Heparin-Like Macromolecules for the Modification of Anticoagulant Biomaterials

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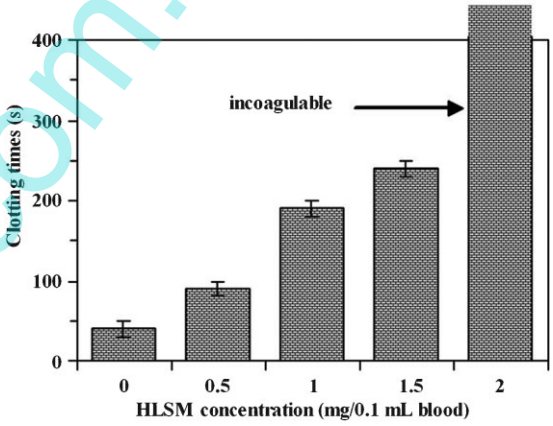
A heparin-like structured macromolecule (HLSM) is synthesized by RAFT polymerization using carboxyl-terminated trithiocarbonate as the RAFT agent. The HLSM can be directly blended with PES in DMAC to prepare flat-sheet membrane by means of a liquid–liquid phase separation technique. The synthesized polymeric material retard blood clotting and the modified membrane exhibits good anticoagulant ability due to the existence of the important functional groups $-SO_3H$, $-COOH$ and $-OH$. The anionic groups on the membrane surface may bind coagulation factors and thus improve anticoagulant ability. The results indicate that the HLSM has potential to improve the anticoagulant properties of biomaterials and to be applied in blood purification including hemodialysis and bioartificial liver supports.



Introduction

Anticoagulant biomaterials are extensively applied in biomedical fields as artificial organs, medical devices and disposable clinical instruments, such as hemodialysis system, extracorporeal circulation system, heart valve prosthesis, heart pacemaker, vascular prosthesis and intravascular stent.[1,2] Although many polymeric biomaterials have been developed, e.g., cellulose acetate (CA), poly(methyl methacrylate) (PMMA), polyethylene (PE), poly(propylene) (PP), poly(acrylonitrile) (PAN), ethylene vinyl alcohol copolymer (EVAL), poly(vinyl alcohol) (PVA), polysulfone (PSf) and poly(ethersulfone) (PES), not all of them are ideal in clinical situation,[3,4] especially as blood contacting materials. In practice, anticoagulant is usually needed for medical devices made from these materials.[5] In order to improve the blood compatibility of these materials, surface modification is an applicable choice.

The modification approaches include grafting negative charges or biological macromolecules (such as bovine serum albumin and heparin), grafting of amphion polymers, designed microphase separation, planting endothelial cell, and so on.[6–11] It was reported that surface heparinization was one of the most effective methods.[12] Many investigators have attempted to develop new synthetic anticoagulants or antithrombogenic materials with heparin-like structure to mimic the activity of heparin. Most of these materials are composed of ionic polymers containing sulfate, sulfamide, and carboxylic acid groups, since it is believed that the anticoagulant activity of heparin is caused by the presence of these ionic functional groups.[13–15] These materials include poly(styrene sulfonate), sulfonated

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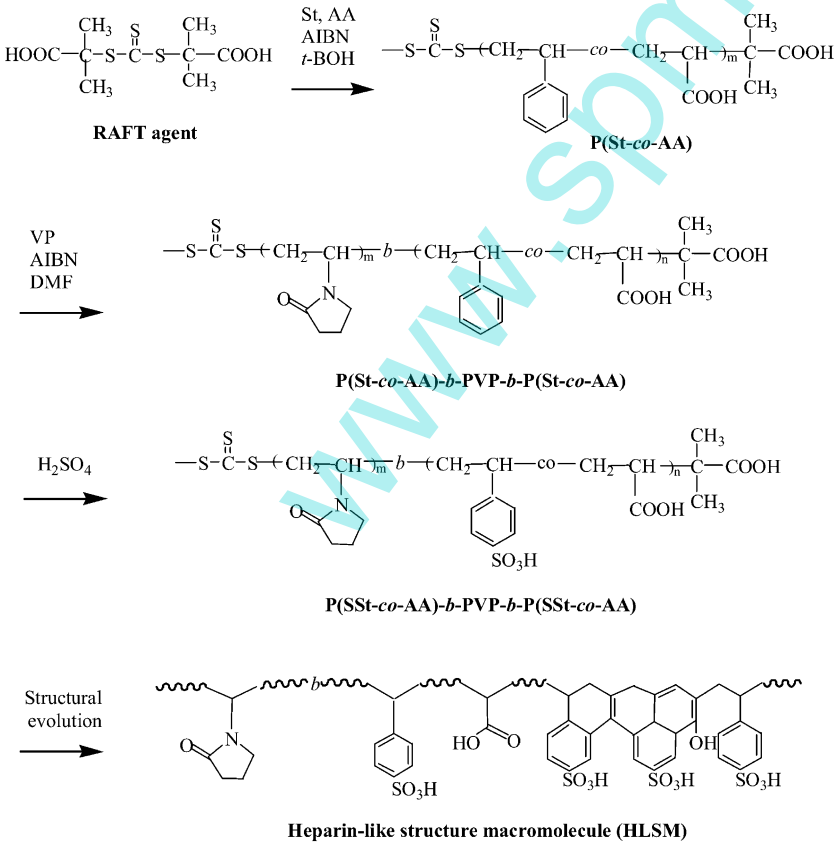
polyurethanes, sulfonated poly(vinyl alcohol/acrylic acid), sulfated poly(vinyl alcohol), sulfonated polyisoprene; sulfonated poly(ethylene oxide), and sulfated silk fibroin. Han et al.[16] investigated the heparin-like anticoagulant effect of sulfonated poly(ethylene oxide). Fougnot et al.[17] and Douzon et al.[18] reported that the modification of polystyrene and crosslinked dextrans by incorporation of sulfonate and carboxylate moieties resulted in heparin-like anticoagulant activity when these insoluble polymers were contacted with blood. Until now, no synthetic polymer with heparin-like structure containing $-SO_3H$, $-COOH$ and $-OH$ synchronously was developed and reported as a surface modifying additive to modify conventional biomaterials.

In this study, a heparin-like structure macromolecule (HLSM, as shown in Scheme 1) was synthesized for the first time by reversible addition fragmentation chain transfer (RAFT) polymerization and was used as an additive to improve the blood compatibility of conventional biomaterials. As one of the most important polymeric materials widely used as membrane contacting blood directly in practice (e.g., hemodialysis),[19,20] PES was selected here as a substrate material to be modified. The as-prepared HLSM was blended with PES directly to improve the biocompatibility, especially the anticoagulant property. It was expected that the surface of the modified PES membrane should be full of $-SO_3H$, $-COOH$, and $-OH$ groups, which could improve the blood compatibility of the membrane for blood purification.



Scheme 1. Synthesis of the heparin-like structure macromolecules.

Experimental Section

Materials

PES (Ultrason E6020P) was obtained from BASF, Germany. Styrene and acrylic acid (St and AA; 99.0%) were purchased from UNI-CHEM, China. *N*-Vinylpyrrolidone (VP; 99.0%) and tetrabutylammonium hydrogen sulfate were purchased from Alfa Aesar, USA. St, AA and VP were pretreated by activated carbons before use. *N,N*-Dimethylacetamide (DMAC; AR, 99.0%) and *tert*-butyl alcohol (*t*-BuOH; 99.0%) were purchased from Chengdu Kelong Inc. (Chengdu, China) and used as the solvent. 2,2′-Azoisobutyronitrile (AIBN) was purchased from Chengdu Kelong Inc. (Chengdu, China), which were used as the initiator. All the other chemicals (analytical grade) were obtained from Chengdu Kelong Inc., China, and were used without further purification.

Synthesis of RAFT Agent

Following to the literature,[21] carbon disulfide (27.4 g, 0.36 mol), chloroform (107.5 g, 0.9 mol), acetone (52.3 g, 0.9 mol), and tetrabutylammonium hydrogen sulfate (2.41 g, 0.0071 mol) were mixed with 120 mL of mineral spirits in a 1 L jacketed reactor cooled with tap water under nitrogen. Sodium hydroxide (50% solution) (201.6 g, 2.52 mol) was added dropwise over 90 min to keep the temperature below 25 °C. The reaction was carried out overnight. Then, 900 mL of water was added to dissolve the solid, followed by 120 mL of concentrated HCl to acidify the aqueous layer, and then stirred for 30 min under a nitrogen purge. After filtering and rinsing thoroughly with water, the solid was dried to constant weight and then 41.3 g of earth-colored product was collected.

Synthesis of Poly(St-*co*-AA)

Copolymerization of St and AA was carried out in a sealed tube. The general procedure is as follows. St, AA, RAFT agent, AIBN, and *t*-BuOH were added into a tube. After bubbling for 30 min with nitrogen, the reaction mixture was allowed to warm under a nitrogen atmosphere to 80 °C, and the polymerization was carried out for 5 h. After precipitating in ethyl ether, the product of Poly(St-*co*-AA) was dried under vacuum at 50 °C overnight.

Synthesis of Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA)

Polymerization of VP was carried out in a sealed tube. The general procedure is as

follows. Poly(St-*co*-AA) (macro-RAFT agent, stirred until fully dissolved in dimethylformamide (DMF)), VP, (AIBN) and DMF were added into a tube. After bubbling for 30 min with nitrogen, the reaction mixture was allowed to warm under a nitrogen atmosphere to 80 °C, and the polymerization was carried out for 8 h. After precipitating in ethyl ether, the product of Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) was dried under vacuum at 50 °C overnight. The obtained product was ground into fine powder and immersed in H_2O and tetrahydrofuran (THF) for one week respectively, and then repeated for three times alternately, and finally dried in vacuum at room temperature for 24 h.

Synthesis of HLSM

The fresh prepared styrene/acrylic acid copolymer was sulfonated using concentrated sulfuric as the sulfonating agent. The weight ratio of the copolymer to the acid was 1:3. The copolymer was stirred at room temperature in the acid for 10 h, then the homogeneous solution was poured into ice-cold water, and yellow colored sulfonated polymeric powders of HLSM were precipitated. The powders were washed with cold water several times to remove the acid and were dried.

Preparation of Evaporation Membranes

The evaporation membrane was prepared by evaporating the solvent in a vacuum oven.[22] PES and HLSM were dissolved in DMAC to obtain a mixed polymer solution, which was then cast onto a glass plate and spread to uniform thickness, and dried in a vacuum oven at 40 °C for 3 d. The concentrations of PES and the additive were 16 and 5 wt%, respectively. The membrane was carefully washed with distilled water and dried at room temperature.

Preparation of Modified PES Membranes

The mixed membranes of PES/HLSM were also prepared by a phase inversion technique. PES and the synthesized HLSM were dissolved in DMAC by vigorous stirring until a clear homogeneous solution was obtained. The concentration of PES was 16 wt%. In the experiments, different membranes were prepared by changing the weight percentage of the copolymer in the casting solutions. The contents of the HLSM in the casting solutions were 0, 1, 3, 5, and 7 wt% (the maximum amount), respectively. After vacuum degassing, the casting solutions were prepared into membranes by spin coating coupled with a liquid-liquid phase separation technique at room temperature. The membranes were rinsed with distilled water thoroughly to remove the residual solvent. All the prepared membranes with the PES/HLSM ratio of 16/0, 16/1, 16/3, 16/5 and 16/7 were in a uniform thickness of about 60–70 μm and were called FSM-0, FSM-1, FSM-3 FSM-5 and FSM-7, respectively.

Characterization

Fourier-transform infrared (FT-IR) spectra were measured with FT-IR Nicolet 560 (Nicol American) instrument. To prepare FT-IR samples, the copolymer was dissolved in DMAC and cast on a potassium bromide (KBr) disc with a thickness of about 0.8 mm. The ^{1}H NMR spectra were recorded on a Varian Unity Plus 300/54 NMR spectrometer using deuterated dimethyl sulfoxide (DMSO-*d*) as the solvent at room temperature. The morphologies of the membranes were observed with an XL 30ESME scanning microscope. The membranes were frozen in liquid nitrogen, and then broken and sputtered with a gold layer before scanning electron microscopy (SEM) analysis. The structures and the elements of the membrane surfaces were investigated by reflected FT-IR and X-ray photoelectron spectroscopy (XPS). Atomic force microscopy (AFM) studies were conducted using CSPM400 (Benyuan Nano-Instruments Ldt., China).

The number-average molecular weight of the copolymer was determined by gel permeation chromatography (GPC) on an HP1100 using two PLgel columns (10 μm, 104 Å; 10 μm, 500 Å) using monodisperse polystyrene as the standard. The mobile phase was tetrahydrofuran (THF). The sample concentration was 1.0 g · L^{-1}. The detector was RID, and the flow rate was 1.0 mL · min^{-1}.

Wettability Measurements

The hydrophilicity of the membrane surface was characterized on the basis of contact angle measurement by means of a contact angle goniometer (OCA20, Dataphysics, Germany) equipped with video capture. A piece of 2 × 2 cm^2 membrane was attached on a glass slide and mounted on the goniometer. For the static contact angle measurements, a total of 3 μL double distilled water was dropped on the air-side surface of the membrane at room temperature, and the contact angle was measured after 10 s. At least eight measurements were averaged to get a reliable value. The measurement error was ±3°.

Platelet Adhesion

Healthy human fresh blood (Dr. Ran F., healthy, man, Chinese, 32 years old) was collected into vacuum tubes containing sodium citrate as an anticoagulant (anticoagulant to blood ratio, 1:9). The blood was centrifuged at 1 500 rpm for 15 min to obtain platelet-rich plasma (PRP) or at 4 000 rpm for 15 min to obtain platelet-poor plasma (PPP).

The PES and modified PES membranes were immersed in phosphate-buffered saline (PBS) solution and equilibrated at 37 °C for 1 h. The PBS solution was removed and then 1 mL of fresh PRP was introduced. The membranes were incubated with PRP at 37 °C for 2 h. Then the PRP was decanted off and the membranes were rinsed 3 times with PBS solution. Finally, the membranes were treated with 2.5 wt% glutaraldehyde in PBS at 4 °C for 1 d. The samples were washed with PBS solution, subjected to a drying process by passing them through a series of graded alcohol-PBS solutions (25, 50, 75 and 100%) and isoamyl acetate/alcohol solutions (25, 50, 75 and 100%). The platelet adhesion was observed using an S-2500C microscope (Hitachi, Japan).

Clotting Time of the HLSM and the Modified Membranes

To evaluate the antithrombogenicity of the HLSM and the modified membranes, the activated partial thromboplastin time (APTT) was

measured by an automated blood coagulation analyzer CA-50 (Sysmex Corporation, Kobe, Japan), and the test method was described as follows: in the beginning of the APTT test, healthy human fresh blood (Dr. Fen R., healthy, man, Chinese, 32 years old) was collected in vacuum tubes containing sodium citrate as an anticoagulant (anticoagulant to blood ratio, 1:9, v/v), and PPP was obtained after centrifuging at 4 000 rpm for 15 min. Synchronously, the HLSM or the membrane ($0.5 \times 0.5\ cm^2$, three pieces) was immersed in 0.2 mL PBS buffer (pH = 7.4) for 1 h. The PBS was removed and then 0.1 mL of fresh PPP was introduced. After incubating at 37 °C for 30 min, 50 μL of the incubated PPP was added into a test cup, followed by the addition of 50 μL APTT agent (Dade Actin Activated Cephaloplastin Reagent, Siemens; incubated 10 min before use), and incubated at 37 °C for 3 min. Thereafter, 50 μL of 0.025 M $CaCl_2$ solution was added, and then the APTT was measured. At least three measurements were averaged to get a reliable value, and the results was analyzed by a statistical method.

Results and Discussion

Preparation and Anticoagulation Performance of HLSM

The random copolymer of Poly(St-*co*-AA) could be conveniently synthesized via RAFT polymerization at 80 °C, in which *S*,*S*′-bis(α,α′-dimethyl-α″-acetic acid)trithiocarbonate acted as a chain transfer agent (CTA), AIBN acted as the initiator, and *t*-BuOH was the solvent. The main advantage of RAFT polymerization is the desirable controllability of molecular weight and low polydispersity index (PDI).[24] As shown from the results (see Table 1), the average molecular weights determined by GPC were consistent with the theoretical values, and the PDI values were all distributed within the desirable range, which confirmed the advantages of the controllable molecular weight and narrow polydispersity for the RAFT polymerization.

The block copolymer Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) was further prepared by RAFT polymerization, for which the synthesized Poly(St-*co*-AA) acted as macro-RAFT agent, and AIBN acted as the free radical initiator. The macro-RAFT agent, initiator and monomer

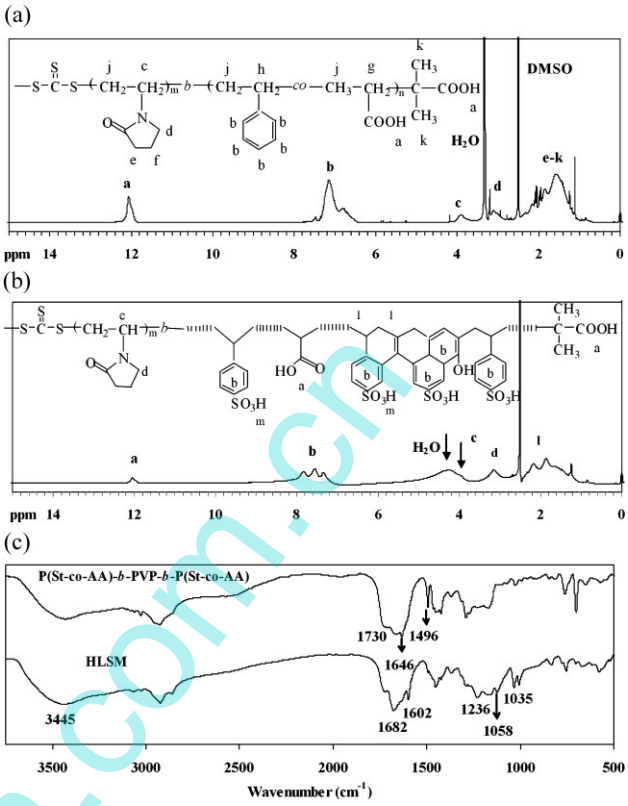


Figure 1. ^1H NMR spectra for (a) Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) and (b) HLSM in DMSO-*d* (b); (c) FT-IR spectra.

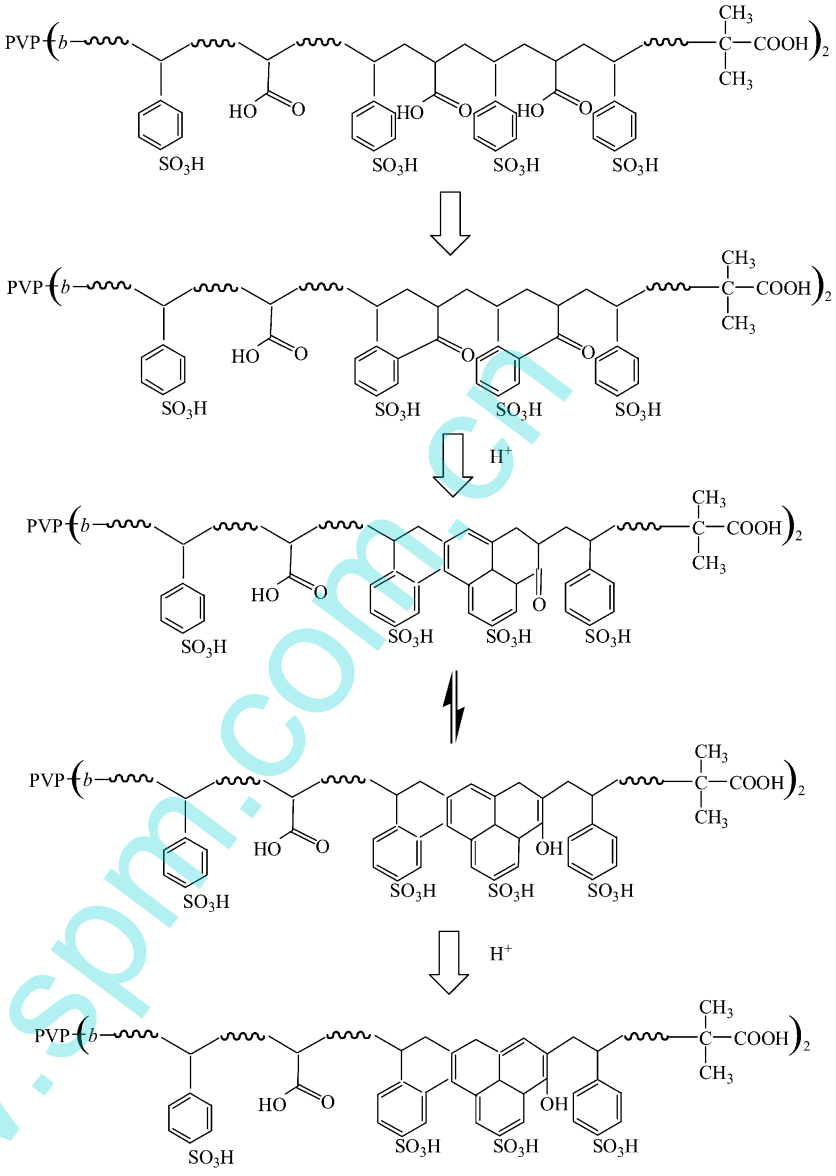
was fed in sequence. During the polymerization, Poly(VP) homopolymer and Poly(St-*co*-AA) copolymer will be formed due to the bi-radical termination. In order to remove the Poly(VP) homopolymer and Poly(St-*co*-AA) copolymer, the prepared product was ground into a fine powder and immersed in H_2O and THF for one week respectively; and this procedure was repeated three times. ^1H NMR spectroscopic analysis indicated that the block copolymer of Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) was successfully synthesized [see Figure 1(a)]. Sulfonation

Table 1. The synthesis condition and characteristic data of copolymer and HLSM.

Polymers	[Monomer]/[CTA]/[AIBN]	$\overline{M}_n$			PDI	Yield[d]
		Theory[c]	GPC	^1H NMR		[%]
PSA-1[a]	160/2.81/1	5 000	5 800	–	1.12	72.4
PSA-2[a]	160/1.40/1	10 000	11 700	–	1.16	74.2
PSA-3[a]	160/0.70/1	20 000	17 800	–	1.61	71.0
HSLM[b]	100/2.50/1	–	–	14 600	–	65.7

[a]Synthesis conditions: $T_p = 80$ °C, $t_p = 6$ h, *t*-BuOH as solvent; [b]Synthesis conditions: $T_p = 80$ °C, $t_p = 6$ h, PSA-2 as macro-RAFT agent, DMF as solvent; [c]Calculated according to ref. [23]; [d]Determined gravimetrically.

of the Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) copolymer with concentrated sulfuric acid is expected to give a structure having both sulfonic acid and carboxylic acid groups, [25] which is obtained through a two-step procedure including the introduction of the $-SO_3H$ on the block copolymer and the structural evolution of the copolymer after sulfonation. Figure 1 (b) shows that in HLSM remained the peak "a" for −COOH. The signals for the aromatic protons (Ar-H) of Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) in the ^{1}H NMR spectrum were well identified as peak "b". However, the peak was shifted to lower fields and split into 3 peaks after the sulfonation, which indicated the existence of the sulfonic acid group and −OH groups located at the aromatic ring.[26,27] These results indicated that there were different ratios of Ar−H before and after sulfonation due to the change of the chemical environment (the neighboring protons in Ar-H had been replaced). The HLSM also shows a very broad peak range from $\delta = 1$ to 3.5 in Figure 1(b), indicating that these protons are deshielded due to some conjugation. The FT-IR spectra of Poly(St-*co*-AA)-*block*-Poly(VP)-*block*-Poly(St-*co*-AA) and HLSM are shown in Figure 1(c). In the spectrum of the HLSM, the absorption peak of sulfonic group at 1 035 cm^{-1} ($-SO_4H$) and the peak at 1 600–1 730 cm^{-1} of carbonyl group indicated that the $-SO_4H$ group was successfully introduced into copolymer. The FT-IR and ^{1}H NMR spectra shown above can not confirm the presence of the hydroxyl group since there are lots of carboxylic groups in HLSM. In order to confirm the fact that there are hydroxyl groups generated during the structural recomposition process of sulfonation, another experiment was conducted: the HLSM powder shows a yellow color in acid form, and it changes to purple at the equivalence point. [28] On the basis of these studies, it could be deduced that the block copolymer during the sulfonation underwent an internal Friedel Crafts acylation followed by cyclic dehydration, leading to a conjugated structure, [28] which endows the block copolymer a heparin-like structure with three important functional group of $-SO_3H$, −COOH and −OH. A possible mechanism is proposed for the formation of HLSM (Scheme 2).



Scheme 2. A route for the formation of heparin-like structure.

APTT was measured to examine the blood coagulation in the presence of different concentrations of HLSM (Figure 2), and the results were analyzed by statistical methods (significant difference, $^*P < 0.05$, compared with the APTT of the pristine PES membrane). The blood clotting time with a small amount of HLSM was longer than that of the pure blood ($^*P < 0.05$), and the blood was incoagulable when the amount of the HLSM was over a concentration of 20 $mg \cdot mL^{-1}$. This suggests that adding the HLSM to blood may retard the blood clotting, since the HLSM has polar functional groups such as $-SO_3H$, −COOH and −OH, similar to heparin. Prolongation of clotting time indicates a reduced generation of coagulation factors.[29,30] Furthermore, the

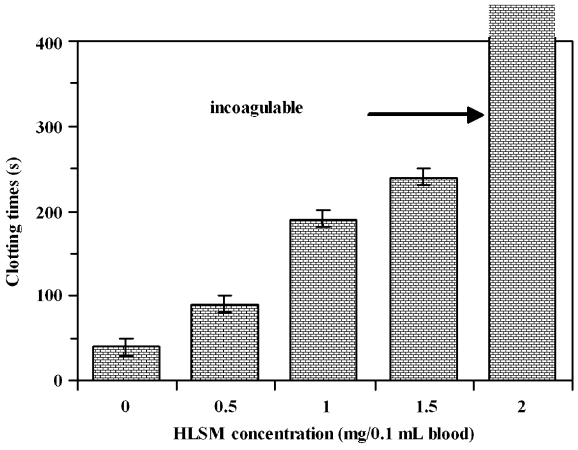


Figure 2. Activated partial thromboplastin times (APTTs) for the HLSM. $n = 3$, ($^{*}P < 0.05$ compared with the APTT of the pure plasma).

hydrophilicity of the materials could be the other important aspect, which might also have an impact on the adsorption of clotting factors and hence affect APTTs as well. [31]

Contact angle is a convenient way to assess the hydrophilic/hydrophobic properties of membrane surfaces. Figure 3 shows the contact angles of the HLSM evaporation membranes. As shown in the figure, the water contact angle was about 51°.

Fabrication and Blood Compatibility of HLSM-Modified Membranes

The HLSM modified PES membranes were also prepared by a liquid-liquid phase separation technique at room temperature after blending 16 wt% PES and 0–7 wt% HLSM in solvent DMAC. Figure 4 shows the cross-sectional SEM micrographs of the PES and PES/HLSM membranes. A characteristic morphology of asymmetric membrane consisting of a dense top-layer and porous sub-layer with a finger-like structure was observed for all the membranes.[32] However, the porous sub-layer of the PES/HLSM membrane was more inerratic and obvious than that of the PES membrane. For the modified membrane, there were large amounts of micro-pores on the surface of the macrovoids. Moreover, with the increase of the HLSM amounts, more pores appeared in the membranes. The microscopic observations of the PES and PES/HLSM membrane surfaces, as shown in Figure 5, revealed that after the blending of HLSM, the surface of the substrate was roughened with lots of tiny grooves and fine holes. The SEM and AFM pictures suggested that the structure of the modified membrane altered after adding the HLSM.

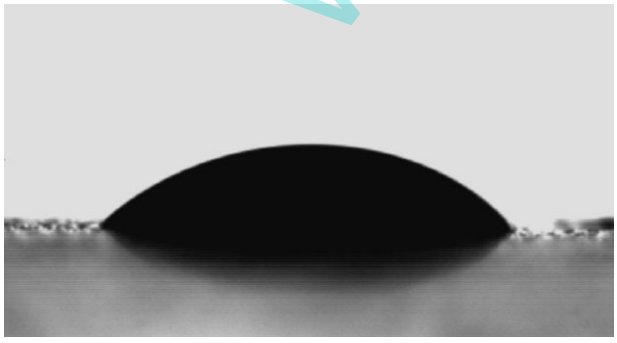


Figure 3. Water contact angle of HLSM.

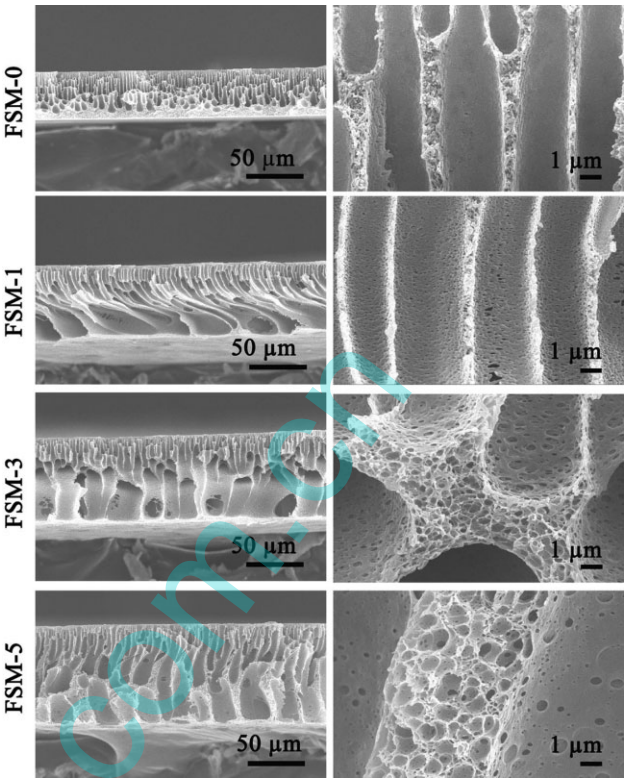


Figure 4. SEM images of the cross-section views of PES and modified PES membranes.

The formation mechanism might be summarized as follows: the initial solution, including PES, HLSM, and DMAC, was a homogeneous phase in which the macromolecules dissolved in the solution homogeneously. Because of the partial hydrophilic property of the copolymer, when phase separation occurred, it had a tendency to move toward the interface of water and dope solution, as shown in Figure 6. Thus, a layer of rich HLSM was formed at the surface of fresh-formed membrane, and pores were left in the bulk. Therefore, there may be abundant HLSM concentrated and a layer of HLSM formed on the surface of

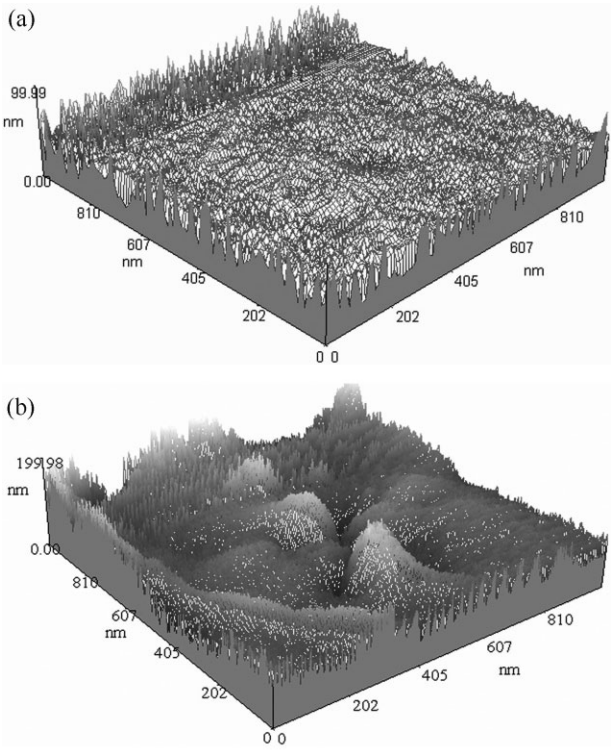


Figure 5. AFM images of PES (FSM-0, a) and modified PES membrane (FSM-5, b).

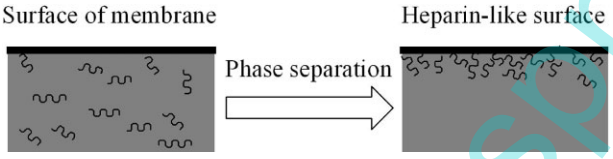


Figure 6. Schematic illustration of HLSM modified membrane formation.

the membrane, which was further confirmed by FT-IR and XPS analysis.

Figure 7 shows the FT-IR and XPS analysis of PES and modified PES membrane. It can be seen that the most significant changes were the appearance of the peaks at 3 450 and 1 676 cm^{-1}, which were the characteristic peaks of the HLSM. Peaks from the atomic composition of the HLSM were observed in the XPS spectra for the modified membrane surface. The element contents of N and S for the modified membrane surface were increased significantly (Table 2).

HLSM is assumed to move toward the surface of the membrane during the liquid/liquid phase separation and might be the dominant component on the surface. To confirm this, a comparison on the FT-IR spectra of the outer and inner parts of the membrane was also studied as shown in Figure 8. From the figure it was found that there are abundant hydrophilic HLSM in the outer surface (preferably getting in touch with water during the liquid/liquid phase

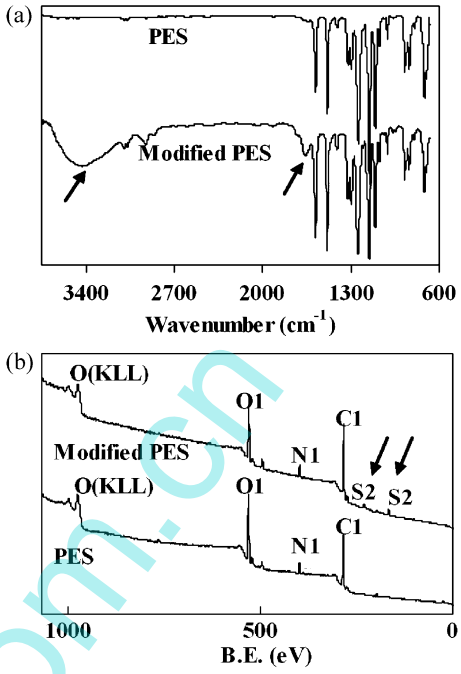


Figure 7. a) FT-IR; and b) XPS spectra for the PES and PES/HLSM membrane surfaces.

Table 2. XPS results for the compositions of the modified membrane surfaces.

Sample	Composition				
	[at%]				
	C	O	N	S	Others
PES	73.49	23.13	–	3.27	0.11
modified PES	67.74	19.98	3.83	8.29	0.06

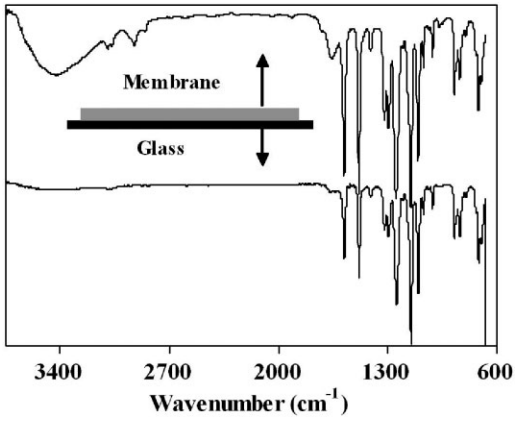


Figure 8. FT-IR spectra for the PES/HLSM membrane surfaces, $n = 3$.

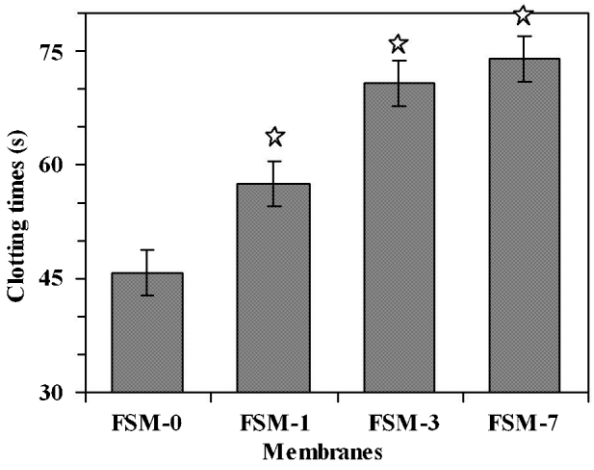


Figure 9. APTT values of the membranes, $n = 3$.

separation), more than in the inner part of the membrane (contacting with the glass).

In order to further confirm the stability of the modified membrane, we put the membrane in water for 1 week and found that there was a similar composition of the membrane surface segregation by FT-IR and XPS testing. However, the wettability measurements indicated that the wetting properties of the blended membrane had changed slightly.

The APTT test of the PES and the modified PES membranes are presented in Figure 9, and the results were analyzed by statistical methods (significant difference, $^{*}P < 0.05$, compared with the APTT of the pristine PES membrane). It was found that the APTTs of the modified membranes had increased compared with the PES membrane ($^{*}P < 0.05$); and with the increase of the HLSM amounts, the APTT values of the modified membranes also increased significantly. For the modified membranes, the anionic or polar groups $-SO_3H$, $-COOH$ and $-OH$, which were abundant on the surface of the membrane, were available for binding of coagulation factors. Similar to the heparin structure and its wonderful anticoagulant ability, the polydisperse anionic HLSM might bind and catalyze the interaction of plasma proteins involved in the intrinsic and extrinsic clotting cascade.

The adhesion of platelets and the morphology of adhering platelets are considered as a key event in thrombus formation on material surfaces. When a foreign material comes into contact with blood, the initial blood response is the adsorption of blood proteins, followed by platelet adhesion and the activation of coagulation pathways, leading to thrombus formation. [33,34] Figure 10 shows the typical scanning electron micrographs of the

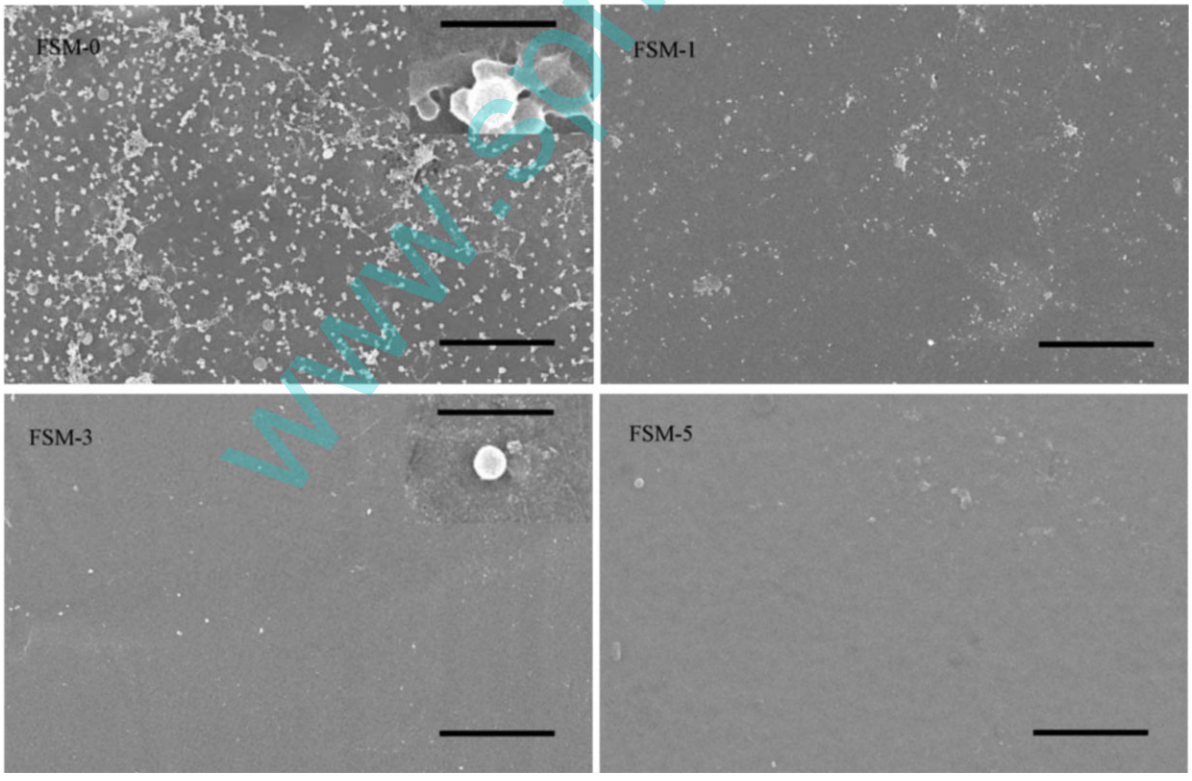


Figure 10. Scanning electron micrographs of the platelets adhering to the membranes. The scale bars correspond to 50 μm in all the images and 5 μm in the insets, $n = 3$.

platelets adhering to the PES and modified PES membranes. By comparing the figures, it was observed that numerous platelets were aggregated and accumulated on the PES membrane surface; and the platelets spread into flattened and irregular shapes. But for the modified membranes, very sparse platelets were observed; and the platelets expressed a rounded morphology with nearly no pseudopodia and deformation. The blended membranes exhibited a much suppressed number of the adhering platelets on the surface compared to the PES membranes, due to the anticoagulant activity of HLSM, as well as the improvement of wetting properties of the materials. The fact that there was an abundance of HLSM aggregated on the surface of the membrane may help to improve many properties of the membrane, especially the hydrophilicity of the membranes. It has been well known that the hydrophobic interaction between the material surface and protein plays a very important role in the non-selective adsorption of proteins. Materials possessing hydrophilic surface normally exhibit relatively low non-selective adsorption to protein or cells. [35]

Taking together the results of clotting assays and platelet adhesion, one can state that the blood compatibility of PES membranes can be considerably improved by blending HLSM.

Conclusion

In conclusion, an HLSM was synthesized for the first time by RAFT polymerization using carboxyl-terminated trithiocarbonate as the RAFT agent. The synthesized copolymer retarded the blood clotting and could be used as a surface modifying additive to improve the blood compatibility of conventional anticoagulant biomaterials. The modified membranes exhibited good anticoagulant ability due to the existence of the important functional groups $-SO_3H$, $-COOH$ and $-OH$. These anionic or polar groups on the membrane surface might bind coagulation factors and thus retard the blood clotting. The results indicate that the HLSM have the potential to improve the anticoagulant properties of biomaterials and might be applied in blood purification including hemodialysis and bioartificial liver supports.

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