, −OC*H*$_2$CH$_2$NH−, 4 H), 3.59 (m, −OC*H*$_2$−, 44.8 H).

$NH_2(CH_2)_6NH-PEG600-NH(CH_2)_6NH_2$. A solution of PEG600−bis(mesylate) (2.0 g, 2.6 mmol) and 1,6-diaminohexamethylene (6.2 g, 53 mmol) in anhydrous ethanol (15 mL) was stirred at 75 °C for 18 h. The solution was cooled to room temperature and diluted with ethanol (50 mL). The resulting white precipitate was filtered off, and the filtrate was condensed and then diluted with CH_2Cl_2 (15 mL). The resulting white precipitate was filtered off. This filtrate was condensed and dried in vacuo to yield 1.3 g (63%) of $NH_2(CH_2)_6NH-PEG600-NH(CH_2)_6NH_2$ as a slightly yellow oil. ^{1}H-NMR: 1.34 (m, −[C*H*$_2$]$_2$−, 8 H), 1.52 (m, −C*H*$_2$CH$_2$NH$_2$ and −C*H*$_2$-CH$_2$NH−, 8 H), 2.63 (t, −C*H*$_2$NH−, 4 H), 2.72 (s, −N*H*−, 2 H), 2.80 (m, −C*H*$_2$NH$_2$ and −OCH$_2$C*H*$_2$NH−, 8 H), 3.62 (m, −OC*H*$_2$−, 48.8 H), 3.79 (s, −N*H*$_2$, 4 H).

***p*-AcNHPhO−PEG600−OPhNHAc.** A solution of PEG600−bis(mesylate) (2.0 g, 2.6 mmol) in ethanol (3 mL) was added dropwise over 5 h to a refluxing solution of 4-acetamidophenol (0.88 g, 5.8 mmol), KOH (0.33 g, 5.8 mmol) and KI (0.88 g, 5.3 mmol) in ethanol (5 mL) and water (3 mL). The solution was heated at reflux for 22 h. The mixture was cooled to room temperature, and the product was extracted with CH_2Cl_2. The CH_2Cl_2 extracts were washed with water and passed through a short column of silica gel using CH_2Cl_2 as the eluant. The solvent was removed and the residue was dried in vacuo to yield 1.9 g (84%) of *p*-AcNHPhO−PEG600−OPhNHAc as a brown oil. ^{1}H-NMR: 2.10 (s, −C*H*$_3$, 6 H), 3.63 (m, −OC*H*$_2$−, 44.8 H), 3.81 (t, −C*H*$_2$OAr, 4 H), 4.06 (t, −C*H*$_2$CH$_2$OAr, 4 H), 6.82 (d, 4 aromatic H ortho to −OCH$_2$−), 7.40 (d, 4 aromatic H ortho to −NHAc), 7.87 (s, −N*H*−, 2 H).

***p*-NH_2PhO−PEG600−OPhNH$_2$.** A solution of *p*-AcNHPhO−PEG600−OPhNHAc (1.9 g, 2.2 mmol) and concentrated HCl (5 mL) in water (3 mL) and ethanol (10 mL) was stirred at reflux for 12 h. Ethanol was removed by rotary evaporation, and the aqueous solution was neutralized with 0.1 M aqueous K_2CO_3. This was extracted with CH_2Cl_2, and the CH_2Cl_2 extracts were washed with water. The product was purified by flash chromatography using silica gel as the stationary phase and first flushing the column with CH_2Cl_2. *p*-NH_2PhO−PEG600−OPhNH$_2$ (0.79 g, 48% yield) was obtained as a brown oil by flushing the column with methanol and drying in vacuo. ^{1}H-NMR: 3.30 (br m, −N*H*$_2$, 4 H), 3.65 (m, −OC*H*$_2$−, 44.8 H), 3.80 (t, −C*H*$_2$OAr, 4 H), 4.03 (t, −C*H*$_2$-CH$_2$OAr, 4 H), 6.63 (d, 4 aromatic H ortho to −NH$_2$), 6.74 (d, 4 aromatic H ortho to −OCH$_2$−).

***p*-NH_2PhNH−PEG600−NHPhNH$_2$.** A solution of PEG600−bis(mesylate) (2.0 g, 2.6 mmol) and 1,4-phenylenediamine (5.7 g, 53 mmol) in anhydrous ethanol (15 mL) was stirred at 80 °C for 24 h. After cooling to room temperature, the solution was diluted with CH_2Cl_2 (50 mL). The resulting precipitate was filtered off, and the filtrate was concentrated. The residue was purified by column chromatography using silica gel as the stationary phase and CH_2Cl_2/methanol (10:1) as the eluant. The solvent was removed in vacuo to yield 1.0 g (49%) of *p*-NH_2PhNH−PEG600−NHPhNH$_2$ as a brown oil. ^{1}H-NMR: 3.22 (t, −C*H*$_2$NH−, 4 H), 3.41 (m, −N*H*$_2$ and −N*H*−, 6 H), 3.64 (m, −OC*H*$_2$−, 48.8 H), 6.55 (d, 4 aromatic H ortho to −NH$_2$), 6.60 (d, 4 aromatic H ortho to −NH−).

3,4-(42-Crown-14)benzaldehyde. 3,4-(42-Crown-14)benzaldehyde was synthesized in 47−73% yield. In a typical example, a solution of 3,4-dihydroxybenzaldehyde (1.5 g, 11 mmol) and PEG600−bis(mesylate) (8.0 g, 11 mmol) in DMF (100 mL) was added dropwise over 1 h to a solution of K_2CO_3 (4.4 g, 32 mmol) in DMF (30 mL) at 70 °C. The temperature was raised to 125 °C and the reaction mixture was stirred for 72 h. It was then cooled to room temperature and filtered to remove the potassium salts. DMF was distilled off under reduced pressure (55 °C/10 mm Hg), and the residue was purified by three reprecipitations from THF (50 mL) into hexanes (350 mL). The resulting brown semisolid residue was purified by flash chromatography using silica gel as the stationary phase and first flushing the column with $CHCl_3$. 3,4-(42-Crown-14)benzaldehyde (3.5 g, 47% yield) was obtained as a light brown oil by flushing the column with methanol and drying in vacuo. ^{1}H-NMR: 3.65 (m, −OC*H*$_2$−, 44.8 H), 3.90 (m, −C*H*$_2$CH$_2$OAr, 4 H), 4.20 (m, −C*H*$_2$OAr, 4 H), 7.01 (d, 1 aromatic H meta to −CHO), 7.40 (s, 1 aromatic H ortho to −CHO, ortho and meta to −CH$_2$OAr), 7.42 (d, 1 aromatic H ortho to −CHO, meta and para to −CH$_2$OAr), 9.81 (s, −C*H*O).

3,4-(42-Crown-14)benzyl Alcohol. 3,4-(42-Crown-14)benzyl alcohol was prepared in 53−87% yield using the following procedure. A solution of sodium borohydride (0.59 g, 16 mmol) in absolute ethanol (50 mL) was added all at once to an ice-cooled solution of 3,4-(42-crown-14)benzaldehyde (10 g, 14 mmol) in absolute ethanol (200 mL). After 24 h at room temperature, the reaction mixture was poured into ice water (250 mL), which was then acidified with 10% aqueous HCl. This mixture was extracted five times with CH_2Cl_2 (125 mL each), and the CH_2Cl_2 extracts were dried over $MgSO_4$. The solution was then filtered and the solvent was removed by rotary evaporation. The final traces of ethanol were removed in vacuo at 75 °C to yield 8.5 g (85%) of 3,4-(42-crown-14)-benzyl alcohol as a brown oil. ^{1}H-NMR: 2.50 (br s, −O*H*), 3.65 (m, −OC*H*$_2$−, 44.8 H), 3.85 (m, −C*H*$_2$CH$_2$OAr, 4 H), 4.12 (m, −C*H*$_2$OAr, 4 H), 4.52 (s, ArC*H*$_2$OH), 6.83 (s, 2 aromatic H ortho to −OCH$_2$−), 6.94 (s, 1 aromatic H ortho to −CH$_2$OH).

3,4-(42-Crown-14)benzyl Dodecyl Ether (MC-12). A solution of 1-bromododecane (2.7 g, 11 mmol) in DMSO (20 mL) was added to a solution of 3,4-(42-crown-14)benzyl alcohol (5.0 g, 7.1 mmol), ground KOH (1.2 g, 22 mmol), and potassium iodide (90 mg, 0.54 mmol) in DMSO (90 mL). The reaction was run at room temperature for 24 h and at 60 °C for another 24 h. It was then cooled to room temperature and poured into ice-cold brine (350 mL), and the product was extracted four times with $CHCl_3$ (300 mL each). The $CHCl_3$ extracts were dried over Na_2SO_4 and filtered, filtered and the solvent was removed by rotary evaporation. The product was dried in vacuo to yield 5.4 g (88%) of MC-12 as a viscous brown oil. The color was removed by flash chromatography using silica gel as the stationary phase, flushing the column first with $CHCl_3$ and then collecting MC-12 with a methanol flush, followed by washing a methanol solution of MC-12 with hexanes. ^{1}H-NMR: 0.87 (t, −C*H*$_3$), 1.25 (m, −[C*H*$_2$]$_9$−), 1.59

(m, $-OCH_2CH_2CH_2-$), 3.42 (t, $-OCH_2[CH_2]_{11}H$), 3.64 (m, $-OCH_2CH_2O-$, 44.8 H), 3.83 (m, $-CH_2CH_2OAr$), 4.14 (m, $-CH_2OAr$), 4.39 (s, $-OCH_2Ar$), 6.86 (s, 2 aromatic H ortho to $-OCH_2-$), 6.90 (s, 1 aromatic H ortho to $-CH_2O-$).

4-(Bromomethyl)benzonitrile. A solution of *p*-tolunitrile (5.0 g, 43 mmol), *N*-bromosuccinimide (7.6 g, 43 mmol), and a few crystals of AIBN in CCl_4 (30 mL) was stirred at reflux for 20 h. The succinimide was filtered off and the solvent was removed by rotary evaporation. The residue was recrystallized from $CHCl_3$/hexanes to yield 4.0 g (48%) of 4-(bromomethyl)-benzonitrile as white crystals; mp 109−110 °C. ^{1}H-NMR: 4.48 (s, $-CH_2Br$), 7.50 (d, 2 aromatic H ortho to $-CH_2Br$), 7.64 (d, 2 aromatic H ortho to −CN).

Methyl 2-Aminoisobutyrate−Hydrogen Chloride. According to a literature procedure,[31] 2-aminoisobutyric acid (0.20 g, 1.9 mmol) was added to a solution of thionyl chloride (2.3 g, 19 mmol) in methanol (5 mL) at 0 °C. The temperature of this heterogeneous mixture was increased to 90 °C in a screw-capped vial, and the resulting homogeneous solution was stirred at 90 °C for 1 h. After cooling to room temperature, the solvent was removed by rotary evaporation. The crude mixture was diluted with $CHCl_3$ (5 mL), and the resulting white precipitate was filtered off. The filtrate was condensed, and the residue was dried under vacuum to yield 0.25 g (84%) of methyl 2-aminoisobutyrate−hydrogen chloride as white crystals; mp 168−169 °C. ^{1}H-NMR: 1.73 (s, $-CH_3$, 6 H), 3.82 (s, $-CO_2CH_3$), 8.93 (br s, $-NH_3^+$).

[*p*-[*p*-Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzonitrile. 4-(Bromomethyl)benzonitrile (0.47 g, 2.4 mmol) was added to a refluxing solution of tris(*p*-*tert*-butylphenyl)-(4-hydroxyphenyl)methane (1.0 g, 2.0 mmol) and NaOH (95 mg, 2.4 mmol) in 95% ethanol (10 mL). The solution was stirred at reflux for 10 min, at which point the entire mixture solidified. After cooling to room temperature, the white precipitate was collected, washed with water and then with methanol, and recrystallized from acetone to yield 1.1 g (87%) of *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzonitrile as white crystals; mp 260−262 °C. ^{1}H-NMR: 1.30 (s, $-CH_3$, 27 H), 5.09 (s, $-OCH_2-$), 6.81 (d, 2 aromatic H ortho to $-OCH_2-$), 7.07 (d, 6 aromatic H meta to *tert*-butyl), 7.11 (d, 2 aromatic H meta to $-OCH_2-$), 7.23 (d, 6 aromatic H ortho to *tert*-butyl), 7.55 (d, 2 aromatic H meta to −CN), 7.66 (d, 2 aromatic H ortho to −CN).

[*p*-[*p*-Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde. Diisobutyl aluminum hydride (0.22 g, 1.5 mmol) was added to a solution of *p*-[[*p*-tris(*p*-*tert*-butylphenyl)-methyl]phenoxy]methyl]benzonitrile (0.96 g, 1.6 mmol) in dry toluene (15 mL) at −70 °C. The reaction mixture was stirred at −70 °C for 30 min and at room temperature for 10 h and then poured into cold dilute aqueous sulfuric acid. The aqueous mixture was extracted with CH_2Cl_2. After the organic solvents were removed by rotary evaporation, the residue was recrystallized from $CHCl_3$/MeOH to yield 0.85 g (88%) of *p*-[[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde as white crystals; mp 244−246 °C. ^{1}H-NMR: 1.30 (s, $-CH_3$, 27 H), 5.12 (s, $-OCH_2-$), 6.83 (d, 2 aromatic H ortho to $-OCH_2-$), 7.07 (d, 6 aromatic H meta to *tert*-butyl), 7.11 (d, 2 aromatic H meta to $-OCH_2-$), 7.23 (d, 6 aromatic H ortho to *tert*-butyl), 7.61 (d, 2 aromatic H meta to −CHO), 7.90 (d, 2 aromatic H ortho to −CHO), 10.02 (s, −CHO).

[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid. [*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid was prepared in 61−81% yield using the following procedure. A mixture of tris(*p*-*tert*-butylphenyl)(4-hydroxyphenyl)methane (0.44 g, 0.88 mmol), tetrabutylammonium hydrogen sulfate (0.12 g, 0.35 mmol), ethyl bromoacetate (0.33 g, 2.0 mmol), and 10 M aqueous NaOH (1.8 mL, 18 mmol) in toluene (6 mL) was stirred at 100 °C for 12 h. After cooling to room temperature, the organic layer was separated and toluene was removed by rotary evaporation. The ethyl [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetate was hydrolyzed by heating the residue in a solution of KOH (0.50 g, 8.9 mmol) in ethanol (8 mL) and water (2 mL) at reflux for 5 h. After cooling to room temperature, the reaction mixture was acidified with concentrated HCl. The precipitate was collected, washed with water, and dried in vacuo to yield 0.30 g (61%) of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetic acid as a white solid; mp 298 °C. ^{1}H-NMR: 1.30 (s, $-CH_3$, 27 H), 4.65 (s, $-OCH_2-$), 6.80 (d, 2 aromatic H ortho to $-OCH_2-$), 7.05 (d, 6 aromatic H meta to *tert*-butyl), 7.13 (d, 2 aromatic H meta to $-OCH_2-$), 7.25 (d, 6 aromatic H ortho to *tert*-butyl).

***N*-((Methoxycarbonyl)isopropyl)-[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide.** *N*-(Methoxycarbonyl)isopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide was prepared in 81−84% yield using the following procedure. A mixture of [*p*-[tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]acetyl chloride (0.38 g, 0.65 mmol), methyl 2-aminoisobutyrate−hydrogen chloride (0.10 g, 0.65 mmol), and triethylamine (0.13 g, 1.3 mmol) in $CHCl_3$ (5 mL) was stirred at room temperature for 10 h. The reaction mixture was washed consecutively with 10% aqueous HCl, 10% aqueous $NaHCO_3$, and water. The solvent was removed using a rotary evaporator, and the residue was dried in vacuo to yield 0.36 g (84%) of *N*-(methoxycarbonyl)isopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide as a white solid which was used without further purification; mp 228−229 °C. ^{1}H-NMR: 1.30 (s, $-C(CH_3)_3$, 27 H), 1.61 (s, $-C(CH_3)_2NH-$), 3.75 (s, $-CO_2CH_3$), 4.28 (s, $-OCH_2-$), 6.81 (d, 2 aromatic H ortho to $-OCH_2-$), 7.07 (d, 6 aromatic H meta to *tert*-butyl), 7.14 (d, 2 aromatic H meta to $-OCH_2-$), 7.25 (d, 6 aromatic H ortho to *tert*-butyl).

***N*-(Carboxyisopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide.** *N*-(Carboxyisopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide was synthesized in 71−80% yield using the following procedure. A solution of *N*-(methyoxycarbonyl)isopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)-methyl]phenoxy]acetamide (0.36 g, 0.55 mmol) and NaOH (37 mg, 0.92 mmol) in a mixture of ethanol (10 mL), THF (2 mL), and water (2 mL) was stirred at reflux for 3 h. After cooling to room temperature, the solution was acidified with concentrated HCl. The resulting precipitate was collected, washed with water, dried and recrystallized from a mixture of $CHCl_3$/hexanes to yield 0.28 g (80%) of *N*-(carboxyisopropyl)[*p*-[tris-(*p*-*tert*-butylphenyl)methyl]phenoxy]acetamide as a white solid which was used without further purification; mp 302−303 °C. ^{1}H-NMR: 1.30 (s, $-C(CH_3)_3$, 27 H), 1.63 (s, $-C(CH_3)_2NH-$), 3.75 (s, $-NH-$), 4.47 (s, $-OCH_2-$), 6.79 (d, 2 aromatic H ortho to $-OCH_2-$), 7.06 (d, 6 aromatic H meta to *tert*-butyl), 7.14 (d, 2 aromatic H meta to $-OCH_2-$), 7.23 (d, 6 aromatic H ortho to *tert*-butyl).

2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl-4,4-dimethylazlactone. 2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone was synthesized in 86−89% yield as in the following example. Triethylamine (67 mg, 0.66 mmol) was added dropwise to a solution of *N*-(carboxyisopropyl)[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]-acetamide (0.28 g, 0.44 mmol) and ethyl chloroformate (48 mg, 0.44 mmol) in dry benzene (10 mL) at 65 °C. The heating bath was increased to 85 °C, and the benzene−EtOH azeotrope (3 mL, bp 68 °C) was collected over 1 h. The reaction mixture was then cooled to room temperature, and triethylammonium hydrochloride was filtered off. The filtrate was condensed under reduced pressure, and the residue was dried in vacuo to yield 0.24 g (89%) of 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]methyl]-4,4-dimethylazlactone as a white solid; mp 220 °C dec. ^{1}H-NMR: 1.30 (s, $-C(CH_3)_3$, 27 H), 1.46 (s, $-C(CH_3)_2N=$), 4.83 (s, $-OCH_2-$), 6.85 (d, 2 aromatic H ortho to $-OCH_2-$), 7.06 (d, 6 aromatic H meta to *tert*-butyl), 7.12 (d, 2 aromatic H meta to $-OCH_2-$), 7.23 (d, 6 aromatic H ortho to *tert*-butyl).

Hydrolytic Stability and Reduction of *p*-NH_2PhNH−PEG600−NHPhNH_2 End-Capped with *p*-[[*p*-Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]benzaldehyde. A solution of *p*-NH_2PhNH−PEG600−NHPhNH_2 (15 mg, 19 μmol) and *p*-[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-benzaldehyde (36 mg, 58 μmol) in toluene (0.50 mL) was stirred at room temperature for 24 h. ^{1}H-NMR demonstrated that the PEG−bis(amine) was quantitatively converted to a PEG−bis(imine) by both the complete shift of the $-CH_2NH-$ resonance from 3.22 to 3.33 ppm and by the complete absence of the aromatic protons of the bis(amine) at 6.55 and 6.60 ppm. The solvent was removed by rotary evaporation and excess *p*-[*p*-tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-

benzaldehyde was isolated by flash chromatography of the crude product using silica gel as the stationary phase and flushing the column with CH_2Cl_2. The PEG derivatives were then isolated by washing the column with methanol. ^{1}H-NMR demonstrated that 31% of the imine bonds had been hydrolyzed back to amine groups. Much of the amine-terminated PEG was removed by washing this solid with methanol: 15% amine end groups; 85% imine end groups.

A solution of this crude mixture (18 mg, 17 μmol of imine groups), 1 drop of 37% aqueous HCl, and $NaBH_3CN$ (0.88 mg, 14 μmol) in THF (0.5 mL) was stirred at room temperature for 24 h. The solution was neutralized with dilute aqueous Na_2CO_3 and then extracted with CH_2Cl_2 (3 mL). After the solvent was removed by rotary evaporation, ^{1}H-NMR demonstrated that 100% of the imine groups were reduced to secondary amine groups: 1.30 (s, $-CH_3$, 54 H), 3.22 (t, $-CH_2CH_2NH-$, 4 H), 3.64 (m, $-OCH_2-$, 48.8 H), 4.71 (s, $ArCH_2NH-$, 4 H), 5.03 (s, $-OCH_2Ar-$), 6.57 (dd, 8 aromatic H ortho to $-NH-$), 6.83 (d, 4 aromatic H ortho to $-OCH_2-$), 7.06 (d, 12 aromatic H meta to *tert*-butyl), 7.10 (d, 4 aromatic H meta to $-OCH_2-$), 7.24 (d, 12 aromatic H ortho to *tert*-butyl), 7.41 (d, 8 aromatic H ortho to $-CH_2O-$ and ortho to $-CH_2NH-$).

PEG3350−Bis(amine) End-Capped with 2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone. A solution of PEG3350−bis(amine) ($-O_2C-NH-(CH_2)_2NH_2$ end groups) (50 mg, 14 μmol) and 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone (46 mg, 73 μmol) in CH_2Cl_2 (5 mL) was stirred at room temperature for 25 h. The solvent was removed in vacuo, and Et_2O (20 mL) was added. After the mixture was stirred for a few minutes, unreacted 2-[[*p*-[tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone was filtered off, and the filtrate was washed with dilute aqueous HCl. The solvent was removed by rotary evaporation and the crude product was purified by flash chromatography using silica gel as the stationary phase and first flushing the column with CH_2Cl_2. The azlactone-end-capped PEG3350−bis(amine) thread (23 mg, 34% yield) was then isolated as a white solid by washing the column with methanol. ^{1}H-NMR: 1.30 (s, $-C(CH_3)_3$, 54 H), 1.57 (s, $-C(CH_3)_2NH-$, 12 H), 3.33 (m, $-NHCH_2CH_2NH-$, 8 H), 3.64 (m, $-OCH_2-$, 302.6 H), 4.00 (t, $-NHCO(CH_3)_2-$, 2 H), 4.34 (t, $-CH_2O_2CNH-$, 4 H), 4.45 (s, $-CH_2OAr-$, 4 H), 5.57 (br s, $-NHCO_2-$, 2 H), 6.81 (d, 4 aromatic H ortho to $-OCH_2-$), 7.06 (d, 12 aromatic H meta to *tert*-butyl), 7.13 (d, 4 aromatic H meta to $-OCH_2-$), 7.23 (d, 12 aromatic H ortho to *tert*-butyl).

Attempted Synthesis and Isolation of the Polyrotaxane of MC-12 Threaded with 2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone−End-Capped PEG3350−Bis(amine). A solution (25.6 wt % MC-12/7.7 wt % H_2O/66.7 wt % toluene, [H_2O/[EO] = 1.1) of MC-12 (0.10 g, 0.11 mmol) and water (30 μL, 1.7 mmol) in toluene (0.30 mL) was stirred at room temperature in a 4 mL vial for 20 min. The PEG3350−bis(amine) thread (34 mg, 9.7 μmol) was added all at once, and the mixture was stirred at room temperature for 24 h. 2-[[*p*-[Tris(*p*-*tert*-butylphenyl)methyl]-phenoxy]methyl]-4,4-dimethylazlactone (18 mg, 29 μmol) was then added, and the solution was stirred at room temperature for another 24 h. The solvents were removed in vacuo, and the residue was introduced to a silica gel column. The column was first eluted with CH_2Cl_2 to remove unreacted 2-[[*p*-[tris-(*p*-*tert*-butylphenyl)methyl]phenoxy]methyl]-4,4-dimethylazlactone; much of the unreacted end-capping agent was contaminated with a small amount of MC-12. The column was then eluted with methanol to remove all other components. The residue of the methanol fraction was dissolved in CH_2Cl_2 and hexane was added dropwise until MC-12 precipitated out of solution as a gummy material. The liquor was decanted off and extracted with water. The solvent was removed from the organic layer, and ^{1}H-NMR showed resonances for MC-12, end-capped thread, and some unreacted end-capping agent; GPC showed that a component elutes at 36.2 min corresponding to the end-capped thread, and a component elutes at 33.0 min corresponding to aggregated MC-12 (and/or polyrotaxane). The solvent was removed from the water-soluble fraction (21 mg), and ^{1}H-NMR showed resonances for MC-12 and end-capped thread; GPC showed that a component elutes at 35.5 min corresponding to the end-capped thread, and a component elutes at 33.2 min corresponding to polyrotaxane and/or MC-12.

Acknowledgment is made to the National Science Foundation for support of this research through DMR-9308485 and an NSF Young Investigator Award to C.P. (1994−1999), with matching funds from Bayer, Dow Chemical, DuPont (DuPont Young Professor Grant), GE Foundation (GE Junior Faculty Fellowship), Pharmacia Biotech, and Waters Corp. We also acknowledge Dr. Babu Narayanswamy for establishing the synthetic route to MC-12.

References and Notes

(1) Rotaxane reviews: (a) Schill, G. *Catenanes, Rotaxanes and Knots*; Academic Press: New York, 1971. (b) Amabilino, D. B.; Stoddart, J. F. *Chem Rev*. **1995**, *95*, 2725. (c) Amabilino, D. B.; Raymo, F. M.; Stoddart, J. F. In *Comprehensive Supramolecular Chemistry*; Lehn, J.-M., Ed.; Pergamon Press: Oxford, England, 1996; Vol. 9; Chapter 3. (d) Vogtle, F.; Dunnwald, T.; Schmidt, T. *Acc. Chem. Res*. **1996**, *29*, 451.

(2) Polyrotaxane reviews: (a) Gibson, H. W.; Marand, H. *Adv. Mater*. **1993**, *5*, 11. (b) Ogino, H. *New J. Chem*. **1993**, *17*, 683. (c) Gibson, H. W.; Bheda, M. C.; Engen, P. T. *Prog. Polym. Sci*. **1994**, *19*, 843. (d) Amabilino, D. B.; Parsons, I. W.; Stoddart, J. F. *Trends Polym. Sci*. **1994**, *2*, 146. (e) Raymo, F. M.; Stoddart, J. F. *Trends Polym. Sci*. **1996**, *4*, 208.

(3) (a) Ogata, N.; Sanui, K.; Wada, J. *J. Polym. Sci., Polym. Lett. Ed*. **1976**, *14*, 459. (b) Maciejewski, M. *J. Macromol. Sci.—Chem*. **1979**, *A13*, 77. (c) Maciejewski, M.; Gwizdowski, A.; Pezak, P.; Pietrazak, A. *J. Macromol. Sci.—Chem*. **1979**, *A13*, 87. (d) Maciejewski, M. *J. Macromol. Sci.—Chem*. **1979**, *A13*, 1175. (e) Ashton, P. R.; Philp, D.; Spencer, N.; Stoddart, J. F. *J. Chem. Soc., Chem. Commun*. **1991**, 1677. (f) Wenz, G.; Keller, B. *Angew. Chem., Int. Ed. Engl*. **1992**, *31*, 197. (g) Shen, Y. X.; Gibson, H. W. *Macromolecules* **1992**, *25*, 2058. (h) Shen, Y. X.; Engen, P. T.; Berg, M. A. G.; Merola, J. S.; Gibson, H.W. *Macromolecules* **1992**, *25*, 2786. (i) Shen, Y. X.; Xie, D.; Gibson, H. W. *J. Am. Chem. Soc*. **1994**, 116, 537. (j) Wenz, G. *Angew. Chem., Int. Ed. Engl*. **1994**, *33*, 803. (k) Sun, X.; Amabilino, D. B.; Ashton, P. R.; Parsons, I. W.; Stoddart, J. F.; Tolley, M. S. *Macromol. Symp*. **1994**, *77*, 191. (l) Loveday, D.; Wilkes, G. L.; Bheda, M. C.; Shen, Y. X.; Gibson, H. W. *J. Macromol. Sci., Chem*. **1995**, *A32*, 1. (m) Preece, J. A.; Stoddart, J. F. *Macromol. Symp*. **1995**, 98, 527. (n) Marand, E.; Hu, Q.; Gibson, H. W.; Veytsman, B. *Macromolecules* **1996**, *29*, 2555. (o) Huff, J.; Preece, J. A.; Stoddart, J. F. *Macromol. Symp*. **1996**, *102*, 1. (p) Asakawa, M.; Ashton, P. R.; Brown, G. R.; Hayes, W.; Menzer, S.; Stoddart, J. F.; White, A. J. P.; Williams, D. J. *Adv. Mater*. **1996**, *8*, 37. (q) Mason, P. E.; Parsons, I. W.; Tolley, M. S. *Angew. Chem., Int. Ed. Engl*. **1996**, *35*, 2238. (r) Owen, G. J.; Hodge, P. *Chem. Commun*. **1997**, 11. (s) Gibson, H. W.; Liu, S.; Gong, C.; Ji, Q.; Joseph, E. *Macromolecules* **1997**, *30*, 3711.

(4) (a) Lipatova, T. E.; Kosyanchuk, L. F.; Gomza, Y. P.; Shilov, V. V.; Lipatov, Y.S. *Dokl. Akad. Nauk. SSSR* (*Engl. Transl.*) **1982**, *263*, 140. (b) Lipatova, T. E.; Kosyanchuk, L. F.; Shilov, V. V.; Gomza, Y. P. *Polym. Sci. USSR* (*Engl. Transl.*) **1985**, *27*, 622. (c) Shaffer, T. D.; Tsay, L.-M. *J. Polym. Sci., Polym. Chem. Ed*. **1991**, *29*, 1213. (d) Kern, J.-M.; Sauvage, J.-P.; Bidan, G.; Billon, M.; Divisia-Blohorn, B. *Adv. Mater*. **1996**, *8*, 580. (e) Whang, D.; Kim, K. *J. Am. Chem. Soc*. **1996**, *118*, 451. (f) Yamaguchi, I.; Osakada, K.; Yamamoto, T. *J. Am. Chem. Soc*. **1996**, *118*, 1811. (g) Zhu, S. S.; Carroll, P. J.; Swager, T. M. *J. Am. Chem. Soc*. **1996**, *118*, 8713. (h) Whang, D.; Jeon, Y.-M.; Heo, J.; Kim, K. *J. Am. Chem. Soc*. **1996**, *118*, 11333. (i) Hoskins, B. F.; Robson, R.; Slizys, D. A. *J. Am. Chem. Soc*. **1997**, *119*, 2952.

(5) (a) Harada, A. *Coord. Chem. Rev*. **1996**, *148*, 115 and references therein. (b) Panova, I. G.; Gerasimov, V. I.; Grokhovskaya, T. E.; Topchieva, I. N. *Dokl. Akad. Nauk. SSSR* (*Engl. Transl.*) **1996**, *347*, 58. (c) Fujita, H.; Ooya, T.; Kurisawa, M.; Mori, H.; Terano, M.; Yui, N. *Macromol. Rapid Commun*. **1996**, *17*, 509.

(6) Agam, G.; Graiver, D.; Zilkha, A. *J. Am. Chem. Soc*. **1976**, *98*, 5206.

(7) Kon-no, K. In *Surface and Colloid Science*; Matijevic, E., Ed.; Plenum Press: New York, 1993; Vol. 15, Chapter 3; and references therein.
(8) Brandys, F. A.; Pugh, C. *Macromolecules* **1997**, *30*, 8153.
(9) Crossland, R. K.; Servis, K. L. *J. Org. Chem.* **1970**, *55*, 3195.
(10) (a) Jacobson, H.; Stockmayer, W. *J. Chem. Phys.* **1950**, *18*, 1600. (b) Jacobson, H.; Beckmann, C. O.; Stockmayer, W. *J. Chem. Phys.* **1950**, *18*, 1607.
(11) Chenevert, R.; D'Astous, L. *J. Heterocycl. Chem.* **1986**, *23*, 1785.
(12) (a) Benedict, D. R.; Bianchi, T. A.; Cate, L. A. *Synthesis* **1979**, 428. (b) Johnstone, R. A. W.; Rose, M. E. *Tetrahedron* **1979**, 2169.
(13) Kon-no, K.; Kitahara, A.; El Seoud, O. A. In *Nonionic Surfactants*; Schick, M. J., Ed.; Marcel Dekker, Inc.: New York, 1987; Chapter 4.
(14) Day, M.; Castoro, J. A.; Wilkins, C. L. *Anal. Chem.* **1995**, *67*, 1575 and references therein.
(15) Pederson, C. J. *J. Am. Chem. Soc.* **1967**, *89*, 7017.
(16) See for example: (a) Mattern, D. E.; Hercules, D. M. *Anal. Chem.* **1985**, *57*, 2041. (b) Bahr, U.; Deppe, A.; Karas, M.; Hillenkamp, F.; Giessmann, U. *Anal. Chem.* **1992**, *64*, 2866. (c) Blais, J. C.; Tessier, M.; Bolbach, G.; Remaud, B.; Rozes, L.; Guittard, J.; Brunot, A.; Maréchal, E.; Tabet, J. C. *Int. J. Mass Spectrom. Ion Processes* **1995**, *144*, 131. (d) Montaudo, G.; Montaudo, M. S.; Puglisi, C.; Samperi, F. *Rapid Commun. Mass Spectrom.* **1995**, *9*, 453. (e) Thomson, B.; Suddaby, K.; Rudin, A.; Lajoie, G. *Eur. Polym. J.* **1996**, *32*, 239.
(17) Fendler, J. H. *Acc. Chem. Res.* **1976**, *9*, 153.
(18) ^{1}H-NMR of MC-12 in toluene-d_8 (ppm): 0.93 (t, $-CH_3$), 1.27 (m, $-[CH_2]_9-$), 1.65 (m, $-OCH_2CH_2CH_2-$), 3.48 (m, $-OCH_2CH_2O-$ and $-OCH_2[CH_2]_{11}H$, 46.8 H), 3.68 (br s, $-CH_2CH_2OAr$, 4 H), 3.97 (br s, $-CH_2OAr$, 4 H), 4.46 (s, $-OCH_2Ar$), 7.00 (m, 3 aromatic H).
(19) (a) Chu, B.; Wu, G. *Macromol. Symp.* **1995**, *90*, 251. (b) Robeiro, A. In *Reverse Micelles*; Luisi, P. L., Straub, B. E., Eds.; Plenum Press: New York, 1984; pp 113−120.
(20) Winsor, P. A. *Solvent Properties of Amphiphilic Compounds*; Butterworths Scientific Publications: London, 1954; Chapter 6.
(21) Penfold, J.; Staples, E.; Tucker, I.; Cummins, P. *J. Colloid Interface Sci.* **1997**, *185*, 424; and references therein.
(22) (a) Harrison, I. T.; Harrison, S. *J. Am. Chem. Soc.* **1967**, *89*, 5723. (b) Harrison, I. T. *J. Chem. Soc., Chem. Commun.* **1972**, 231.
(23) (a) Harrison, I. T. *J. Chem. Soc., Perkin Trans. 1* **1974**, 301. (b) Harrison, I. T. *J. Chem. Soc., Chem. Commun.* **1977**, 384.
(24) Marshall, J. A.; Andersen, N. H.; Schlicher, J. W. *J. Org. Chem.* **1970**, *35*, 858.
(25) Gibson, H. W.; Lee, S.-H.; Engen, P. T.; Lecavalier, P.; Sze, J.; Shen, Y. X.; Bheda, M. *J. Org. Chem.* **1993**, *58*, 3749.
(26) See, for example: March, J. *Advanced Organic Chemistry*, 3rd ed.; Wiley-Interscience: New York, 1985; p 784.
(27) Borch, R. F.; Bernstein, M. D.; Durst, H. D. *J. Am. Chem. Soc.* **1971**, *93*, 2897.
(28) See, for example: Odian, G. *Principles of Polymerization*, 3rd ed.; Wiley-Interscience: New York, 1991.
(29) See, for example: (a) Rasmussen, J. K.; Heilmann, S. M.; Krepski, L. R. In *Encyclopedia of Polymer Science and Engineering*, 2nd ed.; Mark, H. F., Bikales, N., Overberger, C. G., Menges, G., Eds.; Wiley-Interscience: New York, 1988; Vol. 11, p 558 and references therein. (b) Taylor, L. D.; Kolesinski, H. S.; Mehta, A. C.; Locatell, L.; Larson, P.S. *Makromol. Chem., Rapid Commun.* **1982**, *3*, 779. (c) Heilmann, S. M.; Rasmussen, J. K.; Palensky, F. J.; Smith, H. K., II. *J. Polym. Sci., Polym. Chem. Ed.* **1984**, *22*, 1179.
(30) Drtina, G. J.; Heilmann, S. M.; Moren, D. M.; Rasmussen, J. K.; Krepski, L. R.; Smith, H. K., II; Pranis, R. A.; Turek, T. C. *Macromolecules* **1996**, *29*, 4486 and references therein.
(31) Campana Filho, S. P.; Goissis, G. *J. Chromatogr.* **1982**, 236, 197.
(32) Castoro, J. A.; Köster, C.; Wilkins, C. L. *Rapid Commun. Mass Spectrom.* **1992**, *6*, 239.
(33) Yao, J.; Dey, M.; Pastor, S. J.; Wilkins, C. L. *Anal. Chem.* **1995**, *67*, 3638.
(34) Pastor, S. J.; Wilkins, C. L. *J. Am. Soc. Mass Spectrom.* **1997**, *8*, 225.
(35) Castoro, J. A.; Wilkins, C. L. *Anal. Chem.* **1993**, *65*, 2621.

MA9712155