Synthesis and Water Absorbency of Polyampholytic Hydrogels with Antibacterial Activity

Xiaohong Li, Qinzheng Dong, Peixin He

Ministry-of-Education Key Laboratory for the Synthesis and Application of Organic Functional Molecules, School of Chemistry and Chemical Engineering, Hubei University, Wuhan 430062, People's Republic of China

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ABSTRACT: Amphoteric terpolymers of acrylic acid (AA), acrylamide (AM), and *N*,*N*′-dimethyl-*N*-ethylmethacryloxylethylammoniumbromide (DMAEA-EB) with varied compositions P[AA-AM-(DMAEA-EB)] were synthesized by inverse suspension polymerization. The components of P[AA-AM-(DMAEA-EB)] were verified by FTIR spectroscopy. The water absorption ability and antibacterial activity of the copolymer against *Escherichia coli* (*E. coli*) and *Staphylococcus hyicus* (*S. hyicus*) suspended in sterilized physiological saline were investigated. The introduction of $-N^+R_4$ may increase the water absorbency of P[AA-AM-(DMAEA-EB)] in some degree because of the excellent hydrophilicity of $-N^+R_4$. The AA-AM-(DMAEA-EB) hydrogels exhibited high antibacterial activity against bacteria tested. The process of adsorption between live bacteria cells and resins was at least partially reversible. A peak of antibacterial efficiency existed with increasing contact time. The resin killed 96.6% *E. coli* organisms and 90.3% *S. hyicus* organisms, respectively, within 30 min of contact at dosage of 0.1g. The concentration of DMAEA-EB has a special effect on the antibacterial activity of the polyampholytic hydrogels, which is different from polycation. It was observed that the antibacterial activity of the resin with 2 mol % of DMAEA-EB is superior to the copolymers tested with other compositions. © 2009 Wiley Periodicals, Inc. J Appl Polym Sci 112: 439–446, 2009

Key words: superabsorbent; unsaturated quaternary ammonium; acrylic acid; inverse suspension polymerization; antibacterial activity

INTRODUCTION

Superabsorbent polymer (SAP) is a new kind of functional macromolecule developed in the past 30 years. Many kinds of SAPs are commercially prepared and used in a variety of fields such as disposable diapers, feminine napkins, soil for agriculture and horticulture, water-block tapes, and absorbent pads.[1–3] Many methods have been attempted to improve the absorption properties and to expand the application fields of superabsorbents. Recently, research on the superabsorbents as antibacterial materials has attracted great attention. As reported, the percentage of the usage of superabsorbent as hygiene and cosmetic is above 80%. These applications require absorbent materials to have antibacterial activity besides water absorption or water retention properties.[1–5] Compared with conventional antibacterial agents of low molecular weight, polymeric antibacterial agents have the advantages of being nonvolatizable, chemically stable, and hard to permeate through the skin of a man or an animal.[6] For this reason, investigation on SAP with antibacterial activity represents a new direction in the field of SAP.

By the antibacterial activity of polymers, we mean a kind of interaction between polymers and bacteria, i.e., bactericidal or bacteriostatic action of polymers against bacterial cells or capturing bacterial cells by the polymers. In general, antibacterial agents like bactericides or disinfectants are low-molecular weight compounds. Phenols and cationic compounds are two main groups of compounds that are used almost exclusively for disinfectants. The latter covers many kinds of compounds differing considerably in chemical structure. Their common features are the presence of strongly basic groups attached to a fairly massive nonpolar molecule.[6] Among them, quaternary ammonium (QA) salts and biguanides are the best and most widely used antibacterial agents. Interestingly, they kill bacteria and fungi by interaction with the constituents of the cell envelope: interaction with the negative charges of the cell wall, destabilization, and weakening of the cytoplasmic membrane (because of their lipophilic moiety) leading to a loss of cytoplasm constituents because of the high osmotic pressure.[6] On these grounds, it is considered that electrostatic interaction of the

Correspondence to: P. He (qpxhe@hubu.edu.cn).

positive charges on the molecules of the antibacterial agents with the negatively charged species present in the cytoplasmic membranes (such as acidic phospholipids and membrane proteins) can play an important role in the course of the killing of bacteria using these bacterial agents.

From these viewpoints, some researchers studied the antibacterial activities of SAP by introducing antibacterial agents with low molecular weight into polymer chains like P(OADMAC-AM-MBAAm),[3] P(AM-*co*-4VP),[7] P(TRVB-AAm-MBAAm),[8] P(TRVB-NIPAAm-MBAAm),[9] PS-grafting QA groups.[10] These results undoubtedly provide an important and scientific basis for developing a new generation of polymeric antibacterial superabsorbent.

Following the earlier research work, the copolymerization of cationic monomers (QA salts or quaternary phosphonium salts) with nonionic monomers (acrylamide) by solution polymerization were reported. The absorption and antibacterial activity of the copolymer obtained were discussed at the same time.[8,9] However, there were few reports on the absorption and antibacterial activity of polymers consisting of cationic monomers and anionic monomers. In view of these, we introduced cationic groups ($-N^+R_4$) and anionic groups ($-COO^-$) into the polymer chains. The obtained resin is amphoteric polyacrylamide, which is representative of a special sort of polymers that contain both positively and negatively charged groups along the macromolecular backbone. A combination of cationic and anionic groups in a soluble polymer was found to be able to more effectively neutralize the charge in the sludge.[11] In our work, the combination of $-N^+R_4$ and $-COO^-$ for water-insoluble polymer can be found to increase the water absorbency of antibacterial resin.

Inverse suspension polymerization has proved to be a suitable technique for the production of SAP with high water absorbency. Inverse suspension polymerization of water-insoluble polymers in organic media has been studied by many researchers.[12–14] However, only a few studies have carried out inverse suspension polymerization of amphoteric polyelectrolyte in organic media, and there are few reports on amphoteric antibacterial SAP.

In this article, we reported the synthesis of a series of polyampholytic hydrogels with antibacterial activity by inverse suspension polymerization of acrylic acid (AA), acrylamide (AM), and *N*,*N*′-dimethyl-*N*-ethylmeth-acryloxylethylammoniumbromide (DMAEA-EB). The main factors, which affect the water absorbency and antibacterial activity of SAP were discussed, and the antibacterial activity of hydrogels against *Escherichia coli* (*E. coli*) and *Staphylococcushyicu*s (*S. hyicu*s) was studied

EXPERIMENTAL

Materials

AM was purchased from Tianjin Bodi Chemical Factory (Tianjin, China), and it was recrystallized in acetone solvent before use. AA was distilled under reduced pressure. Sodium acrylate was prepared from AA; *N*,*N*′-methylenebisacrylamide (NMBA) and potassium persulfate (KPS) were obtained from Shanghai Chemical Reagent (Shanghai, China) and recrystallized in distilled water; 2-(dimethylamino) ethylmathacrylate (DMAEA) was obtained from Xinyu Chemical Industries (Wuxi, China). Mono-octadecyl phosphate was synthesized in our laborotories.[4] All the reagents were of analytical grade.

Peptone and agar were purchased from Wuhan Tianyi (Hubei, China). Yeast extract was procured from Guoyao Chemical Reagent (Shanghai, China). All reagents were biochemical reagent. *E. coli* (XL10-gold) and *S. hyicus* (NATSEL0502) were provided by the School of Life Sciences of Hubei University.

Synthesis of DMAEA-EB monomer

DMAEA and acetone with equal volume and 0.00034 g/mL inhibitor (according to the dosage of DMAEA) were added into a 100-mL flask with a reflux condenser.[15] The mixture was magnetically stirred until the inhibitor dissolved and then charged with equal volume of bromoethane. After that, it was heated at 45°C in water bath for 10 h, and then cooled in air, filtered, and cleaned with ethyl ether repeatedly. The product was dried under vacuum.

Inverse suspension polymerization

P[AA-AM-(DMAEA-EB)] beads were prepared by inverse suspension polymerization using cyclohexane as the continuous phase and mono-octadecyl phosphate as nonionic surfactant.

NaAAc solution, obtained by neutralizing AA, AM, DMAEA-EB, and NMBA were dissolved in 10 mL of deionized water containing a certain amount of KPS, and the mixture was bubbled with dry nitrogen to remove dissolved oxygen. This solution was slowly poured into 30 mL of cyclohexane containing 0.15 g of mono-octadecyl phosphate, which was previously dissolved at 65°C ± 2°C and purged with dry nitrogen in a 100-mL three-necked round-bottom flask. The reaction mixture was mechanically stirred at 250 rpm under nitrogen atmosphere. The polymerization was allowed to proceed for 2 and a half hours at 72°C. After polymerization, the beads was separated from the oil phase and were washed several times with the mixture of water and ethanol (1 : 9 v/v). The product was then dried in an oven at

60°C to constant weight. Then, fine white particles were obtained.

Measurement of water absorbency of the resin

Suction filtration method

Accurately weighed 100 mg dry SAPs (W) were immersed in a 500-mL beaker containing an excess amount (V_0) of deionized water or 0.9 wt % NaCl (aq) solutions for 24 h to reach the swelling equilibrium at room temperature. The completely swollen copolymer was filtered through a 100-mesh standard screen at least for 15 min until no water being filtered. Then the volume of filtered water V_1. was measured.

Water content (Q_s) was calculated as follows:

$$Q_s = \frac{(V_0 - V_1)}{W} \times \rho_{H_2O}$$

where V_0, V_1 are the initial and filtered volume of solution, respectively, W is the weight of the dry resin, ρ_{H_2O} is the density of water.

Measurement of antibacterial activity

Preparation of nutrient and the initial cell suspension

All procedures in the antibacterial tests for polymers were carried out under aseptic conditions and performed using a batch method.

Freeze-dried ampoules of *E. coli* and *S. hyicus* were opened, and a loopful of each culture was spread to give single colonies on nutrient agar and incubated at 37°C for 24 h. Three representative colonies were selected with a wire loop, placed in 5-mL nutrient broth (peptone, 1.0 g; NaCl, 1.0 g; beef extract, 0.5 g in 100 mL sterile distilled water [pH 7.2]), and then incubated at 37°C 150 r/min overnight. Cell suspension (0.5 mL) was pipetted out from the container and quickly mixed with 4.5 mL of sterilized physiological saline, and then decimal serial dilutions were prepared from this by taking 0.5 mL into 4.5 mL of sterilized physiological saline and was stirred. The cell number was determined by a colony count method. At this stage, the culture of *E. coli* or *S. hyicus* contained about 10^9–10^{10} CFU/mL, respectively.[7–11]

Contact of polymer gels with bacteria

Copolymers (0.1 g) with optimized experimental results and 20 mL sterile 0.9 wt % NaCl solution were placed in 100-mL conical flask with cotton stopper, and the polymers were swollen for 24 h. After that, 0.1 mL of the cell suspension were added to the flask. The mixture was continually shaken at 37°C 150 r/min. At prescribed time intervals, 0.5 mL of the cell suspension was pipetted out from the container and quickly mixed with 4.5 mL of sterilized physiological saline, and then decimal serial dilutions were prepared according to the method described in "Preparation of nutrient and the initial cell suspension" section. The colonies were counted after the inoculated plates were incubated at 37°C for 24 h. Blank test was finished by the same means without polymer. The counting was done in triplicate every time.

Measurement of viable cell numbers after contacting with terpolymer gels

Copolymers (0.1 g) with varying DMAEA-EB content and 20 mL sterile 0.9 wt % NaCl solution were placed in a 100-mL flask, and the polymers were swollen for 24 h, After that, 0.1 mL of the cell suspension (*E. coli* and *S. hyicus*) were added to the flask, which was shaken for 30 min at 37°C 150 r/min. The suspension pipetted was diluted several times and treated with means as reported in "Contact of polymer gels with bacteria" section. Blank test was finished in the same manner without polymers. The antibacterial efficiency η was calculated as follows[2,3]:

$$\eta = \frac{(N_0 - N_1)}{N_0} \times 100\%$$

where N_0 and N_1 are the number of viable cells of blank sample and the tested sample, respectively.

Characterization methods

FTIR spectra of P(AA-AM) and P[AA-AM-(DMAEA-EB)] were recorded using Perkin–Elmer Infrared Spectrometer (FTIR, Model 1760×). The copolymer was ground with the dried KBr. The copolymer–KBr powder was dried, pressed, and the KBr discs obtained were subjected to FTIR spectrophotometer analysis.

RESULTS AND DISCUSSION

FITR analysis of the terpolymer

The FITR spectra of P(AA-AM) and P[AA-AM-(DMAEA-EB)] containing 2 mol % DMAEA-EB are shown in Figure 1. The wide peak centered around 3422 cm^{-1} (Fig. 1) may be assigned to the combination of stretching vibrations of —N—H and —O—H. The corresponding peak on Curve 2 is more intense and wider because of the introduction of the more hydrophilic group of $-N^+R_4$[16] in P[AA-AM-(DMAEA-EB)]. The peaks around 3188 cm^{-1} (—OH

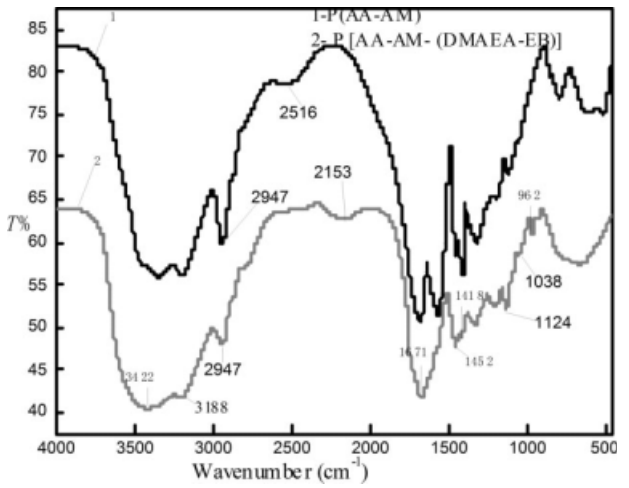

Figure 1 Infrared spectra of P(AA-AM) and P[AA-AM-(DMAEA-EB)].

stretching) and 1124 and 1038 cm^{-1} (C—O stretching) are the characteristic peaks of —COOH and $—COO^-$.[17] The peak at 2947 cm^{-1} is the stretch vibration of —C—H. Compared with the spectrum of P(AA-AM), the band at 2516 cm^{-1} (O—H stretch vibration of the carboxylic group) shifts to 2153 cm^{-1}.[18] Curve 2 also showed peaks at 1671 cm^{-1} for —C=O in $—CONH_2$, 1418 cm^{-1} for $—COO^-$. Comparing the two curves, the IR spectrum of P[AA-AM- (DMAEA -EB)] indicated the presence of QA salt via the peaks at 962 and 1452 cm^{-1}.[11,19]

Effect of reaction factors on water absorbency

Effect of the molar ratio of AA/AM on the water absorbency of the AA-AM-(DMAEA-EB) copolymer gels

According to Flory's theory, the following equation was used:

$$q_m^{5/3} = \frac{(i/2v_u S^{*1/2})^2 + (1/2 - x_1)/v_1}{V_0/v_e}$$

where q_m is the water absorbency of the network at equilibrium, $i/2v_u$ is the concentration of fixed charge in a unswollen network, S^* is the ionic concentration in the external solution, $(1/2 - x_1)v_1$ is the affinity of the gel for water, and V_0/v_e is the crosslinking density of the gel. Hence, the water absorbency of the gel was dependent on the ionic osmotic pressure, the crosslinking density, and the affinity of the gel toward water. Because the crosslinker content, S^*, and ionic osmotic pressure were fixed, the water absorbency of the gels was dependent on the affinity of the gel for water. The results in Figure 2 show that the water absorbency increased with increasing molar ratio of AA/AM when the molar ratio was less than 1 : 1. This result shows that the addition of a small amount of the hydrophilic monomer AA to the copolymer gels increased the affinity of the gels for water, and the water absorbency increased with increasing molar ratio of AA. However, when the molar ratio of AA/AM was greater than 1 : 1, the higher the AA content, the lower was the water absorbency. This is because the excess amount of AA can produce intramolecular or intermolecular hydrogen bonding in two neighboring chains with AM. This may have also caused the gels to shrink.

Effect of DMAEA-EB content on the water absorbency of the AA-AM-(DMAEA-EB) copolymer gels

As shown in Figure 3, the water absorbency of copolymer has a maximum with the increase of the DMAEA-EB content. The water absorbency of gels is dependent on ionic osmotic pressure, crosslinking density, and the affinity of the gel for water. The crosslinking density of copolymer and the ionic concentration in the external solution were fixed. The water absorbency for the gel is dependent on the concentration of the fixed charge in the gel and the affinity of the gel for water. When the cationic monomer DMAEA-EB was introduced into the copolymer, the bromide ion (Br^-) was dissociated and the QA group (R_4N^+) with a positive charge was formed. The affinity of the QA group (R_4N^+) for water is stronger than that of the carboxylate group (COO^-). However, the QA group (positively charged) would bind with the carboxylate group (negatively charged), so the fixed charge concentration of the polymer network decreases. This behavior reduces the negative charge repulsion of the

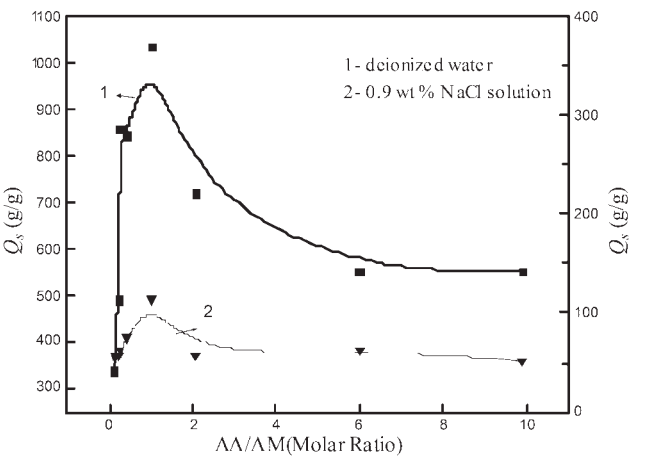

Figure 2 Effect of AA/AM ratio on water absorbency of copolymer in deionized water and 0.9 wt % NaCl solution, respectively. [Conditions: DMAEA-EB, 2 mol %; NMBA, 0.01 mol %; $K_2S_2O_8$, 0.0754 mol %; neutralization degree, 75%; monomer concentration, 5 mol/L.]

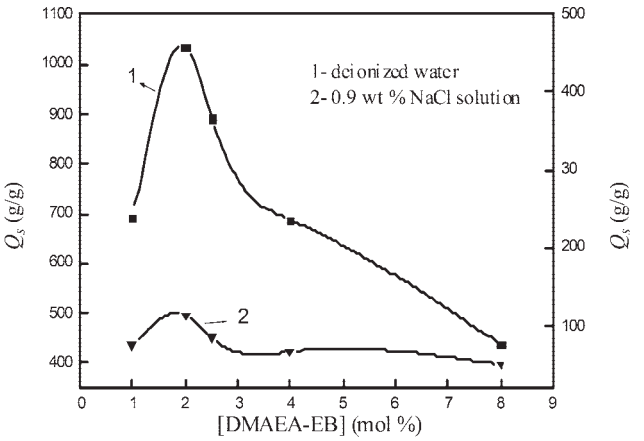

Figure 3 Effect of DMAEA-EB content in the gels on the absorbency of copolymer in deionized water and 0.9 wt % NaCl solution, respectively. [Conditions: AA/AM, 1 : 1 (mol); NMBA, 0.01 mol %; $K_2S_2O_8$, 0.0754 mol %; neutralization degree, 75%; monomer concentration, 5 mol/L.]

polymer network, and the water absorbency of the gel decreases. On the other hand, the results observed from Figure 3 also indicate that the water absorbency in 0.9 wt % NaCl(aq) is lower than that in deionized water. This is attributed to an increase of the concentration of the sodium ion in the external solution, and the difference of ionic osmotic pressure between the gel and the external solution decreases. Thus, the water absorbency of the gel significantly decreased.

Effect of the content of the crosslinking agent on the water absorbency of the AA-AM-(DMAEA-EB) copolymer gels

Table I shows that the water absorbency varies with the increase of crosslinking agent concentration. The water absorbency increased with increasing crosslinking agent concentration and then decreased. The results suggest that there is an optimal value for the water absorbency.

When the molar ratio of NMBA is 0.01(mol %), the water absorbency is 1035 g/g in deionized water. This phenomenon obeys the P. J. Flory theory. Water absorbency is also related to the elastic force between the polymer chains. With more crosslinking agent, the higher the crosslinking density, the stronger was the elastic force of the polymer chain and the lower the water absorbency. In general, crosslinking agents have been employed to help improve the strength of the swollen gel, but they are also very effective in reducing the water absorbency. It is notable that if the crosslinker concentration is too low (for example, less then 0.01 mol %), the solubility of the resin increases as the crosslinking density of copolymer decreases.

TABLE I
Effect of the Content of the Crosslinking Agent on Water Absorbency of Copolymer in Deionized Water and 0.9 wt % NaCl Solution

[NMBA] (mol %)	Q_s(g/g) in deionized water	Q_s(g/g) in 0.9 wt % NaCl
0.005	460	51
0.01	1,035	114
0.03	543	75
0.05	338	55
0.07	251	56

Conditions: AA/AM/(DMAEA-EB), 49 : 49 : 2(mol %); $K_2S_2O_8$, 0.0754 mol %; neutralization degree, 75%; monomer concentration, 5 mol/L.

Effect of initiator (KPS) concentration on the water absorbency of the AA-AM-(DMAEA-EB) copolymer gels

The effect of initiator concentration on water absorbency of the crosslinked copolymers is shown in Figure 4. The water absorption capacity of the copolymers increased with increasing initiator concentration up to an optimum amount of 0.0754 mol % $K_2S_2O_8$ and then decreased.

Based on general kinetics, the rate of polymerization depends on the concentration of monomers and initiators for a bimolecular termination. When heated alone or in the presence of a reducing agent, the persulphate ions ($S_2O_8^{2-}$) in aqueous solution decompose to sulfate radical ions ($2SO_4^{-\cdot}$ or $2KSO_4$). These primary radicals will initiate the monomers to

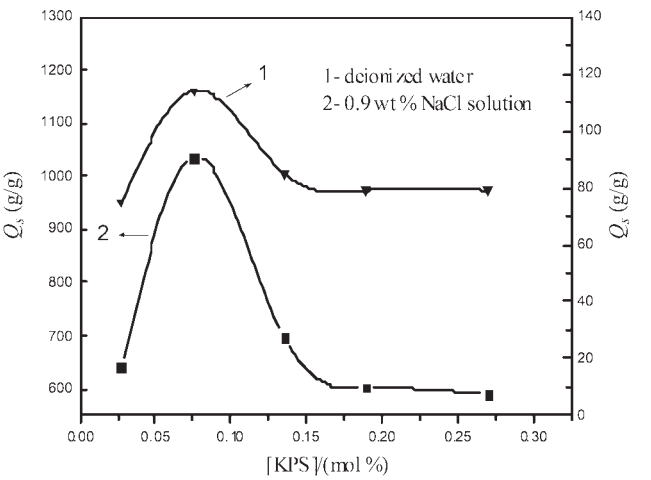

Figure 4 Effect of initiator concentration on water absorbency of copolymer in deionized water and 0.9 wt % NaCl solution, respectively. [Conditions: AA/AM/(DMAEA-EB), 49 : 49 : 2 (mol); NMBA, 0.01 mol %; neutralization degree, 75%; monomer concentration, 5 mol/L.]

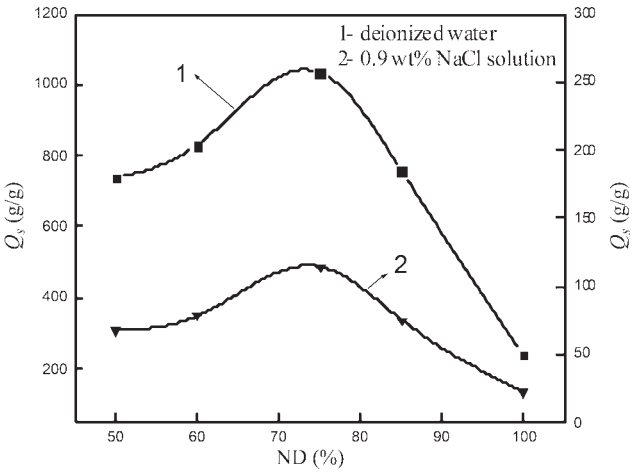

Figure 5 Effect of neutralization degree (ND) of AA on water absorbency of copolymer in deionized water and 0.9 wt % NaCl solution, respectively. [Conditions: AA/AM/(DMAEA-EB), 49 : 49 : 2 (mol); NMBA, 0.01 mol %; $K_2S_2O_8$, 0.0754 mol %; monomer concentration, 5 mol/L.]

form free radicals of monomers, which propagate monomer molecules in succession to form a large polymeric radical and a dead polymer in the termination step. Thus, in free-radical polymerization, the initiator has an effect on both the polymerization rate and the molecular weight of a polymer. In the process of crosslinking polymerization, the initiator also affects the crosslinking degree and molecular weight between two crosslinking points. The lower initiator concentration results in the decrease of the crosslinking degree and the conversion, whereas based on the principle of kinetic chain length, the polymer molecular weight will increase with decreasing initiator concentration. However, when the initiator concentration is higher, the synthesized copolymer has a high crosslinking density, which affects the water absorbency.

Effect of the neutralization degree on the water absorbency of the AA-AM-(DMAEA-EB) copolymer gels

The degree of neutralization of AA not only affected the polymerization rate but also determined the charge number in the three-dimensional network of the P[AA/AM/ (DMAEA-EB)], and therefore it had a marked influence on water absorbency. The degree of neutralization of AA is defined as the molar percentage of carboxyls in AA neutralized by sodium hydroxide. Figure 5 shows that there is a maximum in the dependence of water absorbency on the degree of neutralization. The water absorbency increases as the degree of neutralization increases from 50 to 75% and then decreases. This is because the activity of AA is higher than that of acrylate, so the lower the neutralization, the faster is the polymerization rate. The high polymerization rate would result in the increase of the crosslinking density of hydrogel. At the same time, the charge density of the network would increase with the increase of the degree of neutralization. This would result in the increase of the stretching extent of the hydrogel network and enhance osmotic pressure. When the degree of neutralization was higher than 75%, the polymerization rate would decrease and the content of acrylate would increase with the increasing degree of neutralization. The polymers thus obtained suffered from the alkaline environment, which would affect the dissociation of sodium acrylate. In addition, the concentration of the fixed charge on the gel network decreases, and the negative charge repulsion between the polymeric network becomes weaker. Consequently, the chain segment would freeze and not stretch, so water absorbency decreased.

Antibacterial activity of P[AA-AM-(DMAEA-EB)]

The effect of contact time on antibacterial activity of copolymer gels

Figure 6 shows the influence of different contact time between bacteria and resins on antibacterial efficiency (η). This experiment mainly investigated antibacterial activity of the copolymer with the highest absorbency against *E. coli* and *S. hyicus*. According to Figure 6, the value of η increases as the adding of contact time till the peak appears. This result is similar to that reported in Ref. 10, in which

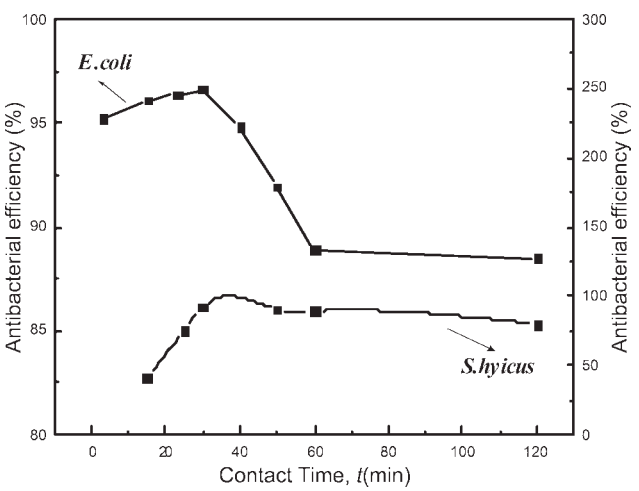

Figure 6 Effect of contact time on antibacterial efficiency η of copolymer gels in 0.9 wt % NaCl solution. [Conditions: AA/AM/(DMAEA-EB), 49 : 49 : 2 (mol); NMBA, 0.01 mol %; $K_2S_2O_8$, 0.0754 mol %; neutralization degree, 75%; initial viable cell concentration of *E. coli* suspension, 4.69×10^{10} CFU/mL; initial viable cell concentration of *S. hyicus* suspension, 4.78×10^9 CFU/mL; copolymer, 0.100 g.]

the antibacterial process was summarized as chemical reactions in the following way:[10]

$$L + P \longleftrightarrow \text{LP physical adsorption of viable cells} \quad (1)$$

$$D + P \rightarrow \text{DP physical adsorption of dead cells} \quad (2)$$

$$L + P \rightarrow \text{DP killing of the viable cells} \quad (3)$$

where L is the viable cells, P is the insoluble ammonium polymers, D is the dead cells, LP is the living cells adsorbed on polymers, and DP is the dead cells adsorbed on polymers. Equation (1) shows a direct decrease in the number of viable cell in suspension, whereas eq. (3) is the key step in killing the bacteria. It is assumed that the adsorption of viable cells is a reversible process, but the adsorption of the dead cells is an irreversible one. Because dead cells lose their mobility, the interaction between dead cells and ammonium groups is strong.

As shown in Figure 6, the copolymer may first absorb the living bacterial cells rather than kill them, but keep them alive in the first 30 min. This may result in the increase of η. As shown in reaction (1), the absorption and release of the viable cells is a reversible process. The agent may also absorb the dead cells, as shown in reaction (2); the process is supposed to be an irreversible one. When the dead cells accumulate on the surface of the agent and shield the cationic center, the ammonium salt cannot absorb the living cells any more, and the previous absorbed viable cells are released from the insoluble ammonium salt. Therefore, the number of viable cells in suspension would increase after contacting with copolymer. This results in the decrease of antibacterial efficiency (η) of copolymer tested after contacting with bacteria for 30 min.

The effect of DMAEA-EB content on antibacterial activity of copolymer gels

At certain contact time (30 min), the antibacterial activity of P[AA-AM-(DMAEA-EB)] with different contents of DMAEA-EA (mol %) against *E. coli and S. hyicus* was investigated. As shown in Figure 7, the concentration of DMAEA-EB has a special effect on the antimicrobial activity of polyampholytic hydrogels, which is different from normal polycation. In general, the higher the content of antimicrobial agent in normal polycation, the better is the antibacterial activity.[10–15] However, the antibacterial activity of polyampholytic hydrogels in our experiment is dissimilar. This result can be confirmed by its influence on the water absorption. After the swelling of P[AA-AM-(DMAEA-EB)], the chains expand and the groups ionize, such as $-COO^-$ and $-N^+(R_4)\ Br^-$, to form two kinds of charges. The cooperation of the charges may be the main reason for the earlier difference, which requires further investigation.

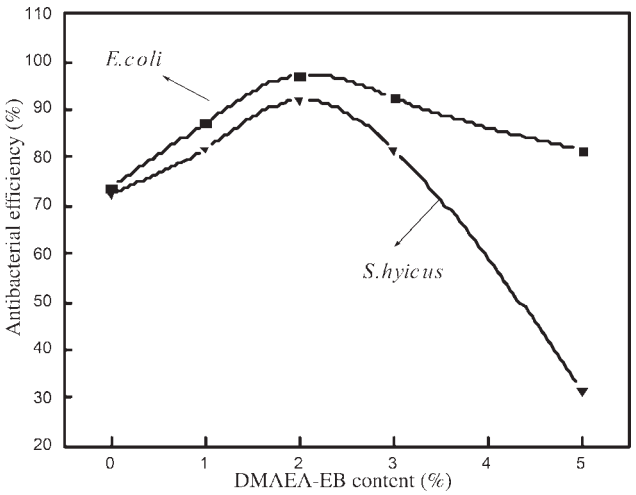

Figure 7 Effect of DMAEA-EB content on antibacterial efficiency η of copolymer gels in 0.9 wt % NaCl solution. [Conditions: AA/AM, 1 : 1 (mol); NMBA, 0.01 mol %; $K_2S_2O_8$, 0.0754 mol %; neutralization degree, 75%; initial viable cell concentration of *E. coli* suspension, 4.69×10^{10} CFU/mL; initial viable cell concentration of *S. hyicus* suspension, 4.78×10^9 CFU/mL; copolymer, 0.100 g; contact time, 30 min.]

CONCLUSION

A series of polyampholytic hydrogels with antibacterial activity were synthesized by inverse suspension polymerization. The effect of various polymerization parameters such as the initiator concentration, crosslinking agent, neutralization degree, and the molar ratio of AA/AM/(DMAEA-EB) on water absorbency of the hydrogels were studied. Experimental results indicated that the introduction of $-N^+R_4$ may increase the water absorption ability of the hydrogels to a certain degree, which can be attributed to the excellent hydrophilicity of $-N^+R_4$.

The antibacterial activity of P[AA-AM-(DMAEA-EB)] in 0.9 wt % NaCl solution was tested against *E. coli* and *S. hyicus*. It was found that the antibacterial efficiency varied according to the contact time and the content of DMAEA-EB in the copolymer. Besides, it also depends on the microorganism examined.

The AA-AM-(DMAEA-EB) terpolymers exhibited high antibacterial activity against *E. coli* and *S. hyicus.* There is a reversible adsorption process between bacteria cells and resins. As the contact time increased, a peak existed before the antibacterial activity reached a balance. Furthermore, the concentration of DMAEA-EB has a special effect on the antibacterial activity of polyampholytic hydrogels; the

antibacterial activity increased to a peak value then decreased with increasing content of DMAEA-EB in the terpolymers. This result is different from the behavior of a normal polycation. The cooperation of $-COO^-$ and $-N^+R_4$ on stretched chain may be the main reason of this character. Furthermore, AA-AM-(DMAEA-EB) terpolymers are water-insoluble polymer hydrogels. These hydrogels can be separated by filtration after contacting with bacterial suspensions. Therefore, residual toxicity of bactericides in water can be prevented.

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