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# Effect of Monomer Composition on the Properties of Biodegradable Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

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**ABSTRACT:** A series of biodegradable hydrogels were prepared from *N*-isopropylacrylamide (NIPAAm), acrylic acid (AA), and a biodegradable crosslinker, poly(caprolactone) diacrylate (PCLdA). The effect of the different molar ratios of NIPAAm and AA in the poly(NIPAAm-*co*-AA) copolymeric hydrogel on the swelling behaviors and physical properties were investigated. The results showed that the swelling ratio in deionized water and in phosphate buffer solution increased with an increase of the content of AA in the copolymeric hydrogel, and the critical gel transition temperature (CGTT) of the copolymeric hydrogel crosslinked with PCLdA was lower than the hydrogel crosslinked with *N*,*N*'-methylenebisacrylamide (NMBA). The compressive modulus decreased with an increase of AA content in the copolymeric hydrogels. The gels showed good pH/temperature sensitive behavior. The results also showed that the hydrogel crosslinked with PCLdA has higher crosslinking density ($\rho_x$) and higher mechanical strength than the hydrogel crosslinked with NMBA. The influence of the copolymeric composition on the drug release behavior in the poly(NIPAAm-*co*-AA) hydrogels was also investigated in this study. © 2012 Wiley Periodicals, Inc. J. Appl. Polym. Sci. 000: 000–000, 2012

**KEYWORDS:** biodegradable; poly(caprolactone) diacrylate; *N*-isopropyl acrylamide; drug release

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## INTRODUCTION

Hydrogels are three-dimensional hydrophilic polymers, which swell but do not dissolve when brought into contact with water, and they sometimes undergo a volume phase change in response to a change in surrounding conditions, such as temperature,[1] pH,[2] ionic strength,[3] and electric field,[4] etc. Thermosensitive hydrogel, one of the environmental stimuli response hydrogels, collapses at elevated temperature through the critical gel transition temperature (CGTT). Poly(NIPAAm) hydrogel is a well-known thermosensitive hydrogel and has a collapsed phase near its critical point between 31 and 35°C.[5] Because the permeability of water through the hydrogel can be changed by an "on-off" switch according to the environmental temperature, such materials can be used in many fields such as drug release and drug delivery system.[6–9] In addition, a small amount of anionic monomer, such as acrylic acid, is incorporated into a thermoreversible hydrogels, the CGTT of the hydrogel depends on the ionization of the pendant carboxyl groups, i.e., the pH of the medium. As the pH of the medium increases above the p$K_a$ of the carboxyl groups of polyanions, CGTT shifts to higher temperatures due to the increased hydrophilicity and charge repulsion.[9,10]

To maintain the three-dimensional structure of hydrogel, prevent from hydrophilic segment dissolving under state of aqueous solution, crosslinker was usually used. As present reports, poly(NIPAAm) hydrogels are usually formed by the covalent crosslinking of poly(NIPAAm) chains with a commercial crosslinking agent like *N*,*N*'-methylene-bis-acrylamide (NMBA). Those hydrogels are not biodegradable, which may restrict their applications as biomaterials. For the application as biomaterials or friendly environmental materials, degradability is especially emphasized on the hydrogel preparation. The bonding formed in gelation is designed to be instability (labile bond), and it can be degrade by enzyme or chemisorptions in the physiological phenomenon, most of the binding is degraded by the way of hydrolysis. Besides being able to be degraded in the organism, the little member degraded must be low toxicity, and in the supersession of the organism won't cause the injury of the kidney.[11–14] Zhang et al. prepared the PNIPAAm/dextran-allyl isocyanate hydrogel particles with modified dextran as crosslinker. The dextran is a biodegradable polysaccharide, which is susceptible to enzymatic digestion in human body.[11] París et al. reported swelling and hydrolytic degradation behavior of pH-responsive hydrogels of poly[NIPAAm-*co*-methacrylic acid] crosslinked by biodegradable poly(caprolactone) (PCL) chains.[14]

PCL, a semicrystalline linear resorbable aliphatic polyester, is subjected to biodegradation because of the susceptibility of its

aliphatic ester linkage to hydrolyze. The products generated are either metabolized via the tricarboxylic acid (TCA) cycle or eliminated by direct renal secretion. At present, PCL is regarded as a soft and hard-tissue compatible material including resorbable suture, drug delivery system, and bone graft substitutes. However, applications of PCL might be limited because degradation and resorption kinetics of PCL are considerably slower than other aliphatic polyester due to its hydrophobic character and high crystallinity. Kweon et al. synthesized PCL macromer by acrylation of acryloyl chloride to build the scaffold and drug delivery matrix.[15]

On drug release, highly swollen ionic hydrogels contain large amounts of unbound water, allowing a lot of solutes release or greater solute transport.[16] When drugs loaded in ionic hydrogels are released, the electrostatic interaction between the drug molecules and the hydrogel matrices has to be considered. For example, the release of the drug solute from the hydrogel with the different charge is affected by electrostatic attraction.[17] Because the drug release behavior of the hydrogels is related to their charge density, swelling ratio, and drug type.

To provide the degradability of poly(NIPAAm) gels, a series of biodegradable porous hydrogels based on thermosensitive NIPAAm and biodegradable crosslinker, polycaprolactone diacrylate (PCLdA), were prepared by photopolymerization at low temperature in our previous study.[18] The results showed that the poly(NIPAAm) gels crosslinked with biodegradable crosslinker-PCLdA have higher gel strength and crosslinking density than that crosslinked with NMBA, but lower the swelling ratio. To improve the hydrophilicity of poly(NIPAAm-PCLdA) hydrogels, acrylic acid (AA) was incorporated into the hydrogels, the effect of the molar ratios of NIPAAm and AA in the poly(NIPAAm-AA-PCLdA) on their swelling behavior and physical properties in deionized water and phosphate buffer solution at 25°C is the main objective of this study. In addition, the effect of different molar ratios of monomers in copolymeric gels on the drug release behavior for the drugs with different charges and molecular size was also studied.

## EXPERIMENTAL

### Materials

*N*-isopropyl acrylamide (NIPAAm) (Wako Pure Chemical, Osaka, Japan) as monomer was recrystallized in *n*-hexane before use to remove an inhibitor. Acrylic acid (AA) (Fluka, St. Gallen, Switzerland) as monomer was purified by distillation. Poly(caprolactone) diol (PCL diol, $\overline{\text{Mw}} = 2000$), (Aldrich chemical, St. Louis, MO), triethylamine (TEA), and acryloyl chloride (Fluka) were used as received. Dimethyl formamide (DMF) and benzene were dried over calcium hydride as solvents. Hexane was used as precipitant. Azobisisobutylonitrile (AIBN) as an initiator was further purified by recrystallization. Caffeine anhydrous (Fluka), vitamin $B_{12}$, crystal violet (CV), indomethacin, sulfanilamide (Sigma chemical, St. Louis, MO) and phenol red (Tokyo Kasei, Tokyo, Japan) as model drugs in the drug release experiment were used as received.

### Preparation of Poly(caprolactone) Diacrylate (PCLdA)

The synthesis and characterization of PCLdA macromer was reported in our previous report.[18] Briefly, 10 g (0.005 mol) of PCL diol ($\overline{\text{Mw}} = 2000$) was dissolved in 50 mL of benzene in a 250 mL of round-bottomed flask, then, 1.53 mL (0.015 mol) of triethylamine and 1.22 mL (0.015 mol) of acryloyl chloride were added into the flask. The reaction was carried out under stirring for 5 h at 40°C. The reaction mixture was filtered to remove triethylamine hydrochloride, then the macromer was obtained by dropping the filtrate into an excess of *n*-hexane. Finally, the precipitated PCLdA macromer was dried at 40°C vacuum oven for 24 h.

### Preparation of Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

NIPAAm and AA with various molar ratios and 2 mol % PCLdA based on total monomer content were dissolved in 10 mL of DMF. To this solution, 1 mol % AIBN as initiator was added, the mixture was immediately injected into the space between two glass plates with a 2-mm silicone rubber as a spacer. Polymerization was carried out at 75°C for 1 day. After the gelation was completed, the gel membrane was cut into disks, 10 mm in diameter, and immersed in an excess of acetone to remove the residual unreacted monomer, then immersed in deionized water for 5 days.

The copolymeric hydrogels were lyophilized for 3 days and then further dried in a 25°C vacuum oven for 1 day. The feed compositions, yields and equilibrium-swelling ratios of the copolymeric hydrogels are listed in Table I.

### Measurement of Swelling Ratio

The preweighed dried gels ($W_d$) were immersed in 10 mL of deionized water (or various volume ratios of EtOH/ $H_2O$ or phosphate buffer solution) at 25°C until swelling equilibrium was attained. Each gel was then removed from the water bath, tapped with filter paper to remove excess surface water, and weighed as the wet weight ($W_w$). The swelling ratio (SR) was calculated from eq. (1):

$$\text{SR} = \frac{W_w - W_d}{W_d} \tag{1}$$

### Measurement of Dynamic Swelling

The dried gels were immersed in an excess amount of deionized water (or phosphate buffer solution, pH = 7.4) at 25°C. The swelling ratio was obtained by the weight of the initial and swollen samples at various time intervals. The amount of water absorbed ($W_t$) was reported as a function of time and the equilibrium absorption at an infinitely long time was designated as $W_8$. Equation (2) was used to calculate the diffusion coefficient ($D$) for $W_t / W_\infty \leqq 0.8$:[19]

$$\frac{W_t}{W_\infty} = \left(\frac{4}{\pi^{0.5}}\right)\left(\frac{Dt}{L^2}\right)^{0.5} \tag{2}$$

where $t$ is the time and $L$ is the initial thickness of the dried gel.

### Measurement of Equilibrium Swelling Ratio at Various Temperatures

Two preweighed dried gels were immersed into 10 mL of deionized water for 2 days at different temperatures from 17 to 40°C.

**Table I.** Feed Compositions, Yield, and Equilibrium Swelling Ratio of the Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

| Sample code | NIPAAm (M) | AA (M) | PCLdA (mol %) | AIBN (mol %) | Yield (%) | SR (eq) (g $g^{-1}$) | *SR (eq) (g $g^{-1}$) |
|---|---|---|---|---|---|---|---|
| N100 | 1 | 0 | | | 88.0 | 0.60 | - |
| N90 | 0.95 | 0.05 | | | 86.1 | 1.79 | 2.68 |
| N85 | 0.85 | 0.15 | 2 | 1 | 86.2 | 2.24 | 3.50 |
| N75 | 0.75 | 0.25 | | | 85.4 | 2.85 | 5.52 |
| N50 | 0.5 | 0.5 | | | 85.1 | 4.16 | 9.81 |

SR (eq): the equilibrium swelling ratio in deionized water at 25°.
*SR (eq): the equilibrium swelling ratio in the phosphate buffer solution, pH = 7.4 at 25°.

After swollen equilibrium was attained, the equilibrium-swelling ratio for the gels at every temperature was calculated as eq. (1).

### Measurement of Physical Properties

The gel strength of these samples was measured by an uniaxial compression experiment with a universal tester (LLOYD LRX, Poole, UK). Equation (3) was used to calculate the shear modulus ($G$):[20,21]

$$\tau = F/A = G(\lambda - \lambda^{-2}) \quad (3)$$

where t is the compression stress, $F$ is the compression load, $A$ is the cross-sectional area of swollen gels, and $\lambda$ is the compression strain ($\Delta L/L_0$) where $\Delta L$ is the difference of the thickness of deformed gel and initial swollen gel $L_0$. At low strains, a plot of shear stress versus -($\lambda - \lambda^{-2}$) yielded a straight line, the slope of which was shear modulus ($G$). The effective crosslink density ($\rho_x$) was calculated from $G$ and the polymer volume fraction ($v_2$) as follows:

$$\rho_x = G/v_2^{1/3}RT \quad (4)$$

where $R$ is the ideal gas constant ($8.48 \times 10^4$ g cm/mol K) and $T$ is the absolute temperature.

### Measurement of Equilibrium Swelling Ratio at Various pH Buffer Solutions

A series of pH buffer solutions (citric acid/$Na_2HPO_4$) were prepared and adjusted to a constant ionic strength of 0.6$M$ through adding of NaCl. The preweighed dried gels were immersed into 10 mL of the buffer solutions to swell. The equilibrium-swelling ratio of the gels in each pH solution was calculated with eq. (1).

### Drug Release Experiment

The model drugs used in the drug-release experiment were caffeine, $B_{12}$, crystal violet (CV), phenol red, sulfanilamide, and indomethacin. The dry gels were equilibrated in 30 mg of drug/10 mL of deionized water at 25°C for 3 days to load the drugs into the gels. On the other hand, the dry gels were equilibrated in 30 mg of drug/10 mL of alcohol solution (alcohol : water = 8 : 2 as volume ratio) at 25°C for 3 days to load indomethacin. The drug-release experiments were carried out by transferring previously incubated drug gels into a 10 mL phosphate buffer solution at 37°C. The gels were repeatedly removed and transferred into a fresh 10 mL buffer solution at each fixed time interval. The released drugs were analyzed with an ultraviolet spectrophotometer (JASCO V530, Tokyo, Japan) for caffeine at 272 nm, for $B_{12}$ at 361 nm, for CV at 588 nm, for phenol red at 430 nm, for sulfanilamide at 258 nm, and for indomethacin at 265 nm, respectively.

### Biodegradability of Gels

The dried polymer discs were equilibrated in phosphate buffer solution (0.2$M$, pH 7.4) containing 0.02 wt % sodium azide to inhibit bacterial growth and 1 mg $mL^{-1}$ of lipase, then incubated at 37°C. Weight loss was monitored gravimetrically at various time intervals.

## RESULTS AND DISCUSSION

### Characterization of the Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

A series of poly(NIPAAm-AA-PCLdA) copolymeric hydrogels were prepared from various molar ratios of NIPAAm and AA with 2 mol % PCLdA based on total monomer content by free radical polymerization in DMF. Table I shows the yields and equilibrium swelling ratios ($SR_{eqs}$). The results showed that the $SR_{eq}$ for poly(NIPAAm) gel crosslinked with PCLdA (N100) in deionized water was very low (0.60 g $g^{-1}$). This low swelling ratio was unfavorable for biodegradation in environmental medium. To improve this defect, a hydrophilic monomer, AA, was selected to incorporate into poly(NIPAAm) gel. The results in Table I indicated that the $SR_{eqs}$ of the present copolymeric hydrogels in deionized water and buffer solution increase with an increase of AA content in the copolymeric gel due to the stronger hydration ability of the carboxyl group in AA.

### Swelling Kinetics of the Copolymeric Hydrogels in Deionized Water and Phosphate Buffer Solution

The swelling kinetic profiles of the present copolymeric hydrogels in deionized water and phosphate buffer solution (pH = 7.4) were shown in Figures 1 and 2, respectively. The $SR_{eqs}$ for the present copolymeric hydrogels in deionized water in Figure 1 indicated that the more the AA content in the gels, the higher the swelling ratio of the gels, that is, N50 (4.16 g $g^{-1}$) > N75 > N85 > N90 > N100 (0.60 g $g^{-1}$). At the same time, the $SR_{eqs}$ for the present copolymeric hydrogels in phosphate buffer solution are higher than those corresponding gels in deionized water solution. This is because carboxylic group of AA is neutralized by the sodium ion ($Na^+$) to form sodium carboxylate (—$COO^-Na^+$). The charge repulsion of the polymeric chain results in the gel network loose and can accommodate much more water. Similar results were observed from our previous report in the poly(NIPAAm-*co*-AA-*co*-Sodium acrylate) gels in which crosslinker used was NMBA.[22]

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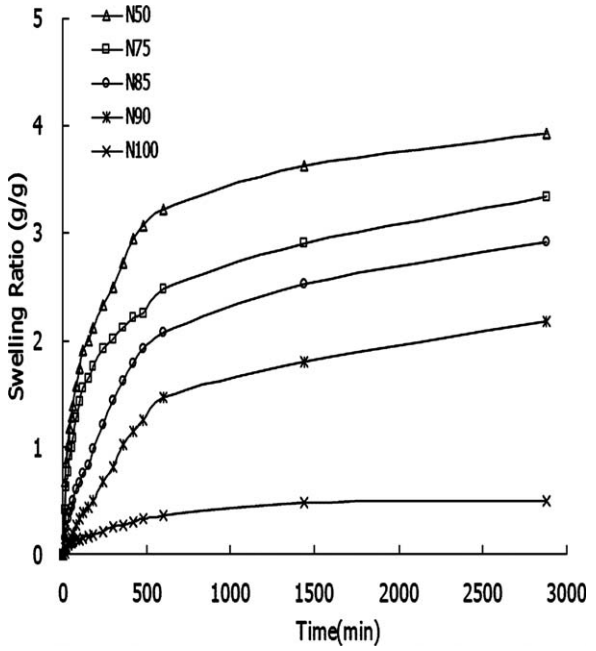


**Figure 1.** Swelling kinetic profile of different compositions of the poly(-NIPAAm-AA-PCLdA) copolymeric hydrogels in deionized water at 25°C.

To elucidate the transport mechanism of the present copolymeric hydrogels in deionized water and in phosphate buffer solution, the initial swelling data were fitted to eq. (2). The $D$ values shown in Table II indicate that the diffusion coefficients in the said copolymeric gels are higher than that of N100 hydrogel and increase with an increase of the content of AA and range from $1.13 \times 10^{-8}$ cm$^2$ s$^{-1}$ to $3.8 \times 10^{-8}$cm$^2$ s$^{-1}$ and $16.6 \times 10^{-8}$ cm$^2$ s$^{-1}$ to $21.2 \times 10^{-8}$ cm$^2$ s$^{-1}$ in deionized water and in phosphate buffer solution, respectively. These results show that the $D$ values for the copolymeric gels in phosphate buffer solution are higher one order than those in deionized water. This result can be confirmed from the initial swelling rate in swelling medium. Because, the initial swelling rate shown in Figures 1 and 2 from the dried state at 25°C increases with an increase of the content of AA in the copolymeric hydrogels and the initial swelling rates in phosphate buffer solution (0.457 g min$^{-1}$ for N50) are higher than those corresponding gels in deionized water (0.0143 g min$^{-1}$ for N50).

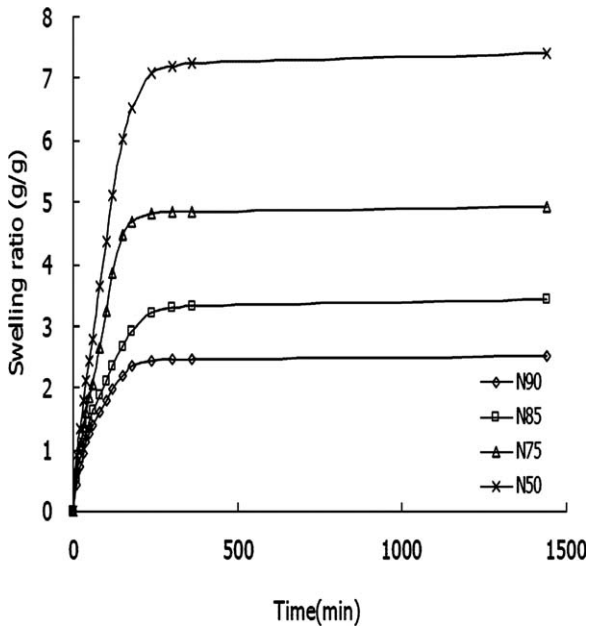


**Figure 2.** Swelling kinetic profile of different compositions of the poly (NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution (pH = 7.4) at 25°C.

**Table II.** Diffusion Coefficient, Gel Strength, and Crosslinking Density of the Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

| Sample code | $G \times 10^{-2}$ (g cm$^{-2}$) | $\rho \times 10^{3}$ (mol cm$^{-3}$) | $D \times 10^{8}$ cm$^2$ s$^{-1}$ |
|---|---|---|---|
| N100[a] | 5.89 ± 1.2 | 0.07 ± 0.01 | - |
| N100 | 412.08 ± 35 | 1.67 ± 0.03 | 0.24 |
| N90 | 389.72 ± 29 | 1.58 ± 0.02 | 1.13 |
| N85 | 351.01 ± 25 | 1.44 ± 0.02 | 2.41 |
| N75 | 319.57 ± 18 | 1.37 ± 0.03 | 2.99 |
| N50 | 292.51 ± 13 | 1.34 ± 0.04 | 3.80 |
| N90[b] | 151.06 ± 26 | 0.81 ± 0.01 | 16.6 |
| N85[b] | 101.83 ± 15 | 0.67 ± 0.01 | 17.3 |
| N75[b] | 77.05 ± 15 | 0.63 ± 0.02 | 20.8 |
| N50[b] | 68.66 ± 15 | 0.60 ± 0.01 | 21.2 |

[a]The gel crosslinked with NMBA.
[b]The gel swollen in PBS buffer solution (pH = 7.4) until swelling equilibrium was attained.

### Mechanical Properties of the Copolymeric Hydrogels

The gel strength was evaluated by the shear modulus ($G$) obtained from eq. (3). According to eq. (4), an increase in the swelling ratio is usually accompanied by a decrease in $G$ and a decrease in $\rho_x$ for hydrogels. The results listed in Table II indicate that the $G$ value for the poly(NIPAAm) gel crosslinked with NMBA (N100*) is $5.89 \times 10^2$ g cm$^{-2}$, much lower than that ($412.08 \times 10^2$ g cm$^{-2}$) for poly(NIPAAm) gel crosslinked with PCLdA (N100). This is because the gel crosslinked with a hydrophobic and crystalline PCLdA showed a lower swelling ratio than that crosslinked with NMBA. Table II also shows that the $G$ and $\rho_x$ values of the copolymeric gels swelling in deionized water and in phosphate buffer solution decreased with an increase of the content of AA. At the same time, the $G$ and $\rho_x$ values for the copolymeric hydrogels in deionized water were higher than those hydrogels in phosphate buffer solution. This is due to the higher swelling ratio of the hydrogels in phosphate buffer solution. Hence, the copolymeric hydrogels crosslinked with PCLdA can enhance the gel strength and offer biodegradability.

### Effect of Temperature on Swelling Ratio

It is well known that the critical gel transition temperature (CGTT) of the poly(NIPAAm) hydrogels is around 32°C. For NIPAAm gel, the hydrophilic group (amido, —NHCO—) in the polymer structure would form an intermolecular hydrogen bond with surrounding water at low temperature (below gel transition temperature). Hence, water penetrated into the

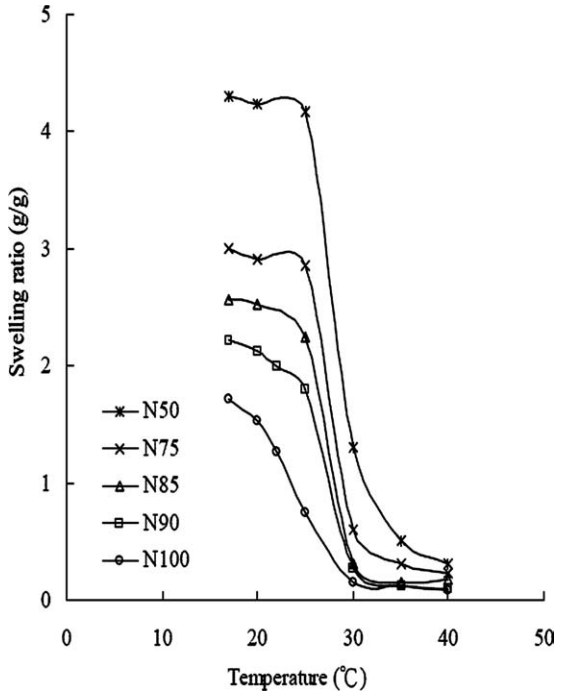


**Figure 3.** Swelling ratio as a function of temperature for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in deionized water.

NIPAAm gels is in a bound state at low temperature. The effect of temperature on the $SR_{eqs}$ for the present copolymeric gels shown in Figure 3 indicates that the swelling ratio decreases with an increase in temperature. The CGTTs for the copolymeric gels shift to higher temperatures due to the increased hydrophilicity and charge repulsion after adding AA into poly(NIPAAm) gel.[22] But, the results also show that the deswelling rate dramatically increases as the more AA was incorporated into the gels under around CGTT. This behavior conforms to our previous report for the poly(NIPAAm-AA-sodium acrylate) gels.[22]

### Effect of pH on Swelling Ratio

The effect of pH on $SR_{eqs}$ for the present copolymeric hydrogels shown in Figure 4 indicates that the swelling ratio increases with an increase in pH. However, the gel transition occurring at around pH = 5 ($pK_a$ = 4.75 for AA) is not obviously affected by the addition of more content of AA in the gel. Although the gel transition of the copolymeric hydrogel does not change, the gels still possess excellent pH response under higher AA content. The results shown in this figure also indicate that the swelling ratio of the copolymeric gels is almost the same and very small at lower pH (pH = 3), but the swelling ratio increased with an increase in pH. At higher pH (pH = 8) the swelling ratio rapidly increases with an increase in more AA content. This is attributed that when the pH of the medium increases above the $pK_a$ of the carboxyl groups of polyanions, the carboxylic acid groups are easily ionized at pH = 8 and their charge repulsion increase.

According to these results, we can control the gel deswell in low pH like in stomach to protect the drug, and swell in high pH like in intestine to release the drug. Hence, we can use this result to develop a smart drug delivery system to biomedicine.

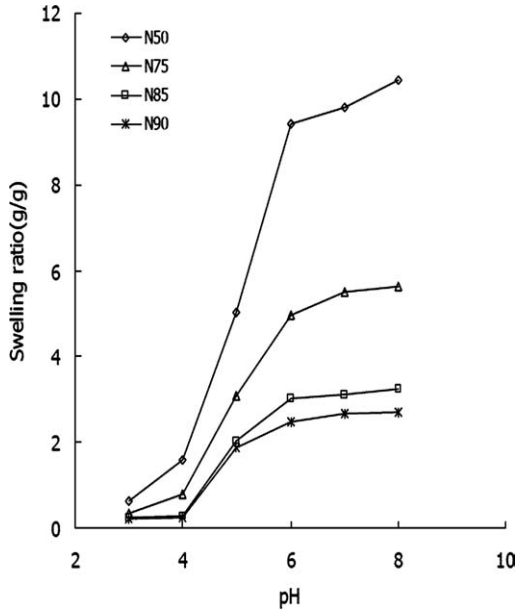


**Figure 4.** pH dependent swelling ratio of different compositions of the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in various buffer solutions (citric acid/$Na_2HPO_4$) at 25°C.

### Effect of Alcohol Aqueous Solution on Swelling Ratio

Figure 5 shows the effect of AA content on swelling ratio for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in various alcohol aqueous solutions at 25°C. The result in Figure 5 shows that the more the content of AA in the hydrogels, the higher the

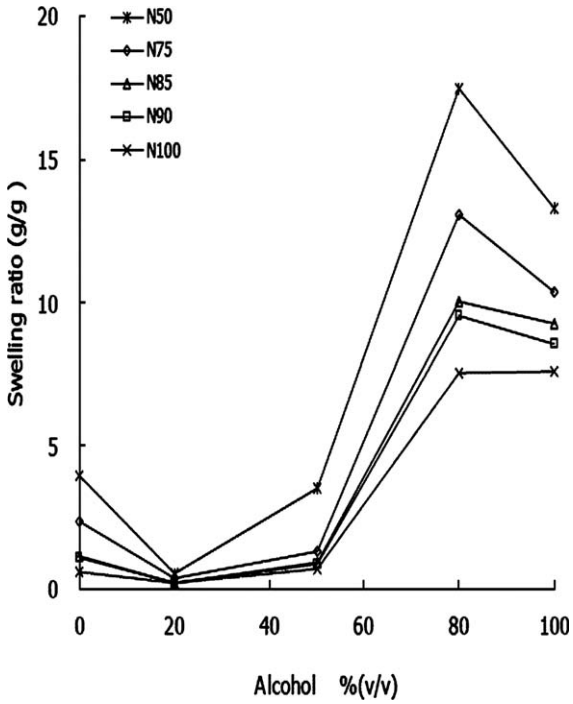


**Figure 5.** Effect of AA content on swelling ratio in various alcohol aqueous solutions with different concentrations for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels at 25°C.

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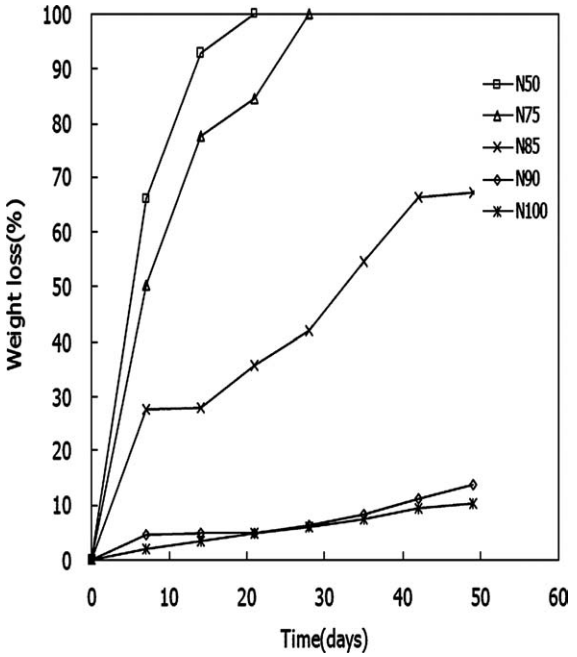


**Figure 6.** Degradation curves of the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution with lipase (1 mg $mL^{-1}$) (pH 7.4 and 37°C).

swelling ratio of the hydrogels in alcohol aqueous solutions and the swelling ratios of the hydrogels increase with an increase in the concentration of alcohol [80% alcohol >50% alcohol >20% alcohol]. In 80% alcohol aqueous solution, the hydrogels have the highest solvent content; but in 20% alcohol aqueous solution, the hydrogels have the lowest solvent content. We conjectured that when the gels swollen in 80% alcohol aqueous solution, the polarity of solvent is closed to the gel and the gel has a best affinity with solvent at this moment. But in 20% alcohol aqueous, the gel has lowest solvent content due to the large interaction force among the solvent molecules. According to this result, we dissolved indomethacin in 80% alcohol aqueous solution to let the gel have the highest loading amount of indomethacin.

**Biodegradability**

Generally, degradation kinetics of macromolecule was affected by their chemical structure and structural characteristics. PCL has great hydrophobic and crystalline properties; it does not allow fast water penetration into the PCL bulk. Though the mechanism of PCL degradation was known as random hydrolytic chain scission of the ester linkage,[23] the rate of degradation was relatively lower than other degradable polyester including poly(lactic acid) and its copolymer due to the chemical and structural characteristics of PCL segment. In our previous reports, poly(NIPAAm-PCLdA) copolymeric hydrogels[18] and PEG/PCL random block copolymer[24] showed the weight loss percentage only 15 and 28 wt % under PBS buffer solution (pH = 7.4) with lipase 1 mg $mL^{-1}$ at 37°C for 49 days. But, in the present poly(NIPAAm-AA-PCLdA) copolymeric hydrogels, Figure 6 shows the weight loss percentage of the present hydrogels against incubation time in PBS buffer solution (pH = 7.4) with lipase 1 mg $mL^{-1}$ at 37°C. The results show that the weight loss percentage approaches 100% (that is, completely degradation) at 21 and 28 days for N50 and N75 hydrogels, respectively. For N85 hydrogel, the weight loss also can approach 65 wt % for 49 days. Comparing these systems, the present poly(NIPAAm-AA-PCLdA) copolymeric hydrogels showed faster degradation behavior than poly(NIPAAm-PCLdA) copolymeric hydrogels[18] and PEG/PCL random block copolymer[24] due to the incorporation of acrylate groups into PCL.

The reason is that the AA component was added into the poly(-NIPAAm-PCLdA) copolymeric hydrogels, the swelling ratio are increased from 0.60 (g $g^{-1}$) for N100 hydrogel to 4.16 (g $g^{-1}$) and 9.81(g $g^{-1}$) for N50 hydrogel in deionized water and in phosphate buffer solution (pH = 7.4), respectively (shown in Table I). This made the hydrogel become more porous structure and let the water be easier penetration into the hydrogel network such as the $D$ value increased from $0.24 \times 10^{-8}$ ($cm^2$ $s^{-1}$) for N100 to $21.2 \times 10^{-8}$ ($cm^2$ $s^{-1}$) for N50 in phosphate buffer solution (pH = 7.4) (also see Table II). These results showed that the main influence on degradation rate is the incorporation of hydrophilic AA into the hydrogels.

**Table III.** Drug-Loading Amount and Fractional Release of the Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogels

| Drug | | N100 | N90 | N85 | N75 | N50 |
|---|---|---|---|---|---|---|
| Caffeine | Loading amount (ppm $g^{-1}$) | 251.12 | 277.23 | 292.84 | 318.44 | 354.27 |
| | Fractional release at 1440 min (%) | 21.9 | 58.6 | 66.5 | 76.7 | 88.2 |
| B12 | Loading amount (ppm $g^{-1}$) | 224.34 | 258.4 | 267.14 | 278.66 | 309.52 |
| | Fractional release at 1440 min (%) | 17.6 | 27.9 | 33.0 | 53.6 | 71.8 |
| CV | Loading amount (ppm $g^{-1}$) | - | 2961.5 | 3010.1 | 4558.6 | 4857.1 |
| | Fractional release at 1440 min (%) | - | 8.8 | 9.4 | 10.7 | 17.7 |
| Phenol Red | Loading amount (ppm $g^{-1}$) | - | 95.42 | 174.76 | 334.38 | 378.79 |
| | Fractional release at 1440 min (%) | - | 62.4 | 31.9 | 14.8 | 10 |
| Sulfanilamide | Loading amount (ppm $g^{-1}$) | 402.60 | 364.24 | 340.14 | 328.00 | 247.79 |
| | Fractional release at 1440 min (%) | 23.9 | 64 | 73 | 78.9 | 85.4 |
| Indomethacin | Loading amount (ppm $g^{-1}$) | 828.95 | 932.20 | 934.55 | 1030.3 | 1068.7 |
| | Fractional release at 1440 min (%) | 24.3 | 45.6 | 43.3 | 48.6 | 53.0 |

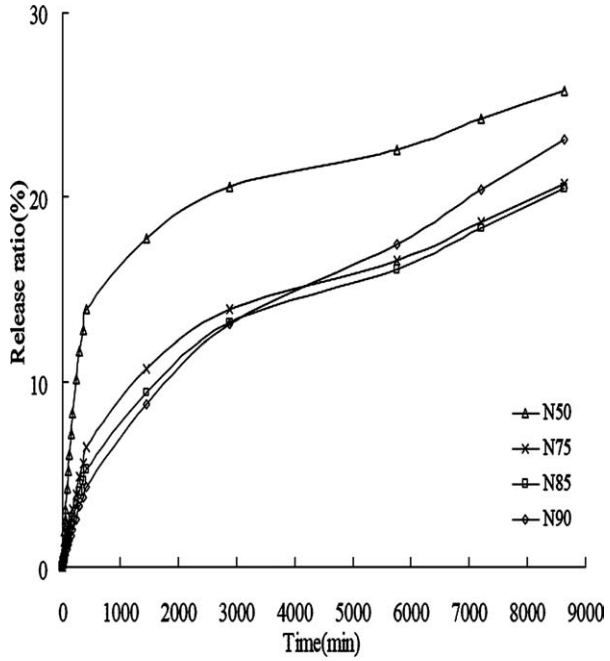


Figure 7. CV release profile under loading at 25°C and releasing at 37°C for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution (pH = 7.4).

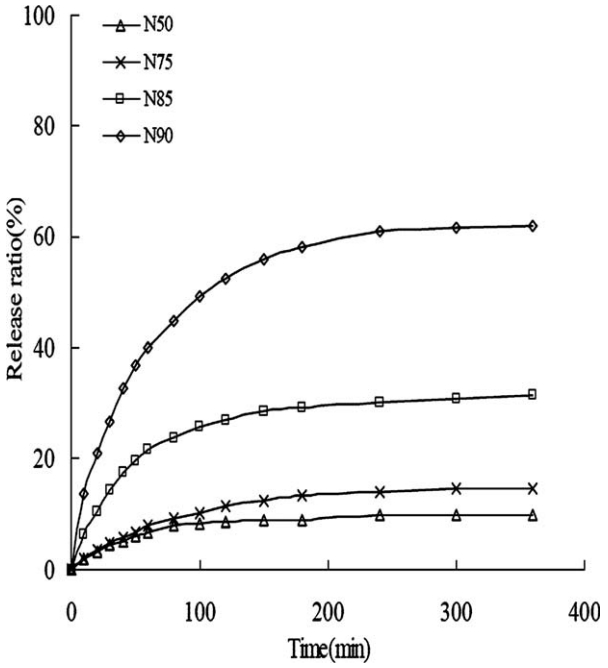


Figure 8. Phenol red release profile under loading at 25°C and releasing at 37°C for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution (pH = 7.4).

### Drug Release Behavior of Poly(NIPAAm-AA-PCLdA) Copolymeric Hydrogel

Some mechanisms of drug release are considered such as swelling, diffusion, degradation, or simultaneous diffusion/degradation controlled mechanism etc., depending on composition of the hydrogels, but effect of degradation-controlled mechanism of the copolymeric hydrogels on drug release can be negligible according to aforementioned results due to their very low degradation in 1 day. Hence, we can only consider the swelling and diffusion controlled effect in this study. The main effect of molecular size, charge, and hydrophilicity or hydrophobicity of drug on the drug release behavior in the present copolymeric gels under phosphate buffer solution (0.2 M and pH 7.4) at 37°C was investigated. The loading amount and fractional release of the model drugs for the hydrogels at 24 h are listed in Table III.

The results shown Table III indicate that the loading amount and fractional release of caffeine and vitamin $B_{12}$ increase with an increase of AA content in the copolymeric hydrogels. This can be attributed to the swelling-diffusion controlled effect of the hydrogels. The loading amount and fractional release of caffeine in the copolymeric hydrogels are higher than those of vitamin $B_{12}$. This is due to the molecular size of caffeine is smaller than that of vitamin $B_{12}$.

The copolymeric hydrogel containing AA content made the net charges of the hydrogels become negative. Therefore, the more the content of AA in the copolymeric hydrogel, the higher the anionic charge density of the gels. Hence, the loading amounts of cationic CV in the gels shown in Table III rapidly increase with an increase in AA content due to charge attraction. The fractional release profiles of CV for the copolymeric gels in phosphate buffer solution are shown in Figure 7. The release profile is related to the swelling ratio of the copolymeric gels in buffer solution. When the content of AA in the copolymeric gel increases, the swelling ratio of the gel increases, hence the

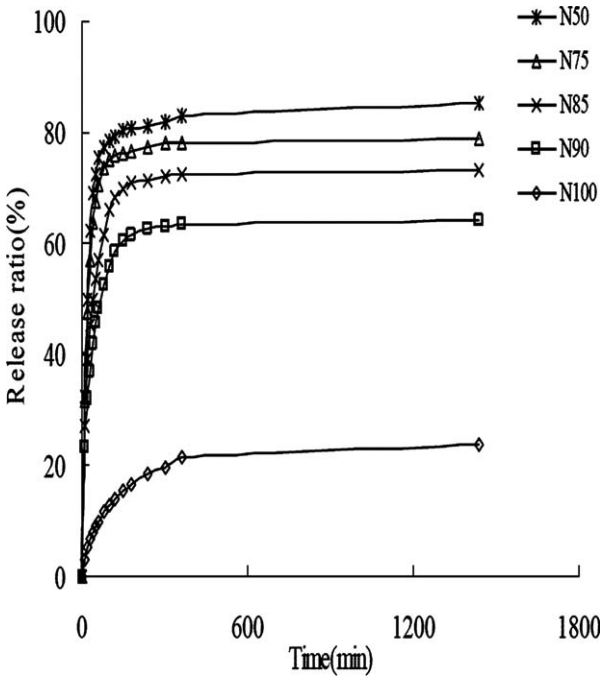


Figure 9. Sulfanilamide release profile under loading at 25°C and releasing at 37°C for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution (pH = 7.4).

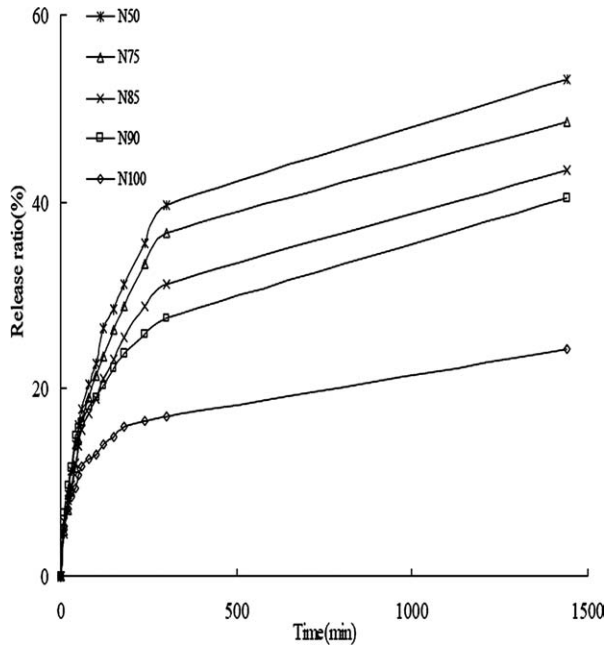


**Figure 10.** Indomethacin release profile under loading in 80% alcohol aqueous solutions at 25°C and releasing at 37°C for the poly(NIPAAm-AA-PCLdA) copolymeric hydrogels in PBS buffer solution (pH = 7.4).

fractional CV release increases, but the fractional CV release is very low due to the negative and positive charge attraction of hydrogel and cationic drug. On the contrary, the loading amounts of anionic phenol red in the hydrogels increase with increase in AA content due to increased charge repulsion with AA content in deionized water. In addition, because the charge repulsion occurs between anionic gel and anionic drug, the loading amount is lower than CV. This could be observed from loading amount of the hydrogels (see Table III). The release profile of anionic phenol red in the copolymeric hydrogel is shown in Figure 8. The results showed that with an increase of the content of AA, the fractional release of phenol red rapidly decreased, this is because the incubated-gels loading in deionized water would be reswollen in the PBS buffer solution (pH 7.4). The reason is that the carboxylic acid (COOH) group would be neutralized into sodium carboxylate group ($COO^-Na^+$) in pH 7.4 buffer solution. This condition caused the anionic drug inside the hydrogels to be more difficultly released, hence, the fractional release of phenol red in the gels decreased with an increase in AA content. This also can be observed from fractional release amount of phenol red in the gels (see Table III). This result is contrary to our previous study for the charge effects on drug release behavior for ionic thermosensitive hydrogels in deionized water.[17]

The release profiles of nonionic sulfanilamide and indomethacin from the hydrogels with different content of AA in phosphate solution were shown in Figures 9 and 10. Sulfanilamide is an uncharged small molecular drug, and its solubility in water is very small, so the bonding force between sulfanilamide and hydrogels is small. The loading amount of sulfanilamide decreased with an increase of the content of hydrophilic AA, because of the gel has good affinity toward water than drug. This could be observed from the loading amount of sulfanilamide in the hydrogels (see Table III). The release ratio is still related to the swelling ratio of hydrogels in buffer solution. In addition, indomethacin is also difficult to dissolve in water, but in alcohol. The dry gels were equilibrated in drug of 80% alcohol aqueous solution for loading indomethacin but the gels were immersed in phosphate buffer solution. Hence, the loading amount was related to the affinity of the gel toward alcohol. But the fractional release was affected by the swelling ratio of the hydrogel. Because the hydrogel and indomethacin had a good affinity toward alcohol but indomethacin has a poor solubility in buffer solution.

Consequently, the loading amount of indomethacin in the gels increased with an increase in AA but the fractional release doesn't very high. We also know that the more the content of AA in the gels, the higher the swelling ratio of the hydrogel in buffer solution. Thus, the fractional release amount of indomethacin in the copolymeric gels still increased with an increase in AA content.

## CONCLUSIONS

The hydrogels poly(NIPAAm-PCLdA) prepared with NIPAAm and polyester-PCLdA have higher mechanical strength than those crosslinked by NMBA, but the PCL segment hydrophobic group in the poly(NIPAAm) gel made the water hard to diffuse into the network of the hydrogels. An addition of hydrophilic AA into poly(NIPAAm-PCLdA) hydrogels made the hydrogels have more water content, pH sensitivity, lower crystallinity, and better biodegradability and high mechanical properties. In drug release behavior, the loading amount and release ratio of caffeine and $B_{12}$ were affected by the swelling ratio of the gel and molecular size of the drug. The loading amount and the release ratio of CV increased with an increase of the content of AA in the hydrogel, but the release ratio of phenol red in the hydrogels is contrary to CV. The loading amount of sulfanilamide decreased with an increase of the content of AA in the hydrogel, but the release ratio was affected by the swelling ratio of the hydrogel in phosphate buffer solution. Results showed that the loading amount and release ratio of indomethacin were affected by the affinity in the alcohol solution and the swelling ratio in phosphate buffer solution, respectively.

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## REFERENCES

1. Hoffman, A. S. *J. Control. Release* **1987,** *6*, 297.
2. Hirokawa, Y.; Tanaka, T. *J. Phys. Chem.* **1984,** *81*, 6379.
3. Ricka, J.; Tanaka, T. *Macromolecules* **1984,** *17*, 2916.
4. Kwon, I. C.; Bae, Y. H.; Okano, T.; Kim, S. W. *J. Control. Release* **1991,** *17*, 149.
5. Heskins, M.; Guillet, J. E. *J. Macromol. Sci. Chem.* **1968,** *1*, 1441.
6. Dong, L. C.; Hoffman, A. S. *J. Control. Release* **1991,** *15*, 141.

7. Yoshida, R.; Sakai, K.; Okano, T.; Sakurai, Y. *Polym. J.* **1991,** *23*, 1111.
8. Okuyama, Y.; Yoshida, R.; Sakai, K.; Okano, T.; Sakurai, Y. *J. Biomater. Sci. Polym. Ed.* **1993,** *4*, 545.
9. Bea, Y. H.; Okano, T.; Kim, S. W. *Makromol. Chem. Rapid. Commun.* **1987,** *8*, 481.
10. Park, H.; Robinson, J. R. *Pharm. Res.* **1987,** *4*, 457.
11. Zhang, X. Z.; Sun, G. M.; Chu, C. C. *Eur. Polym. J.* **2004,** *40*, 2251.
12. Kim, S.; Healy, K. E. *Biomacromolecules* **2003,** *4*, 1214.
13. Namkung, S.; Chu, C. C. *J. Biometry Sci. Polym. Ed.* **2007,** *18*, 901.
14. París, R.; Quijada-Garrido, I. *Polym. Int.* **2009,** *58*, 362.
15. Kweon, H. Y.; Yoo, M. K.; Park, I. K.; Kim, T. H.; Lee, H. C.; Lee, H. S.; Oh, J. S.; Akaike, T.; Cho, C. S. *Biomaterials* **2003,** *24*, 801.
16. Am Ende, M. T.; Hariharan, D.; Peppas, N. A. *React. Polym.* **1995,** *25*, 127.
17. Lee, W. F.; Chiu, R. J. *Mater. Sci. Eng. C* **2002,** *20*, 161.
18. Lee, W. F.; Cheng, T. S. *J. Appl. Polym. Sci.* **2008,** *109*, 1982.
19. Kabra, G.; Gehrke, S. H.; Hwang, S. T. *J. Appl. Polym. Sci.* **1991,** *42*, 2409.
20. Peppas, N. A.; Barr-Howell, B. D. Hydrogels in Medicine and Pharmacy; CRC Press: Boca Raton, **1986.**
21. Treloar, L. R. G. The Physics of Rubber Elasticity; Clarendon Press: Oxford, **1975.**
22. Lee, W. F.; Shieh, C. H. *J. Appl. Polym. Sci.* **1999,** *73*, 1955.
23. Pitt, C. G.; Gratzei, M. M.; Kimmei, G. L.; Surles, J.; Schindler, A. *Biomaterials* **1981,** *2*, 215.
24. Lee, W. F.; Cheng, T. S. *J. Biomater. Sci. Polym. Ed.* **2009,** *20*, 2023.