Supplementary Material for Chemical Communications

Supplementary Material

Part 1

Experimental

Materials

Chitooligosaccharide (M_n=1800, the degrees of deacetylation is 96%), purchased from Shenzhen Bright Way Novel Bio-Material Tech. Co. Ltd.(P. R. China), was dried at 60□ under vacuum before using. ε-caprolactone (from Acros Organics, 99%) was purified by vacuum distillation over CaH_2 and the fraction collected at 96-98□(5mmHg) was used in polymerization. Stannous octoate ($Sn(oct)_2$) (from Sigma, 95%) was used as received. Xylene, CH_3Cl and DMSO were purified by the usual distillation method. Hexamethyldisilazan(HMDS), chlorotrimethylsilane, and other regents were used as received without further purification.

Trimethylsilylation of COS

A typical procedure was as follows. COS (8.0g, 49.5mmol as momosaccharide residue containing 98.9mmol hydroxyl group and 47.5mmol amino group) was firstly dissolved in DMSO (concentrations of ca. 15%) and then Hexamethyldisilazan (40ml, 191.0mmol) and chlorotrimethylsilane was slowly dropped into this solution at 80□ under a dried nitrogen stream. The reaction was stirred for 12 hours. After cooling slowly the mixtures was precipitated in water and washed three times with water to remove the unreacted COS. This crude product was purified by repeated dissolution in acetone and precipitation in water and then dried for 72h at 50□ under vacuum. The trimethylsilyl substitution of COS (D_{TMS}) was determined to be 2.3 based on the basis of the ratio of the integral areas of the signals for the methyl protons of the TMS groups at around δ=0.1ppm to those for the methylene protons(H-2) of monosaccharide residue at around δ=2.7ppm.

IR(KBr, cm^{-1}): 3450, 2960-2900, 1680, 1580, 1250, 1100-1000, 841, 748. ^{1}H-NMR($CDCl_3$, ppm): 4.3(H-1), 4.0-3.1(H-3, 4, 5, 6), 2.7(H-2), 0.1(TMS).

Graft copolymerization and deprotection

TMSCOS (1.0g, 3.1mmol as momosaccharide residue containing 1.8mmol hydroxyl group and amino group) were dissolved in fresh, purified mixture of CH_3Cl xylene (concentration~20%), and a desired amount of ε-caprolactone monomer and a drop of $Sn(oct)_2$ were added under N_2. The mixture in a capped vial under N_2 was placed in a preheated oil bath at 120□ and stirred for 24 h. The resulting polymer was dissolved in 10 mL of $CHCl_3$ and precipitated twice with CH_3OH to give the purified TMSCOS-*g*-PCL. The obtained TMS-protected graft copolymer was stirred in an isopropyl alcohol/H_2O/HCl mixture at room temperature. The solid was isolated, washed, and dried for 72 h at 50□ *in vacuo*. The degree of ε-caprolactone grafted on every glucose unit of COS backbone (D_p) was calculated as the ratio of the integral areas of the methylene signal of PCL at δ=2.3 ppm and the methylene protons(H-2) signal of monosaccharide residue at around δ=2.7ppm.

IR (KBr, cm^{-1}): 3440, 2950-2870, 1730, 1650, 1540, 1100– 1000. ^{1}H-NMR (DMSO-D_6, ppm): 4.9, 4.1, 3.8-3.5, 3.0, 2.3, 1.7, 1.4. ^{13}C-NMR (DMSO-D_6, ppm): 176.8, 67.5, 37.4, 31.9, 29.0, 28.1.

Self-assembly procedure of COS-g-PCL

Aggregates of COS-g-PCL in water were prepared as follows. COS-g-PCL copolymers were initially dissolved in THF. Deionized water was dropwise added to the COS-g-PCL solutions under vigorous stirring until pre-determined water contents were reached. After that, a large amount of water was added to the solutions to quench the resulting morphologies. The solution was then dialyzed against water to remove the organic solvent.

Measurements

Fourier-transform infrared (FTIR) spectra were obtained on a BIO-RAD FT3000 spectrometer using KBr pellets. ^{1}H-NMR and ^{13}C-NMR analyses were carried out by means of a JOEL JNM-ECA300 spectrometer in $CDCl_3$, D_2O, CF_3COOH or DMSO (solvents without TMS). Molecular weight (M_n) and molecular weight distribution (M_w/M_n) were measured with a Viscotek TDA 302 GPC instrument equipped with tetrahydrofuran (THF) as the mobile

phase and polystyrene as calibration standards. TEM was performed on a JEOL JEM-1200 electron microscope at an acceleration voltage of 120 kV. A copper grid with a carbon film was used. The copper grid was immersed in a drop of the aqueous polymer solution for 10 min and then removed and dried. A drop of phosorous tungstenic acid in water (2wt%) was placed on the copper grid for 2 min. The copper grid was then dried at room temperature prior to measurement.

Part 2

Results

Synthesis and Characterization

PCL copolymers are usually obtained with the ring-opening copolymerization of ε-caprolactone (CL) using hydroxyl groups as initiating points[1]. Amounts of hydroxyl groups on COS backbone can act as the initiating points of the ring-opening copolymerization of CL. Unfortunately, the poor solubility of COS in common organic solvents precluded the reaction in a homogenous solution and then COS-g-PCL with narrow molecular weight distribution was difficult to be obtained by heterophase copolymerization. At the same time, only part hydroxyl groups on COS backbone are required to participate in the copolymerization in order to preserve the hydrophilic properties of the COS backbone. Thus, COS-g-PCL with controlled structure was difficultly obtained by directly heterophase ring-opening copolymerization of CL onto COS backbone.

Trimethylsilyl(TMS) group is widely used in the protection of hydroxyl functional groups as its easily deprotection in wild condition[2]. The modification of TMS groups not only make regioselective reaction of polysaccharides possible but also solve the problem of poor solubility existing in polysaccharides by significantly improving the solubility in organic solvents[3]. Recently, the protection method of TMS groups was successfully used in the preparation of polysaccharide-based graft copolymers with controlled structures. Ohya and Cecile Nouvel synthesized Pollulan-g-poly(lactic acid) and Dextran-g-poly(lactic acid) via ring-opening graft copolymerization of lactide onto the corresponding trimethylsilyl (TMS) protected polysaccharides and subsequent removal of the TMS groups[4].

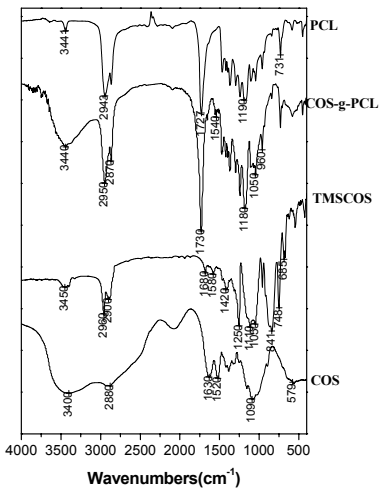

Figure 1. IR spectra of COS, TMSCOS, COS-g-PCL and PCL.

In this work, we synthesized amphiphilic graft copolymers, COS-g-PCL, using the protection and deprotection technique of part hydroxyl group via trimethylsilyl (TMS) groups and the homogenous ring-opening copolymerization of CL using $Sn(oct)_2$ as catalyst.

Trimethylsilylation of COS

The trimethylsilylation of COS took place first onto the hydroxyl groups and amino groups, in which the

protected-OH group number was controlled by the adjustment of the molar ratio of COS to HMDS.

Compared with that of parent COS, FTIR spectrum of TMSCOS appeared new peaks at 1250cm^{-1} and around 841–748cm^{-1}, corresponding to Si-Me groups (Fig. 1). A weaker -OH stretching peak around 3450 cm^{-1} can still be observed in the FTIR spectrum of TMSCOS, indicating that the hydroxyl groups of COS were not completely substituted. Therefore, these remaining free hydroxyl groups became the initiating points of the subsequent grafted polymerization. The introduction of the TMS group was further confirmed by the appearance of the methyl proton signal at 0.10 ppm next to the broad methine and methylene proton signals of the parent COS at 3.0–5.5 ppm in the ^{1}H NMR spectra (Fig. 2). The trimethylsilyl substitution of COS (D_{TMS}) was determined from the ratio of the integral areas of the methyl protons signals of TMS groups around 0.1 ppm to those of the methine protons (H-2) of monosaccharide residue at around δ=2.7ppm. In the experiment, D_{TMS} was approximately 2.3, indicating that the hydroxyl and amino groups of COS were only partly protected by the TMS groups.

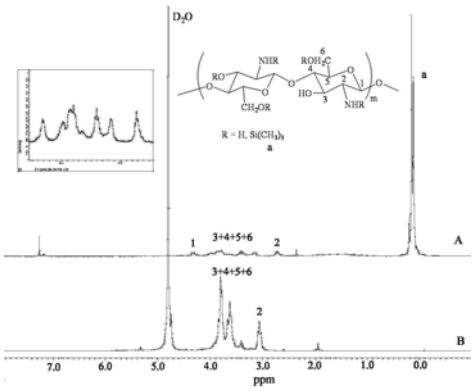

Figure 2. ^{1}H-NMR spectra of COS and TMSCOS. (A) COS in D_2O containing a drop of CF_3COOD and (B) TMSCOS in $CDCl_3$.

Graft Copolymerization and Deprotection

The obtained TMSCOS achieved solubility or swellability in a variety of organic solvents, such as pyridine, chloroform, THF, and xylene. Therefore, the homogeneous ring-opening copolymerization of ε-caprolactone grafted onto TMSCOS was successfully carried out in mixed solvent of chloroform and xylene with $Sn(oct)_2$ as catalyst. The TMS groups of TMSCOS-g-PCL were deprotected by incubation in an isopropyl alcohol/H_2O/HCl mixture.

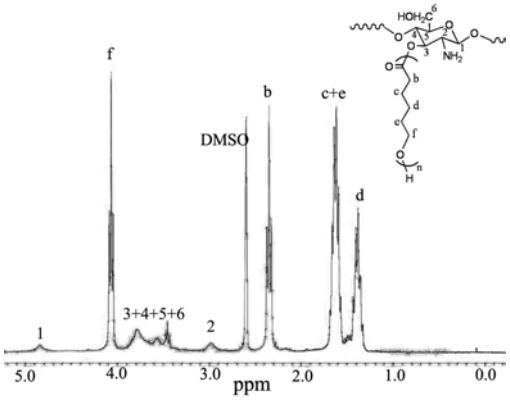

Fig. 3. ^{1}H NMR spectrum of TMS-deprotected graft copolymer COS-g-PCL (in DMSO-D_6, the average molar ratio of CL and glucose unit of COS is about 6).

As shown in Figure 1, the disappearance of the Si-Me groups peaks means that the TMS groups were successfully cleaved. A new peak at 1730 cm^{-1} can be attributed to the C=O stretching of the PCL segments in

COS-g-PCL. Figure 1 also shows a shift of the associated -OH groups in the whole synthetic process. The incorporation of PCL onto COS resulted in the shifting upward of the associated -OH groups frequency, meaning that the ability to form hydrogen bonds became weak after –OH on COS were partly substituted by PCL.

^{1}H NMR characterization of TMS-deprotected COS-g-PCL was in good agreement with the expected structure as shown in Figure 3. The deprotection of TMS groups was also confirmed by the disappearance of methyl proton signals from TMS at 0.10 ppm. The methylene proton signals of PCL can be observed at 4.1 ppm (three peaks), 2.3 (three peaks), 1.7 ppm (multipeaks), and 1.4 ppm (multipeaks). The methine and methylene proton signals of COS are at 3.0–5.0 ppm.

The average degree of ε-caprolactone grafted on every glucose unit of COS backbone (D_p) was calculated from the ratio of the integral areas of the methylene signal of PCL at 2.3 ppm to the methine proton signal(H-2) of COS at 3.0 ppm (Table 1).

GPC Curves of COS-g-PCL

Figure 4 shows the GPC curves of obtained COS-g-PCL with different PCL branch length. The contamination of the PCL homopolymer cannot be observed in the GPC curves.

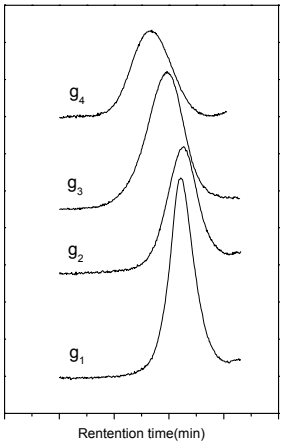

Figure . GPC curves of graft copolymers with different PCL branch length.

Supramolecular Self-assembly from COS-g-PCL in Water

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The self-assembly behavior of the synthesized copolymers is not the same, especially the polymer with large differences in COS/PCL ratios. In the case of the polymer (COS_{11}-g-PCL_{66}), due to the very low solubility of this polymer in THF, no self-assembly experiment was performed in our work. In the case of the polymer (COS_{11}-g-PCL_{132}), however, under the same self-assembly conditions like those for the polymer (COS_{11}-g-PCL_{253}), only sphere and rod-like micelles were observed. And, when the selective solvent is acid aqueous solution, the polymer COS_{11}-g-PCL_{132} can also form vesicle morphology. Figure 5 showed the TEM images of the self-assembled aggregates of COS_{11}-g-PCL_{132}.

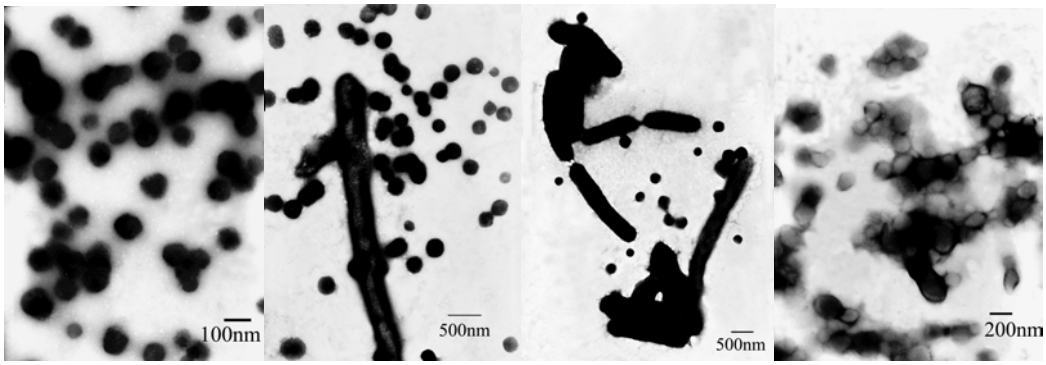

Figure 5. TEM pictures of 0.5% aggregates from 0.5% COS_{11}-g-PCL_{132} (THF) under different conditions. (A) 5% water; (B) 20% water (C) 30% water (D) 30% hydrochloric acid(0.3M)

Figure 6 shows the aggregate images from the polymer COS_{11}-g-PCL_{520} (0.5% polymer in THF, 5% water) before and after dialysis. Before dialysis the formed spherical micelles well disperse in the used solution due to the existence of THF (Figure 6A). After dialysis, however, a complicated network morphology, which clearly consists of the spherical micelles through fusion, was observed (Figure 6B). Additionally, it was observed that the micelles swelled with THF are larger than those after the removal of THF.

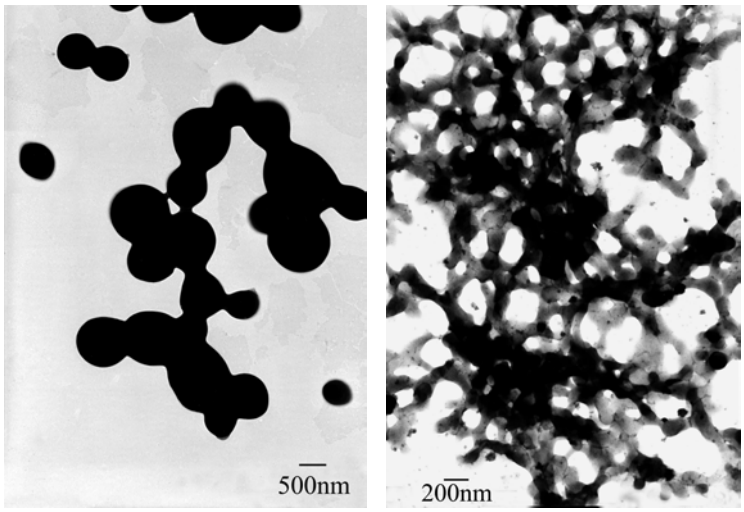

Figure 6. Transmission electron micrographs of the aggregates from COS_{11}-g-PCL_{520} (A) before dialysis; (B) after dialysis (0.5% copolymer in THF, 5% water).

References

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