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Incorporating α-Allyl Glucoside into Polyacrylonitrile by Water-Phase Precipitation Copolymerization To Reduce Protein Adsorption and Cell Adhesion

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ABSTRACT: α-Allyl glucoside (AG) was incorporated into polyacrylonitrile by water-phase precipitation copolymerization (WPPCP) for the first time with $K_2S_2O_8-Na_2SO_3$ as initiator system to improve the resistance properties of protein adsorption and cell adhesion for acrylonitrile-based polymer. The effects of initiator concentration, reaction time and temperature, and total monomer concentration on the copolymerization were studied, and some results were compared with those of solution copolymerization using AIBN as initiator. FT-IR, ^{1}H NMR, and ^{13}C NMR spectroscopes, element analysis, and DSC measurement were used to characterize the copolymers. It was found that both the yield and molecular weight for the WPPCP were higher than those for solution polymerization. The AG content in the resulting copolymers and the AG conversion for WPPCP were also higher than those of solution polymerization. The surface properties of the carbohydrate-containing copolymers were studied by pure water contact angle, protein adsorption, and cell adhesion measurements. It was found that the contact angle of the copolymer films decreased from 68° to 30° with the increase of AG content in the copolymer. The adsorption amount of bovine serum albumin (BSA) and the adhesive number of macrophage on the film surface also decreased significantly with increasing α-allyl glucoside content from 0 to 42 wt % in the copolymer. These results revealed that both the hydrophilicity and biocompatibility of polyacrylonitrile-based membranes could be improved by copolymerization acrylonitrile with vinyl carbohydrates.

Introduction

Polyacrylonitrile and acrylonitrile-based copolymers have been successfully applied as membrane materials in the fields of dialysis, ultrafiltration, enzyme immobilization, and pervaporation.[1−11] However, the relatively poor hydrophilicity and biocompatibility for this type of membrane limit their further applications in aqueous solution and biomedical usage. Ultrafiltration membranes are widely used in the biotechnology industry for the recovery of biological products in steps such as cell broth clarification, cell harvesting, concentration or diafiltration of protein solution prior to separation, and final concentration.[12] Nevertheless, one major problem with ultrafiltration is the loss of permeation flux caused by adsorptive fouling of biological molecules such as proteins on the surface and even inside the pores of typically used hydrophobic membranes such as polyacrylonitrile. Fouling reduces productivity due to longer filtration times and shortens membrane life due to the harsh chemicals necessary for cleaning. What is more, it can also alter membrane selectivity and lead to significant product loss through denaturation. Generally, two distinct types of fouling phenomena are considered:[13] (1) macrosolute (such as proteins) adsorption, which refers to specific intermolecular interactions between the macrosolute and the membrane that occur even in the absence of filtration, and (2) filtration-induced macrosolute or particle deposition, which is over and above that observed in a static system. Filtration-induced macrosolute or particle deposition is often reversible, nonadhesive fouling. A variety of methods have been reported to reduce this type of fouling for a range of different applications, as summarized recently by Ma et al.[14] Macrosolute adsorption is generally irreversible, adhesive fouling. In bioseparation and water-treatment applications involving proteins, cells, colloids, microbes, and undissolved hydrocarbons, the foulents are often adhesive and exhibit irreversible fouling due to hydrophobic interactions, hydrogen bonding, van der Waals attractions, extracellular macromolecular interactions, and other effects.

Increasing the hydrophilicity of the membrane surface can reduce fouling and improve biocompatibility for the membranes.[11−16] Therefore, there are many surface modification methods that have been reported to make ideal hydrophilic and fouling-resistant surfaces. Among them, the grafting of hydrophilic monomers on the membrane surface shows some promise.[10,12−16] However, grafting polymerizations induced by radical, plasma, electron-bean, γ-radiation, and ultraviolet may result in production of a significant amount of homopolymer or cross-linked polymer. The undesired homopolymer wastes expensive starting materials, and cross-linked polymer is detrimental to membrane filtration since the membrane pores may become blocked. Moreover, the grafting density (number of grafting sites per area) and grafting polymer chain length cannot be determined independently, much less controlled. Copolymerizing hydrophilic monomers such as acrylic acid, acrylamides, vinylpyrrolidone, 2-hydroxyethyl methacrylate, glycidyl methacrylate, and 4-vinylpyridine with acrylonitrile have also been used to improve the properties of acrylonitrile-based polymeric membranes.[3,5−8,11] However, much attention[3,5−6,8] was focused on the pervaporation properties and enzyme immobilization for these copolymer membranes. Only fragmentary data[11] were reported concerning the improvement of fouling reducing.

The primary objective for our work is the preparation of novel polyacrylonitrile-based membranes possessing the properties of fouling reducing, especially the resistance of protein adsorption and cell adhesion. Compared

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with the above-mentioned hydrophilic monomers used for surface modification and copolymerization, vinyl carbohydrates may meet these demands. The fundamental concept was inspired by the fact that carbohydrates exist in many forms and play important roles in natural living systems. Their highly hydrophilic characteristics together with their innate compatibility with biomolecules have led to considerable interest in their polymers synthesis.[17−27] Up to date, many artificial carbohydrate-containing polymers (so-called glycopolymers) have been prepared by means of the vinyl polymerization[17−23] or ring-opening polymerization[24,25] of the corresponding monomers, where the carbohydrate moieties are bound to the polymer chains by ester, amide, ether, or glycoside bond. Some applications or potential use of these polymers are hydrogels, surface modification, biomolecule, or cell recognition.[26−31] Although the carbohydrate-containing homopolymers have the properties of perfectly hydrophilicity and biocompatibility, they are also highly polar, water-soluble, fragile, and biodegradable.[32] Furthermore, because of the incompatibility of hydroxyl groups from the carbohydrate moieties with either initiators or normal organic solvents for polymerization, the synthesis procedures are relatively complex because all of these approaches require the use of protected monomers and the subsequent deprotection of polymer chains to generate the desired glycopolymers. Therefore, they cannot be used directly as backbone of membranes.

To increase the hydrophilicity and reducing the fouling of polyacrylonitrile-based membranes, it is one of the most effective methods to copolymerize vinyl carbohydrates with acrylonitrile.[33] In this paper, the copolymerization of acrylonitrile (AN) and α-allyl glucoside (AG) was carried out for the first time by water-phase precipitation copolymerization (WPPCP) method with $K_2S_2O_8-Na_2SO_3$ as initiator system and water as reaction medium. Because the carbohydrate monomer, α-allyl glucoside, is highly soluble in water, the synthesis procedure can be simplified by leaving out the protection and deprotection steps. Pure water contact angle, bovine serum albumin (BSA) absorption, and the macrophage adhesion onto the films fabricated from the AN/AG copolymers were studied to reveal the potentiality of the copolymerization acrylonitrile with vinyl carbohydrates in improving both the hydrophilicity and biocompatibility of polyacrylonitrile-based membranes.

Experimental Section

Materials. All chemicals were analytical grade. Acrylonitrile (AN) and dimethyl sulfoxide (DMSO) were commercial products and were purified by vacuum distillation before used. Potassium persulfate ($K_2S_2O_8$), anhydrous sodium sulfite (Na_2SO_3), and azobis(isobutyronitrile) (AIBN) were recrystallized by usual procedures. Allyl alcohol, anhydrous glucose, and tris(hydroxymethyl)aminomethane were used as received without further purification. Bovine serum albumin (BSA) was purchased from Sino-American Biotechnology Co. and used as received.

Monomer Synthesis. α-Allyl glucoside (AG) was synthesized with the method reported by Talley et al.[34] Dry hydrogen chloride (24.00 g, 0.66 mol) was dissolved in 800 g (13.79 mol) of dry allyl alcohol and then stirred with 400 g (2.22 mol) of anhydrous glucose at 70 °C for $4^1/_2$ h. After cooling, the mixture was treated with 92 mL of concentrated ammonium hydroxide and then with decolorizing carbon. The solution was concentrated at reduced pressure to a thick sirup, which was extracted by stirring vigorously with 2 L portions of dry acetone at room temperature until no more glucoside was removed. The acetone solution was concentrated in a vacuum, seeded, and placed in refrigerator to crystallize. The crude crystals were recrystallized three times with acetone. The melting point was measured as 101.5 °C. Anal. Calcd for $C_9H_{16}O_6$: C, 49.09; H, 7.27; O, 43.64%. Found: C, 48.80; H, 7.42; O, 43.78%. ^{1}H NMR of AG (500 MHz, DMSO-d_6, TMS): δ = 3.27−3.31 (t, HOC*H*⟨, 1H$_f$), 3.43−3.46 (m, HOC*H*⟨, 1H$_g$), 3.58−3.60 (bd, HOC*H*$_2$−, 2H$_i$), 3.62−3.66 (m, −C*H*⟨, 1H$_h$), 3.73−3.76 (m, HOC*H*⟨, 1H$_e$), 3.97−4.12 (dq, −C*H*$_2$O−, 2H$_c$), 4.85−4.86 (d, −OC*H*O−, 1H$_d$), 5.14−5.28 (dd, C*H*$_2$═CH−, 2H$_a$), and 5.82−5.90 (m, CH$_2$═C*H*−, 1H$_b$).

Copolymerization of AN and AG. Copolymerization of AN and AG was performed by using varying molar ratios of the two monomers. For 15/85 AG/AN feed, 100 mL of distilled and deionized water, 9.50 g of AG, and 11.50 g of AN were added into a four-necked round flask equipped with mechanical stirrer, thermometer, and nitrogen inlet tube. 45.6 mg of $K_2S_2O_8$ and 21.2 mg of Na_2SO_3 were added into the stirring solution while maintaining the reaction temperature at 60 °C under a nitrogen atmosphere. The copolymerization was continued for a designated period of time, and the precipitated copolymer was filtered and washed with excess distilled and deionized water and ethanol to remove residual monomers. The obtained copolymer was dried under vacuum at 60 °C to constant weight. The yield (conversion of monomers) was calculated by the mass ratio of the copolymer product to the total monomers in the feed. The oxygen contents of the copolymers were measured by elemental analysis (EA1110) and used to calculate the weight fractions of AG in the copolymers. The conversion of AG was calculated by the mass ratio of the monomer existing in copolymer to the monomer in the feed. The solution copolymerizations of AN and AG initiated by AIBN in DMSO were carried out at 70 °C with the usual procedure[33] for comparison.

Characterization. IR spectra were measured on a Bruck Vector 22 spectrometer. ^{1}H and ^{13}C NMR spectra were measured on a Bruck (Advance DM×500) nuclear magnetic resonance spectrometer. The solvent is dimethyl-d_6 sulfoxide, and three drops of D_2O were added to the solution. The composition of the copolymers was determined by element analysis (EA1110). Differential scanning calorimetry (DSC) analysis of the copolymers was conducted by a STA409PC thermal analysis system. The measurements were run under an Ar atmosphere at 10 °C/min heating rate to 500 °C. The static contact angle of the copolymer films was determined on a KRUSS DSA10-MK machine. Viscosity measurements were made in a thermostatic water bath at 30 ± 0.1 °C using a Ubbelchde viscometer. Copolymer was dissolved in DMSO that had been exhaustively dried over molecular sieves. For each copolymer, the viscosity of five concentrations was measured. Intrinsic viscosity was obtained by extrapolation of a plot of specific viscosity/concentration vs concentration to infinite dilution using linear least squares. Such analysis yield regression coefficients ≥0.999. Estimates of the copolymer molecular weight were obtained from the relationship for PAN in DMSO at 30 °C:[35]

$$[\eta] = 2.865 \times 10^{-2} M_v^{0.768}$$

where $[\eta]$ is the intrinsic viscosity and M_v is the viscosity-average molecular weight.

Preparation of AN/AG Copolymer Films. AN/AG copolymer films were prepared by casting the DMSO solution of the copolymers (7 wt %) onto glass plates followed by drying at 60 °C for 24 h and then at 150 °C under vacuum for a week to completely remove the residual solvent. The copolymer films thus obtained were removed from the glass plates by immersing in water and followed by drying for another 24 h at 100 °C under vacuum. All films for contact angle, protein adsorption, and cell adhesion measurements were treated according to the same procedure. The thickness of the dried film was approximately 100 ± 5 μm.

Adsorption of BSA. Bovine serum albumin (BSA) adsorption was carried out by the following method. BSA was

dissolved in a Tris-HCl buffer (pH = 8) that can facilitate the hydrophobic interaction and inversely depress the electrostatic binding between the protein and the polymer surface. To perform equilibrium experiments, copolymer film with 44 cm^2 external surface area was introduced into a tube containing 5 mL of Tris-HCl buffer at 30 °C, before being exposed to the protein solution. Any air bubbles that would adhere to the sample were removed by allowing the samples to cross the air/buffer interface several times. 10 mL of BSA solution was then introduced into the tube. After the protein solution remained in contact with the sample for 24 h at 30 °C, the film was taken out from the protein solution and was further rinsed gently until the surface remained constant. The amount of adsorbed protein was determined by measuring spectrophotometrically the difference between the concentration of albumin in the solution before and after contact with the polymer film. The spectroscopic analytical method utilized in this work for protein dosage was based on the reaction of albumin with Coomassie brilliant blue (Fluka) dyestuff to record the absorbance of the albumin−Coomassie brilliant blue complex according to Bradford's method.[36] The reported data were the mean value of triplicate samples for each copolymer.

Macrophage Adhesion. The murine macrophage suspension was prepared with the method reported by Li et al.[37] The suspension was isolated from freshly killed mice using chloroform. The skin was sprayed with alcohol and the abdomen opened. 10 mL of Roswell Park Memorial Institute (RPMI) 1640 containing 10% foetal bovine serum (FBS), 100 g/mL penicillin, and 100 μm/mL streptomycin was injected into the peritoneal cavity, and then abdomen was gently massaged by fingers for 5 min. The peritoneum was carefully punctured, and then the washings were removed by a sterile pipet and placed in a sterile container to be centrifuged at 1000 rpm for 10 min to collect the macrophages. The macrophages obtained were grown in RPMI 1640 to obtain the macrophage suspension in which the cell concentration was 1×10^6 cells/mL.

The copolymer films (10×10 mm^2) used were cleaned sequentially in an ultrasonic bath of ethyl alcohol solution for 10 min and then rinsed in phosphate buffered saline (PBS). Then the films were immersed in physiological saline (PH 7.4) to recondition for several hours. The cell suspension was inoculated on the surface of the films to assess the cell attachment. The incubation period was 48 h for the cell attachment test in a humidified atmosphere of 5% CO_2 in air at 37 °C. Then the supernatant was removed, and the films were washed cautiously five times using PBS (pH 7.2) prior to fixation. The adherent cell density on the films was quantified on the basis of measurements obtained visually from at least five randomly selected fields (0.75×1.00 mm^2) using an Olympus TE300 phase contrast optical microscope. The mean values of triplicate samples for each polymer with the standard deviation were reported.

Results and Discussion

Copolymerization Behaviors of AN/AG. The effects of initiator concentration, reaction time and temperature, and total monomer concentration on the copolymerization in water were studied using an 85/15 AN/AG monomer feed ratio. The results are presented in parts a, b, c, and d of Figure 1, respectively. It can be seen that the copolymerization of AN and AG behaved in a typical free radical chain polymerization fashion. At a constant 20 wt % total monomer concentration, the yield (overall monomer conversion) increased rapidly with the initiator concentration and reaction time as well as reaction temperature at first and then became almost stable (Figure 1a−c). Increasing the total monomer concentration raised the yield remarkably at 60 °C and a constant initiator/monomer ratio (1/500). On the other hand, the molecular weight of the resultant copolymer decreased with the initiator concentration and reaction temperature while it in-

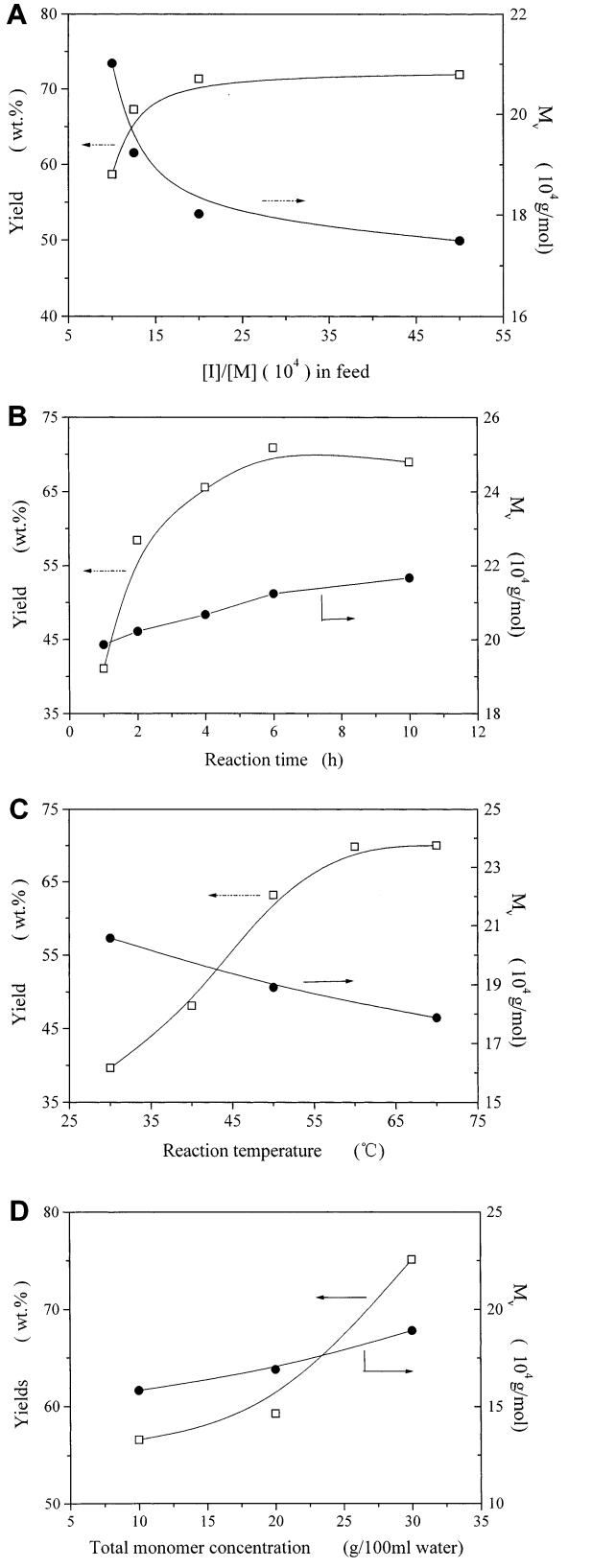

Figure 1. Typical results for the water-phase precipitation copolymerization of acrylonitrile/α-allyl glucoside. (a) Effects of initiator concentration ([AN]/[AG] mole ratio = 85/15, total monomer concentration = 0.20 g/mL, 60 °C, 6 h); (b) effects of reaction time ([I]/[M] = 1/500, [AN]/[AG] = 85/15, total monomer concentration = 0.20 g/mL, 60 °C); (c) effects of reaction temperature ([I]/[M] = 1/500, [AN]/[AG] = 85/15, total monomer concentration = 0.20 g/mL, 6 h); (d) effects of total monomer concentration ([I]/[M] = 1/500, [AN]/[AG] = 85/15, 60 °C, 6 h).

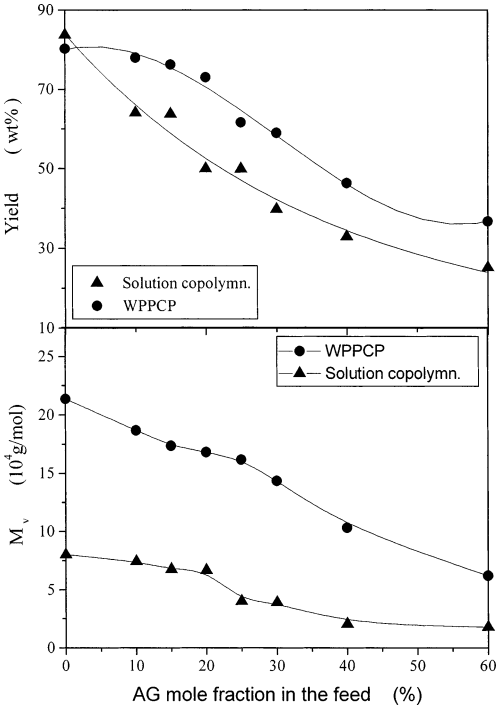

Figure 2. Effects of AG mole fraction in monomer feed ([I]/[M] = 1/500, 60 °C, 6 h) on the yield and molecular weight of the copolymer synthesized by water-phase precipitation and solution copolymerization.

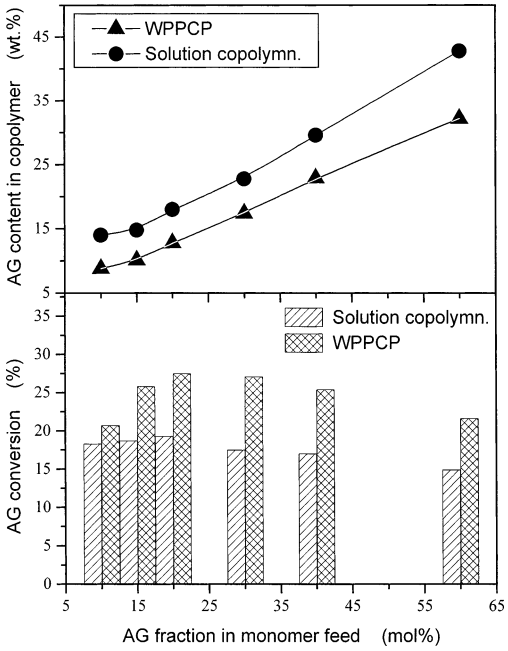

Figure 3. Effects of AG mole fraction in monomer feed on AG content in the copolymer and AG conversion for both water-phase precipitation and solution copolymerization.

creased slightly with reaction time and total monomer concentration. From these results, the optimal condition for AN/AG (85/15) copolymerization in water was obtained as follows: total monomer concentration, 20 g/100 mL water; initiator/monomer ratio, 1/500; reaction temperature, 60 °C; reaction time, 6 h.

The influences of monomer feed ratio on the copolymer yield and molecular weight are shown in Figure 2. The results of solution copolymerization conducted in DMSO using AIBN as initiator are also included for comparison. It was found that both the yield and molecular weight for the WPPCP were higher than those for solution copolymerization. The M_v value for the copolymers initiated by AIBN was all in the 10^4 g/mol range, while it increased to 10^5 when WPPCP method was used, as is shown in Figure 2. Water-phase precipitation polymerization is a unique process that affords the advantage of increasing the molecular weight of the AN-based polymers,[38] which, in turn, benefits the mechanical strength of corresponding membranes.[39] When the slightly water-soluble monomer AN was added to the medium, a small fraction of AN dissolved in the continuous aqueous phase where initiator was present. Then, the polymerization was first taken place in water. After the chains in water grew big enough, they would precipitate from the water medium. At this time the polymerizations were transferred from homogeneous polymerization to heterogeneous polymerization, which means that the polymerizations were taken place both in water and at the surface of the microparticles precipitated from the water. In this condition, the chances of the chain termination and chain transfer became difficult; therefore, the molecular weight and the yield of the polymer increased.

From Figure 2, it can be also seen that the yield of the copolymer decreased obviously with increasing the AG feed concentration. It might be due to that the C═C bond of AG is linked with an alkoxy bond that has a negative effect on the radical polymerization, which leads to a low reactivity of AG. The low reaction ability of AG also led to the decrease of the M_v of the copolymer with increasing the AG content in feed.

As shown in Figure 3, increasing the mole fraction of AG in monomer feed mainly increased the AG content in the copolymers for both WPPCP and solution polymerization. Taking advantages of the WPPCP method, both the AG content in the copolymers and the AG conversion for WPPCP were higher than those of solution copolymerization in DMSO. This might be reasonable since AG was highly soluble and AN was slightly soluble in water. As mentioned above, for the WPPCP method, the initiator presented in water, therefore, the highly soluble AG had more chance to be initiated and polymerized than in the solution copolymerization method; in turn, the AG conversion and content were higher than those of solution copolymerization.

Characterization of the Copolymers. The copolymers of AN with AG were characterized by IR and NMR spectra. The IR spectra of polyacrylonitrile and the copolymers are shown in Figure 4. Compared with the spectrum of polyacrylonitrile, several new absorption bands appeared near 3400 and 1040 cm^{-1} for the copolymers. These bands were due to the hydroxyl (−OH) and the alkoxy bond (−O−CH_2−) in the carbohydrate moieties, respectively.

Figure 5 and Figure 6 are the ^1H NMR and ^{13}C NMR spectra of the AG monomer and a typical AN−AG copolymer. From these figures it can be seen that the ^1H NMR and the ^{13}C NMR all gave the information on both AN and AG monomer unit in the polymer chain, which confirmed that the sample was an AN−AG copolymer. Compared with the spectrum of the α-allyl glucoside monomer (Figure 5a, ^1H NMR: $\delta = 3.27-3.31$ (t, HOC$H\langle$, $1H_f$), 3.43−3.46 (m, HOC$H\langle$, $1H_g$), 3.58−3.60 (bd, HOCH_2−, $2H_i$), 3.62−3.66 (m, −C$H\langle$, $1H_h$), 3.73−3.76 (m, HOC$H\langle$, $1H_e$), 3.97−4.12 (dq, −CH_2O−, $2H_c$),

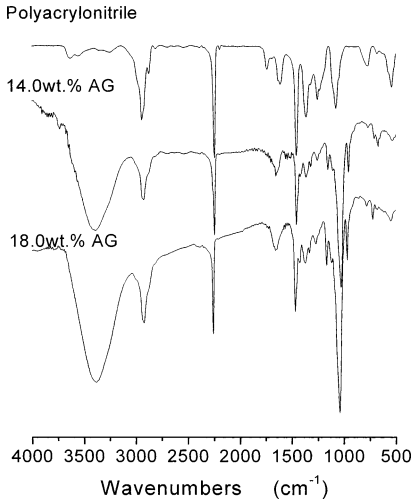

Figure 4. IR spectra of polyacrylonitrile and acrylonitrile/α-allyl glucoside copolymers.

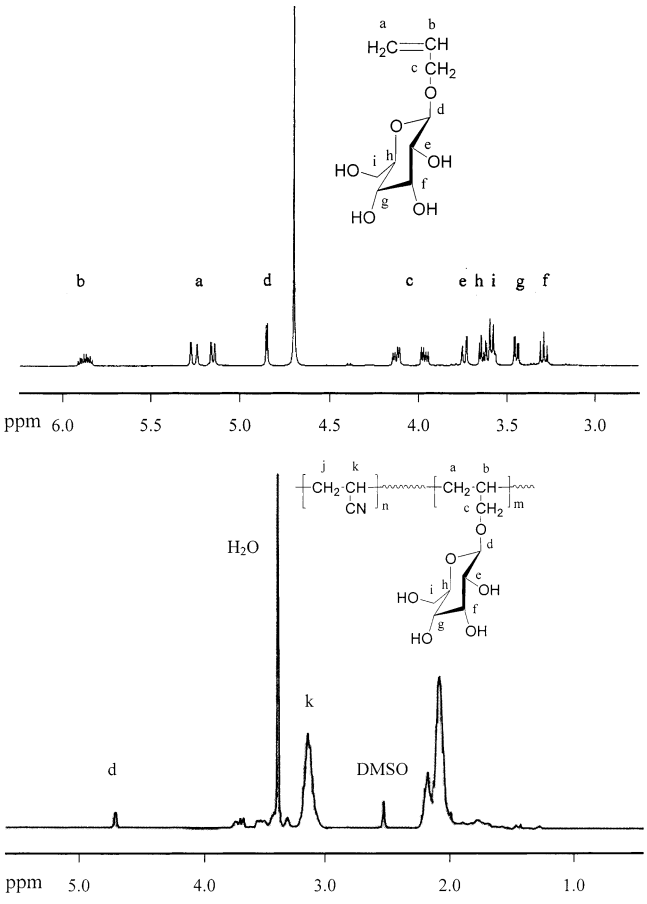

Figure 5. ^{1}H NMR spectra: (a, top) AG monomer; (b, bottom) acrylonitrile/α-allyl glucoside copolymer.

4.85−4.86 (d, −OCHO−, 1H$_d$), 5.14−5.28 (dd, CH$_2$═CH−, 2H$_a$), and 5.82−5.90 (m, CH$_2$═CH−, 1H$_b$)), it is obvious that the peaks of the vinyl group in monomer disappeared, and fine structures could not be observed for the corresponding copolymer. Other protons in the main chain gave broad peaks from 1.97 to 2.17 ppm. However, from this spectrum it was seen that the peaks at 3.12−3.15 and 4.70 ppm could provide the quantitative information about the AG content in the copolymers because the proton of ⟩CH−CN showed broad peaks in the range 3.12−3.15 ppm while the d proton in the AG unit gave a single peak around 4.70 ppm. The composition of the copolymers can be calculated according to the following equation:

$$\text{AG (wt \%)} = 220I_{4.70}/(220I_{4.70} + 53I_{3.12-3.15}) \times 100$$

where I is the intensity of the peaks in the NMR spectrum. The AG weight content value for the 85/15 monomers feed copolymer calculated by the ^{1}H NMR spectrum was 14.8 wt %, which was relatively consistent with the value 17.4 wt % calculated by element analysis method. The ^{13}C NMR spectrum of the copolymer (Figure 6) gave more information about the carbohydrate moieties than the ^{1}H NMR spectrum. The designations (δ = 60.72 (−C$_i$H$_2$O−), 68.00−69.50 (HOC$_g$H$_2$−), 70.04 (HOC$_c$H⟨), 71.69 (HOC$_e$H⟨), 72.87 (HOC$_h$H⟨), 73.15 (HOC$_f$H⟨), and 98.92 (−OC$_d$HO−)) revealed the existence of carbohydrate-carrying monomer in the copolymer, while the −CH$_2$−CH− (a and b carbons) of this monomer on the polymer chain seems overlap with those of acrylonitrile at 26.72−33.52 ppm.

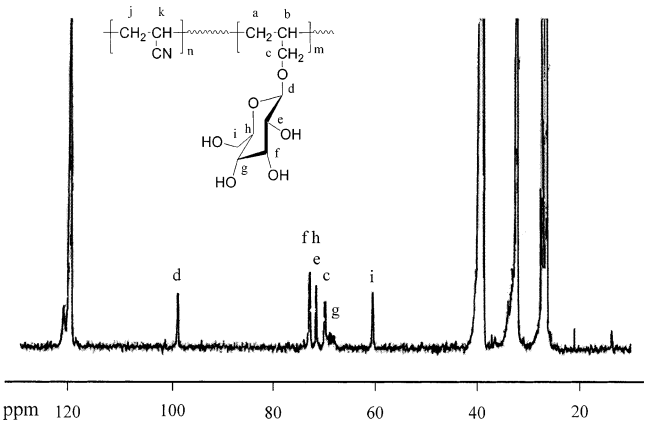

Figure 6. Typical ^{13}C NMR spectrum of acrylonitrile/α-allyl glucoside copolymer.

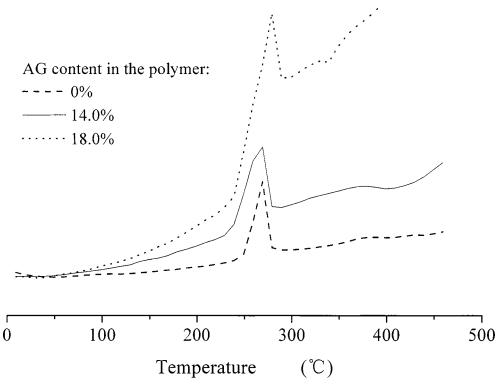

Figure 7. DSC curves of acrylonitrile/α-allyl glucoside copolymers.

DSC thermal diagrams of the AN homopolymer as well as the AN/AG copolymers are shown in Figure 7. Polyacrylonitrile showed a sharp exothermic peak at 269.7 °C, which was due to the cyclization reaction of nitrile groups. No obvious changes were observed for the copolymers. This could be ascribed to the relatively low AG content and the monodistribution of AG in the polymer chains.

Surface Properties of the Copolymer Films. Figure 8 shows the contact angle curve of pure water on the films fabricated from the acrylonitrile/α-allyl glucoside copolymers. It was found that the contact angle decreased gradually from 68.5° to 29.7° with

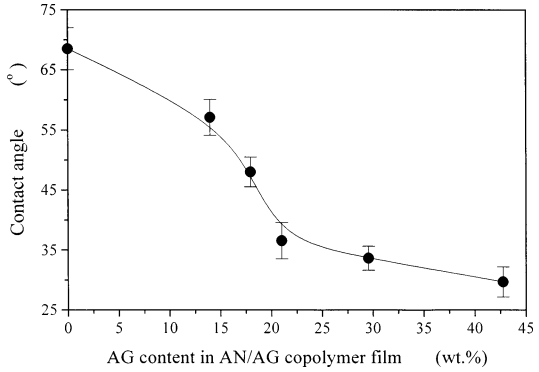

Figure 8. Relationship between the contact angle and AG content of the acrylonitrile/α-allyl glucoside copolymer films at 25 °C.

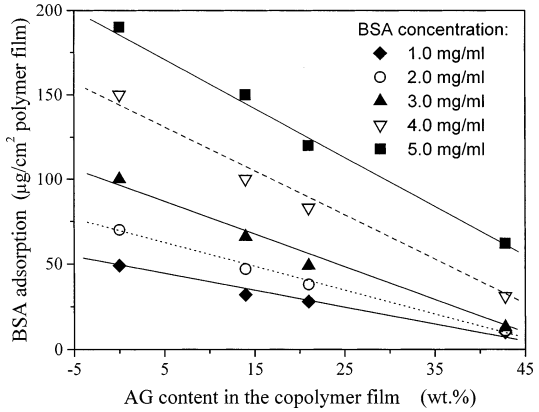

Figure 9. Adsorption of BSA onto the acrylonitrile/α-allyl glucoside copolymer films at 25 °C.

increasing the AG content in the copolymers from 0 to 42 wt %. This was due to the contribution of the hydroxyl groups in AG moieties.[27] The decreasing of the contact angle indicated that a kind of highly hydrophilic copolymer was obtained.

It is well-known that the hydrophobic interaction between material surfaces and proteins plays a very important role for nonselective adsorption of protein onto biomaterials. Usually, materials possessing hydrophilic surface show relatively low nonselective adsorption for proteins or cells. Carbohydrate-containing polymers are highly hydrophilic materials; however, some of them have the recognition function to the biomolecules or cells because of the cluster effect.[26,27] Therefore, the hydrophilicity and the recognition function of the carbohydrate moieties will have a different effect on the biomolecules or cells adsorption. Figure 9 illustrates the results of BSA adsorption on the AN/AG copolymer surface. It can be seen that the adsorbed amount BSA decreased almost linearly with the increase of AG content in the copolymer, and the high BSA concentration could lead to the larger amount of BSA adsorbed onto the copolymer films. The decrease of the BSA adsorption could be mainly ascribed to the improvement of the hydrophilicity by the carbohydrate moieties for the copolymer surface.

Macrophage is a kind of immune cell and performs various functions such as migration, phagocytosis, secretion, antigen presentation, and survival through precisely modulated adhesion, in living bodies. However, the molecular mechanism in macrophage adhesion is complex, dynamic, and not yet fully understood. Generally speaking, the fewer amount of macrophages adhered onto the materials surface indicates that the better of blood compatibility for the material, which is to say that the immunological reaction or immunological rejection will decrease after the materials are planted into the living body. The results of macrophages adhesion on the acrylonitrile/α-allyl glucoside copolymer surface are shown in Figure 10. It was clearly demonstrated that the number of macrophages adhered on the acrylonitrile/α-allyl glucoside copolymer was significantly decreased when compared with that on the polyacrylonitrile film, which means that the incorporating of α-allyl glucoside into polyacrylonitrile induced the reduction of macrophage adhesion. However, it seems that the macrophage adhesion number increased again when the AG content in the copolymer was exceeded around 20 wt %. This might be due to the cluster effect of the carbohydrate moieties in the copolymer chains. It was reported that the carbohydrate density strongly affects the specific interaction between carbohydrate moieties and cells, and the phenomenon that the binding affinity is drastically enhanced by multivalent carbohydrate ligands is called the cluster effect.[26]

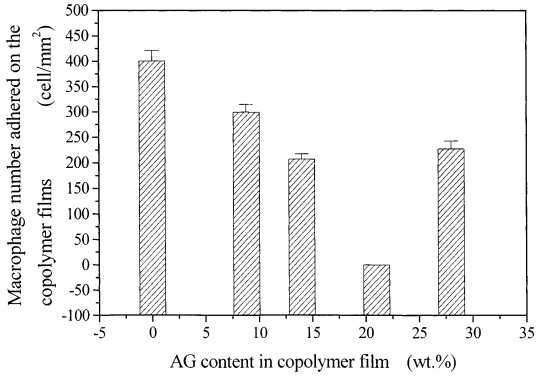

Figure 10. Relative macrophage number vs α-allyl glucoside content in the copolymer films at 25 °C.

Conclusions

Carbohydrate containing acrylonitrile-based copolymers could be simply synthesized by water-phase precipitation copolymerization of α-allyl glucoside with acrylonitrile. The contact angle of pure water, the amount of BSA absorption, and macrophages adhesion on the copolymer surface could be decreased to a certain level by increasing the carbohydrate monomer content in the copolymers. These preliminary results revealed that both the hydrophilicity and biocompatibility of polyacrylonitrile-based membranes could be improved by copolymerization acrylonitrile with vinyl carbohydrates. Further work concerning the membrane fabrication, characterization (especially fouling resistance), and applications from the resulted copolymers has been carrying out in our lab.

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