

Preparation and Characterization of Thermosensitive Beads with Macroporous Structures

Caihua Ni,^{1,2} Zheng Wang,^{2,3} Xiao Xia Zhu^{2,3}

¹School of Chemical and Materials Engineering, Southern Yangtze University, Wuxi, China

²State Key Laboratory of Functional Polymers for Adsorption and Separation, Nankai University, Tianjin, China

³Department of Chemistry, University of Montreal, C.P. 6128 Succursale, Centre-ville, Montreal, QC, H3C 3J7 Canada

Received 3 December 2002; accepted 27 June 2003

ABSTRACT: Thermosensitive copolymers of poly(*N*-isopropyl acrylamide)-*co*-2-hydroxyethyl methacrylate (NIPA-*co*-HEMA) macroporous resins were synthesized in the form of beads by inverse suspension polymerization. The copolymerization was carried out in aqueous solutions of the comonomers dispersed by cyclohexane with stabilizers. A series of resins with different molar ratios of NIPA : HEMA, and different crosslinking degrees was obtained. The compositions of the copolymers were determined by elemental analysis. The results showed that the content of HEMA in a copolymer was higher than that of the corresponding feed mixture from which the copolymer was made. IR spectra also confirmed the structure of the copolymers. The porous

parameters such as true densities, apparent densities, pore volumes, and porosities of the resins were measured by means of pycnometry. The determination of equilibrium swelling ratios and measurement of differential scanning calibration indicated that the resins exhibited thermosensitivity in aqueous solutions. Finally, the loading of hydroxyl groups was determined by titration. The resins have potential applications as supports in solid-phase synthesis after being functionalized. © 2003 Wiley Periodicals, Inc. *J Appl Polym Sci* 91: 1792–1797, 2004

Key words: macroporous polymers; functionalization of polymers; resins; dispersions; thermosensitive hydrogels

INTRODUCTION

Thermosensitive hydrogels have attracted much attention since Tanaka first observed the gel collapse phenomenon.¹ Aqueous solutions of poly(*N*-isopropyl acrylamide) [poly(NIPA)] and poly(*N,N'*-diethylacrylamide) exhibit a lower critical solution temperature.^{2,3} When crosslinked they show discontinuous volume phase transition in water in response to a change of temperature.^{4–7} Because of this unique property thermosensitive hydrogels have received wide applications in solute recovery, drug delivery, immobilized enzyme reactors, and superabsorbents.^{8–16}

Since Marrifield first introduced the solid-phase synthesis technique for preparation of polypeptides, resins based on polystyrene backbone have already been extensively developed as polymeric supports, although reports of hydrogels used for this purpose have been scarce. Usually a desirable polymeric support should have two important characteristics: its backbone should have good swellability in solvents,

and it should contain reactive functional groups so as to be readily functionalized for further reactions. To satisfy these requirements we designed the copolymer as our resultant polymeric support. When the reaction was completed the solution was heated to above the phase-transition temperature of poly-NIPA, the copolymer support therefore shrinks, so that the solid support is separated from the reaction system.

There were two reports about redox copolymerization of NIPA and 2-hydroxyethyl methacrylate (HEMA).^{17,18} Three-dimensional gel structures were obtained in the studies. The objective of this study was to synthesize new types of thermosensitive resins that have potential applications in solid-phase synthesis. In this work we synthesized the beaded copolymer of NIPA-*co*-HEMA by inverse suspension polymerization, optimized the synthetic conditions, characterized the structures, and investigated the thermosensitive properties of the copolymers. Figure 1 shows the reaction scheme.

EXPERIMENTAL

Materials

N-Isopropylacrylamide (NIPA), 2-hydroxyethyl methacrylate (HEMA), *N,N'*-methylene(bis)acrylamide (BIS), *N,N,N',N'*-tetramethylethylenediamine (TEMED), ammonium persulfate (APS), and cyclohexane were purchased from Aldrich Chemical Company (Milwaukee,

Correspondence to: C. Ni (caihuani@yahoo.com).

Contract grant sponsor: Natural Science and Engineering Research Council (NSERC) of Canada.

Contract grant sponsor: The Educational Committee of Hubei Province.

Contract grant sponsor: China Scholarship Council.

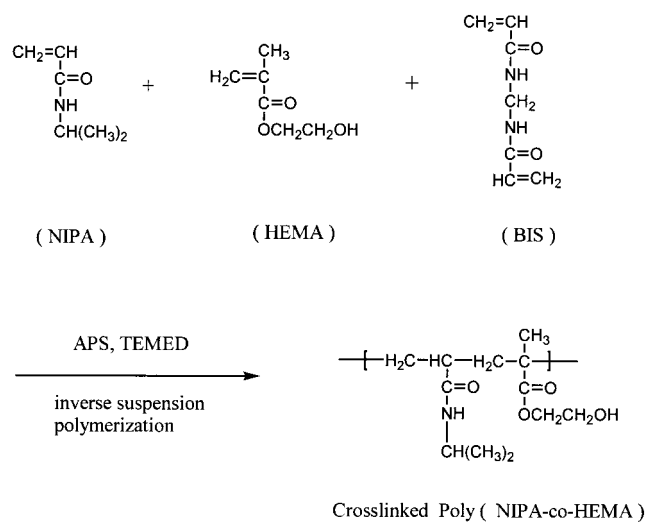


Figure 1 Scheme of polymerization.

WI). HEMA was redistilled before use to remove inhibitor. The others chemicals were used as received without further purification.

Preparation of copolymers

The resins were prepared by free-radical copolymerization of NIPA-co-HEMA using BIS as the crosslinker. Cyclohexane (150 mL) was charged to a three-neck flask equipped with a mechanical stirrer, condenser, and thermometer. Sorbitan monopalmitate (span 40; 0.4 g) and sorbitan monooleate (span 80) were added as stabilizers. Nitrogen gas was passed through the solution for 20 min. In a small beaker 2.26 g (0.02 mol) of NIPA, 2.60 g (0.02 mol) of HEMA, and 0.185 g of BIS were dissolved in 25 mL deionized water. APS solution (1.2 mL; 0.04 g/mL) and 0.1 g TEMED were introduced to initiate the reaction. The mixture was promptly poured into the experimental flask at a stirring speed of 300 rpm. The reaction temperature rose slowly, within 1 h, from 22 to 72°C, at which temperature it was kept constant for 6 h until completion of the reaction. The product was washed with acetone and distilled water successively, after which it was freeze-dried. The resin was largely beaded but contained a slight amount of amorphous polymers. Other analogous copolymer resins with various molar ratios and amounts of crosslinker were prepared under similar conditions.

Characterization

Compositions of some copolymers were determined by elemental analysis (EA 1108 CHN; Fisons Instruments; Isomass Scientific, Inc., Calgary, Canada) and the structure was also confirmed with FTIR spectrom-

etry (Bomen MB-100, ABB Bomen, Inc., Quebec, Canada). Potassium bromide pellets were used.

True density and apparent density were determined by pycnometry. An accurately weighed sample was placed in a 10-mL volumetric flask weighed previously. Cyclohexane (9 mL) was added and the resin was soaked for 24 h. Cyclohexane was refilled up to 10 mL. The flask with beads and cyclohexane was weighed. Apparent density could be estimated with the method described in the literature.¹⁹ The cyclohexane-absorbed resin was then filtrated and centrifuged for 2 min at 2000 rpm. The wet resin was weighed. True density (ρ_t) and apparent density (ρ_a) were calculated as follows:

$$\rho_t = \rho \frac{W_d}{10\rho - (W_2 - W_d)} \quad (1)$$

$$\rho_a = \rho \frac{W_d}{10\rho - (W_2 - W_w)} \quad (2)$$

where W_d , W_w , and W_2 are the weights of dry beads, wet beads, and the sum of beads and cyclohexane in the flask, respectively. ρ is the density of cyclohexane at the given temperature. Porosity (P) and pore volume (V_p) were calculated by the following formulas²⁰:

$$P = 1 - (\rho_a / \rho_t) \quad (3)$$

$$V_p = (1 / \rho_a) - (1 / \rho_t) \quad (4)$$

The swelling ratios of the resins were measured at various temperatures. The dried resins to be tested were placed in a tea bag made of nonwoven fabric and then were soaked in excess deionized water for 8 h at 20°C. The water unabsorbed into resin was removed by filtration and water on the surface of the tea bag was immediately wiped with filter paper, after which they were weighed. An empty tea bag without sample was treated in the same way as a blank. When they were measured at different temperatures the resins were soaked at each temperature for 8 h. The swelling ratio was calculated using the equation

$$S_R = \frac{W_w - W_d}{W_d} \quad (5)$$

where S_R is the swelling ratio and W_w and W_d are the weights as indicated above.

Thermal analysis of the volume-phase transition was performed using a differential scanning calorimeter (DSC 2910; TA Instruments, New Castle, DE). The samples were placed and sealed in DSC cells and were scanned against an empty reference cell from 0 to 55°C at a heating rate of 10°C/min.

TABLE I
Synthetic Results of Poly(NIPA-co-HEMA) Resins^a

Resin	NIPA		HEMA		Molar ratio	BIS		Yield (%)
	(g)	(mol)	(g)	(mol)		(g)	(mol %)	
PNH-1	4.07	0.036	0.52	0.004	9 : 1	0.185	3	71.5
PNH-2	3.62	0.032	1.04	0.008	8 : 2	0.185	3	75.0
PNH-3	1.58	0.028	0.56	0.012	7 : 3	0.185	3	77.9
PNH-4	2.71	0.024	2.08	0.016	6 : 4	0.185	3	80.9
PNH-5	2.26	0.020	2.60	0.020	5 : 5	0.185	3	79.4
PNH-6	1.36	0.012	3.64	0.028	3 : 7	0.185	3	76.7
PNH-7	2.26	0.020	2.60	0.020	5 : 5	0.062	1	68.3
PNH-8	2.26	0.020	2.60	0.020	5 : 5	0.123	2	73.1
PNH-9	2.26	0.020	2.60	0.020	5 : 5	0.246	4	78.4
PNH-10	2.26	0.020	2.60	0.020	5 : 5	0.308	5	81.2

^a The reactions were carried out at 72°C for 8 h; initiator APS, 1.0%; accelerator, TEMED 2.1 wt %; water, 25 mL; cyclohexane, 150 mL; surfactant, 0.6 g.

Loading of hydroxyl groups was determined by titration after the resins reacted with acetic anhydride. Resin (0.2 g), acetic anhydride (1.0 mL), and pyridine (4.0 mL) were mixed in a small flask. The reaction was carried out at 22°C for 3 h and 40°C for 30 h. After that 5.0 mL of water was introduced to convert excess acetic anhydride into acetic acid. The quantity of acetic acid was titrated by 0.5N NaOH aqueous solutions with indicator phenolphthalein.

RESULTS AND DISCUSSION

Preparation of the copolymers

Inverse suspension polymerization of spherical hydrogels has been studied less often than normal suspension polymerization, especially with respect to thermosensitive polymers.²¹⁻²⁴ In previous studies paraffin oil, dichloromethane, and xylene were used for the continuous phase. To minimize the solubility of NIPA in organic solvent we selected cyclohexane as a continuous phase for facilitating the reaction and increasing the yield.

To stabilize monomer droplets in continuous phase, surfactants of water in oil type are needed for inverse

suspension polymerization. In this case nonionic surfactants are desirable. Suitable hydrophilic-lipophilic balance (HLB) values of surfactants for an ordinary water/oil system are 6-7.²⁵ It is known that blend of surfactants with different HLB values can stabilize emulsion more efficiently than can a single surfactant.²⁶ Some nonionic surfactants were tested in this study. It was found that the blend of sorbitan monooleate (HLB 4.3) and sorbitan monopalmitate (HLB 6.7) was optimal surfactants for our purpose.

BIS was used as a crosslinker in the study. The amount of crosslinker was changed to investigate the effects of crosslinker on the yields and properties of the products. We noticed that the higher the amount of crosslinker used, the easier the reaction and the higher the yields obtained.

The yield of the resultant resins depends on many factors such as molar ratio of reactants, amount of crosslinker or initiator, concentration, reaction temperature, and time. The relationship between yield and time was studied. Yields were 52, 67, 76, 82, and 83%, respectively, when the reaction time was through 2, 4, 6, 10, and 15 h, respectively, under the following conditions: molar ratio of NIPA to HEMA, 6 : 4;

TABLE II
Elemental Analysis and Composition of Poly(NIPA-co-HEMA)

Resin	Molar ratio of NIPA / HEMA		Analysis (%)					
			Calc ^a			Experimental		
	In feed	In copolymer ^b	N	C	H	N	C	H
PNH-1	0.9 : 0.1	0.73 : 0.27	11.3	62.5	9.4	9.1	58.2	9.3
PNH-2	0.8 : 0.2	0.66 : 0.34	9.7	61.8	9.2	8.2	57.4	9.5
PNH-3	0.7 : 0.3	0.63 : 0.37	8.7	60.7	9.0	7.8	58.2	9.6
PNH-4	0.6 : 0.4	0.50 : 0.50	7.4	59.9	8.8	6.2	56.8	8.7
PNH-5	0.5 : 0.5	0.34 : 0.66	5.9	59.2	8.6	4.3	55.9	9.0
PNH-6	0.3 : 0.7	0.29 : 0.71	3.9	58.5	8.5	3.8	55.6	8.4

^a The calculation was based on the molar ratio in feed.

^b Molar ratio in copolymers was calculated based on analysis of experimental values.

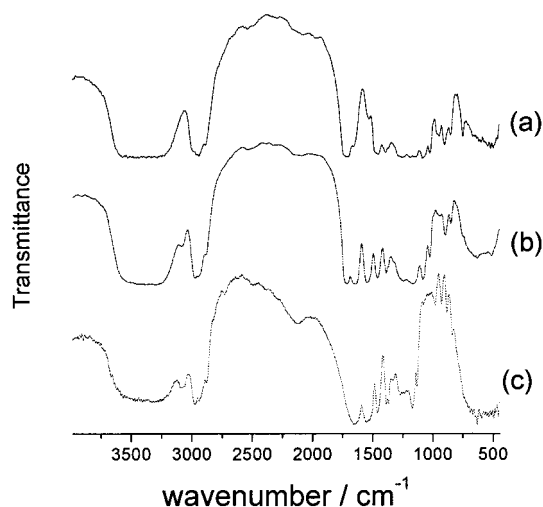


Figure 2 IR spectra of polymers and copolymer: (a) poly(HEMA); (b) poly(NIPA-co-HEMA); (c) poly(NIPA).

amount of BIS, 3 mol %; volume ratio of water to cyclohexane, 1 : 5; amount of APS 1 wt % and TEMED 2.1 wt % based on total amount of monomers.

Apparently the yield increased as the reaction time increased within the first 10 h, but no longer increased after that. It is explained that reactants have a certain solubility at a given temperature in a mixture of water and cyclohexane. According to the result we controlled the reaction time for 8 h. The synthetic conditions and results are listed in Table I.

Structural characterization

Compositions of the copolymers could be readily determined according to the nitrogen contents from the results of elemental analysis because nitrogen was one of the elements in NIPA, which distinguished it from HEMA. The relationship between nitrogen percentage (n) and molar fraction of NIPA (x) is as follows:

$$x = (134.62n - 84) / (1400 + 17n) \quad (6)$$

This formula could be used to determine the composition of a copolymer by converting nitrogen percentage into molar ratio of NIPA/HEMA, or calculating theoretical nitrogen contents according to feed compositions. It may be observed from Table II that the experimental value of nitrogen content in a copolymer was lower than that in the feed mixture for each sample, indicating that there was less NIPA component in a copolymer than in the feed mixture from which the copolymer was made. It is probably because NIPA is more soluble in cyclohexane and less reactive than HEMA in the reaction.

The structure of poly(NIPA-co-HEMA) was also confirmed by characteristic absorption peaks of the infrared spectrum. In Figure 2(b), for poly(NIPA-co-

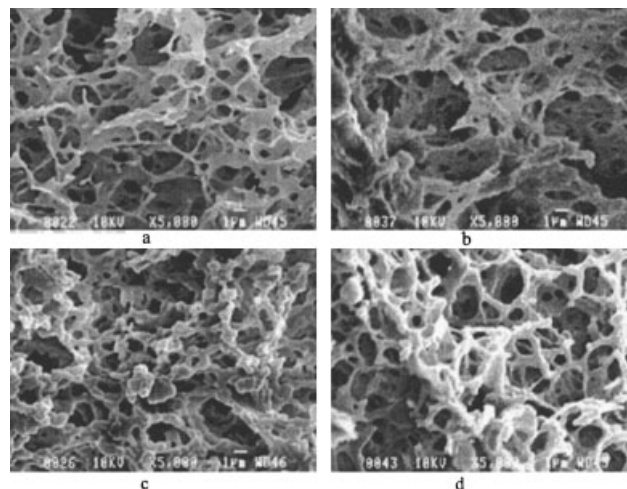


Figure 3 Scanning electron micrographs of: (a) PNH-1; (b) PNH-2; (c) PNH-3; (d) PNH-4.

HEMA) there was a weak peak appearing at 3100 cm^{-1} , which was the $\nu_{\text{C-N}}$ of the substituted amide. Compared to Figure 3(a), pure poly-HEMA did not have this absorption. The peak at 2900 cm^{-1} was the evidence of CH_2 and CH_3 stretching; 1735 cm^{-1} was attributed to C=O of the ester structure in copolymer; whereas peaks at 1650 and 1580 cm^{-1} showed the existence of C=O group in the amide structure. The peak around 1040 cm^{-1} was attributed to C-O stretching in the ester structure. Therefore all the characteristic absorption peaks suggested that two component units of NIPA and HEMA coexist in the copolymer. Figure 2 also depicts spectra of pure poly-NIPA and poly-HEMA for comparison with the copolymer containing the two components.

Certain loading of hydroxyl groups on a resin is indispensable for the resin to be further functionalized. Thereby measurements of hydroxyl loading were carried out by means of titration. A clear trend may be seen in Table III. Hydroxyl loading increased from 1.5 to 4.2 mmol/g when the molar ratio fraction of HEMA in resin varied from 0.27 to 0.71. This trend is reasonable given that HEMA provides hydroxyl groups. However, it was found that loading of titration was lower than that predicted for each sample.

TABLE III
Hydroxyl Loading in Poly(NIPA-co-HEMA)

Resin	Molar ratio of NIPA : HEMA (mol)	Loading by calculation (mmol/g)	Loading by titration (mmol/g)
PNH-1	0.73 : 0.27	2.2	1.5
PNH-2	0.66 : 0.34	2.7	2.1
PNH-3	0.63 : 0.37	3.0	2.5
PNH-4	0.50 : 0.50	3.9	3.1
PNH-5	0.34 : 0.66	5.1	3.8
PNH-6	0.29 : 0.71	5.5	4.2

TABLE IV
Particle Size Distribution of Copolymer Beads (%)^a

Sample	Particle size (μm)				
	840–601	600–421	420–251	250–150	<150
PNH-a	24.5 \pm 0.4	39.4 \pm 0.6	22.9 \pm 0.5	8.5 \pm 0.2	4.7 \pm 0.1
PNH-b	21.9 \pm 0.3	40.7 \pm 0.7	23.5 \pm 0.5	8.4 \pm 0.2	5.5 \pm 0.1
PNH-c	17.2 \pm 0.2	41.8 \pm 1.0	25.3 \pm 0.3	9.2 \pm 0.3	6.5 \pm 0.2

^a Amounts of surfactants are 0.4, 0.5, and 0.6 g for PNH-a, PNH-b, and PNH-c, respectively.

The reasons could be attributed to the fact that the resin was less swellable in pyridine, which was used as the solvent for reactions between resins with acetic anhydride before titration. The esterification was depressed inside the beads because of internal diffusion limitations. Thus the measured loading with this method was lower than the calculated contents. However, the hydroxyl contents in these beads were still substantial, a result that ensured that the copolymer beads would be a potential candidate as a support for solid-phase synthesis.

Physical characterization

Particle size distribution was determined by sieving. Particle sizes are affected by some factors such as stirring speed in reaction and amount of surfactants. We investigated the relationship between particle sizes and amount of surfactants at a constant stirring speed of 300 rpm. The reaction conditions were: molar ratio of NIPA to HEMA, 1 : 1; crosslinker, 3 mol %; volume ratio of water/oil, 1/5; surfactants were a blend of sorbitan monooleate and sorbitan monopalmitate in a weight ratio of 1 : 2. The particle size distribution is shown in Table IV. The average sizes became slightly smaller and the distribution became slightly narrower with increasing amounts of surfactants.

Pycnometry studies provided some information about porous structures. Cyclohexane can permeate

into pores of resins but does not swell them. Therefore pore structure can be detected by cyclohexane. The true density, apparent density, porosity, and pore volume were estimated and are listed in Table V. We have seen that the true density and apparent density increase with the increase of HEMA in the resins from PNH-1 to PNH-6, whereas porosity and pore volume decrease in the same sequence. This is because the inherent molecular structures of NIPA and HEMA are different. NIPA has a branched isopropyl group in its molecule, so it is less dense. When the degree of crosslinking increased from 1% in PNH-7 to 5% in PNH-10, the true density and apparent density increased but porosity and pore volume decreased. These results may be readily understood. Further investigations of the particle surface property are currently under way.

To investigate morphology structures inside resins, scanning microscope tests for some samples were conducted with a JEOL JSM 840 scanning microscope (JEOL, Peabody, MA). It was seen that macroporous network structures existed within the resins.

To investigate thermosensitivity, equilibrium swelling ratios of the resins were measured as a function of temperature, varying from 20 to 55°C. Resins with

TABLE V
Pore Structural Parameters of the Resins

Resin	ρ_t (g/mL)	ρ_a (g/mL)	P (%)	V_p (mL/g)
PNH-1	1.068	0.367	65.7	1.79
PNH-2	1.102	0.413	62.5	1.51
PNH-3	1.151	0.481	58.2	1.21
PNH-4	1.228	0.526	57.2	1.09
PNH-5	1.257	0.595	52.7	0.89
PNH-6	1.283	0.624	51.4	0.82
PNH-7	1.104	0.401	63.7	1.59
PNH-8	1.215	0.497	59.1	1.19
PNH-9	1.302	0.623	52.2	0.84
PNH-10	1.324	0.624	51.5	0.80

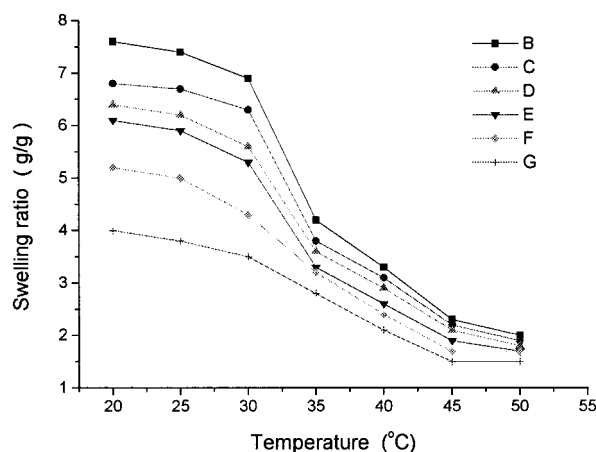


Figure 4 Dependency of swelling ratio on temperature for resins with molar ratio of NIPA/HEMA: (B) 0.73/0.27; (C) 0.66/0.34; (D) 0.63/0.37; (E) 0.55/0.45; (F) 0.34/0.66; (G) 0.29/0.71.

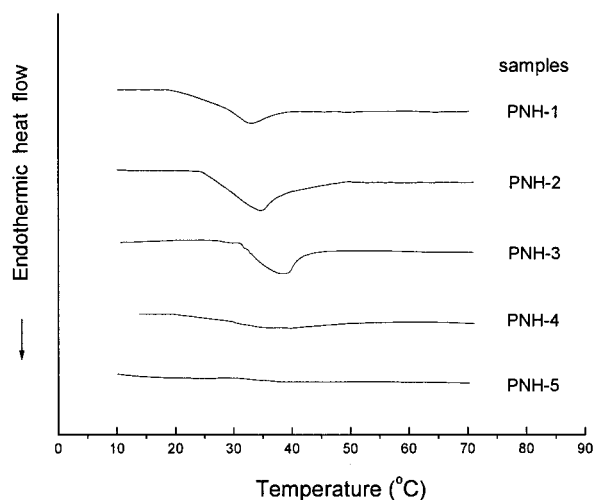


Figure 5 Differential scanning calibration thermogram of poly(NIPA-co-HEMA) at a heating rate of 10°C/min.

different molar ratios and degrees of crosslinking were tested. The results are presented in Figure 4. As shown in the figure, resins exhibited various thermosensitivities. The higher the NIPA component in a resin, the more thermosensitive the resin. If the major component in the copolymer is HEMA, the thermosensitivity will be very weak. We also observed through measurements that the swelling ratio varied with different degrees of crosslinking. The swelling ratio decreased as the degree of crosslinking increased. This phenomenon corresponds to the behavior of common hydrogels.

A differential scanning calorimeter confirmed thermosensitivity (Figure 5). Some endothermic peaks between 30 and 40°C were observed. These peaks corresponded to phase-transition temperatures of the copolymers. When the molar ratio of NIPA/HEMA was high (0.73/0.27), the phase-transition temperature was 33°C (close to that of pure poly-NIPA). As the proportion of the HEMA component increased, the peaks shifted slightly toward high temperature.

CONCLUSIONS

Beaded thermosensitive resins of poly(NIPA-co-HEMA) were synthesized by inverse suspension polymerization. In the synthesis cyclohexane was selected as the dispersion medium and a blend of sorbitan monooleate and sorbitan monopalmitate as the surfactant.

Elemental analysis showed that there was less NIPA component in a copolymer than in its relevant feed mixture. The structure of the copolymer was con-

firmed by infrared spectroscopy. Measurements of porous structures revealed that the resins had a macroporous network, a finding that was further confirmed by SEM micrographs. The swelling ratio of most resins varied as a function of temperature, which indicated the resins were thermosensitive to a certain extent. Volume phase-transition temperatures were determined using a differential scanning calorimeter. Loading of hydroxyl groups was estimated by titration, and showed a certain amount of hydroxyl loading. The conclusions drawn above suggested that thermosensitive resins might have potential applications as supports in solid-phase syntheses.

The authors are grateful for the financial support of this work from the Natural Science and Engineering Research Council (NSERC) of Canada. C. H. Ni also thanks The Educational Committee of Hubei Province and the China Scholarship Council for a visiting scholarship to Université de Montréal.

References

1. Tanaka, T. *Phys Rev Lett* 1978, 40, 820.
2. Heskins, M.; Guillet, J. E. *J Macromol Sci Chem* 1968, A2, 1441.
3. Liu, H. Y.; Zhu, X. X. *Polymer* 1999, 40, 6985.
4. Matsuo, E. S.; Tanaka, T. *J Chem Phys* 1988, 89, 1.
5. Inomata, H.; Goto, S. *Macromolecules* 1990, 23, 4887.
6. Hirotsu, S. *J Chem Phys* 1988, 88, 427.
7. Li, Y.; Tanaka, T. *J Chem Phys* 1989, 90, 5161.
8. Liu, F.; Zhuo, R. X. *Polym Mater Sci Eng* 1998, 14, 54.
9. Shin, Y.; Chang, J. H.; Liu, J.; Williford, R. *J Controlled Release* 2001, 73, 1.
10. Mi, F. L.; Wong, T. B.; Shyu, S. S.; Chang, S. F. *J Appl Polym Sci* 1999, 71, 747.
11. Allan, S. H.; Ali, A.; Dong, L. C. *J Controlled Release* 1986, 4, 213.
12. Nikolaos, A.; Peppas, D.; Ende, J. A. *J Appl Polym Sci* 1997, 66, 509.
13. Nonaka, T.; Watanabe, T.; Kawabata, T.; Kurihara, S. *J Appl Polym Sci* 2001, 79, 115.
14. Bune, Y. V.; Barabanova, A. I.; Yu, B. S.; Gromov, V. F. *Eur Polym J* 1997, 33, 1313.
15. Zhang, J.; Nicholas, A. P. *Macromolecules* 2000, 33, 102.
16. Chen, G.; Allan, S. H. *Bioconjugate Chem* 1993, 4, 509.
17. Lee, W. F.; Huang, Y. L. *J Appl Polym Sci* 2000, 77, 1769.
18. Cicek, H.; Tuncel, A. *J Polym Sci Part A: Polym Chem* 1998, 36, 527.
19. Greig, D. C.; Sherrington, J. A. *Polymer* 1978, 19, 163.
20. He, B.; Huang, W. *Ion Exchanger and Adsorption Resin*; Shanghai Scientific Technology and Education Press: Shanghai, 1992.
21. Makino, K.; Agata, H.; Ohshima, H. *J Colloid Interface Sci* 2000, 230, 128.
22. Dowding, P. J.; Vincent, B.; Williams, E. *J Colloid Interface Sci* 2000, 221, 268.
23. O'Connor, S. M.; Gehrke, S. H. *Polymer* 2001, 42, 1541.
24. Omidian, H.; Hashemi, S. A.; Sammes, P. G.; Meldrum, I. *Polymer* 1999, 40, 1753.
25. Hirokawa, Y.; Tanaka, T. *J Chem Phys* 1984, 81, 6379.
26. Abe, T.; Egawa, H.; Ito, H.; Nitta, A. *J Appl Polym Sci* 1990, 40, 1223.