Fast Responsive Thermosensitive Hydrogels as Drug Delivery Systems

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ABSTRACT

Ionic and non-ionic temperature sensitive hydrogels of poly(*N*-isopropylacrylamide) and calcium alginate were prepared as interpenetrating networks. Calcium alginate was used as a mould to prepare uniform, large size, spherical beads and then extracted to leave a macroporous structure. Swelling behaviour, and drug release kinetics of these thermosensitive hydrogels were studied in aqueous media. The swelling observations indicated that the equilibrium swelling degree of homopolymer gel increased after calcium alginate extraction, and besides calcium alginate had no effect on lower critical solution temperature (LCST). In addition, equilibrium swelling degree of copolymer composite hydrogels containing Na^+, and Ca^{2+} cations were greater than that of the extracted hydrogel containing only monovalent cations. It was observed that, swelling kinetics of hydrogels followed a Fickian behaviour. Drug release experiments indicated that drug release from these hydrogels was fast and in Fickian manner.

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Key Words:
hydrogels;
N-isopropylacrylamide;
fast responsive;
drug delivery;
calcium alginate.

INTRODUCTION

During the past recent years most of the pharmaceutical researches have been concentrated on the discovery or synthesis of the novel drugs and drug administration systems. In this way, controlled release drug delivery systems have an outstanding place. Although long acting drug delivery systems are more desirable, but for some applications fast responsive delivery systems are also required, such as wound treatment (burst release followed by a diminishing need for drug), encapsulated flavors,

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targeted delivery (triggered burst release) and pulsatile release [1].

Among various kinds of polymeric systems, which have been used as drug containers or release rate controlling barriers, are hydrogels. Hydrogels are cross - linked, three-dimensional hydrophilic networks that swell but not dissolve when brought into contact with water. Hydrogels sometimes undergo a volume change in response to a change in surrounding conditions, such as pH [2,3], temperature [4,5], ionic strength [6,7], and electric field [8,9]. Stimuli responsive hydrogels, especially those sensitive to temperature and pH are attractive, because these factors are variables that change in typical physiological, biological, and chemical systems.

The thermosensitive hydrogel demonstrated a volume transition and associated phase transition from low temperature (a highly swollen gel), to high temperature (a collapsed gel near its critical point) [10,11].

During the recent years, *N*-substituted acrylamide homopolymers, and in particular poly(*N*-isopropylacrylamide) [poly(IPAAm)] gel have been studied by many researchers [12-18]. Poly(IPAAm) is one of the best thermotropic polymers and exhibits a LCST behaviour, collapsing and shrinking above the LCST. Molecular transition of poly(IPAAm) structure from hydrophilic to hydrophobic state occurs at LCST. Experimental values of LCST lay between 30 to 35°C and exact temperature is a function of molecular microstructure of gel. Common method in synthesizing poly(IPAAm) is inverse suspension polymerization, which yields small beads with broad size distribution. Hoffman and Park [19], in order to prepare large beads with narrow size distribution, used calcium alginate as a mold for polymerization of (IPAAm). Clinical applications of thermosensitive hydrogels based on poly(IPAAm) and its derivatives have limitations. The monomers and cross-linkers used in the synthesis of hydrogels are not known to be biocompatible, i.e. they may be toxic, carcinogenic or teratogenic. In addition, the polymers of poly(IPAAm) and its derivatives are not biodegradable. The observation that acrylamide-based polymers activate platelets upon contact with blood, together with the unclear metabolism of poly (IPAAm), requires extensive toxicity studies before clinical applications can emerge [20].

In this study, thermosensitive hydrogels of poly(*N*-isopropylacrylamide) and poly(*N*-isopropylacrylamide-*co*-sodium acrylate) were prepared as interpenetrating polymer networks (IPNs) with calcium alginate gel. Calcium alginate was used as a mold to prepare uniform, large size, spherical beads and then extracted to leave a macroporous structure. Sodium acrylate was used for evaluating the effects of the presence of the ionic species on the swelling and drug release behaviour. The aim of this research was preparation of the uniform, large size, spherical beads of thermoresponsive hydrogels as fast responsive drug delivery systems. The main advantage of the synthesized porous drug carriers is their suitability as fast responsive systems for certain applications, such as wound treatment, encapsulated flavors, targeted delivery and pulsatile release.

EXPERIMENTAL

Materials and Methods

N-Isopropylacrylamide (IPAAm) and sodium acrylate (SA) were purchased from Ubichem Ltd. (England), *N,N,Ní,Ní*-methylene-bisacrylamide (MBAAm), ammonium persulphate (APS), sodium metabisulphite (SMB) and calcium chloride from Aldrich Chemical Co. (England), tetramethylethylenediamine (TEMED) from Pharmacia LKB (Sweden), phosphate buffer (PBS 9887 Titrisol, pH 7) and ethylenediaminetetracetic acid (EDTA) from Merck AG (Germany). Sodium alginate was obtained from internal market. IPAAm, SA, and MBAAm were used without any further purification. Dilthiazem hydrochloride was prepared from Daropakhsh Co. (Iran).

Hydrogel Synthesis

As shown in Table 1, two solutions of reactants were prepared and then bubbled with pure nitrogen gas for 10 min. For ease of presentation, they are called monomer and initiator solutions. Accelerator (TEMED) and initiator (APS) were dissolved in monomer and initiator solutions, respectively, because bulk addition of reactant agent to initiator solution increases the rate of generation of free radicals and causes premature consumption of initiator [21]. In all experiments molar ratio of monomer (or monomer and comonomer) to cross - linking agent was equal to 30. In

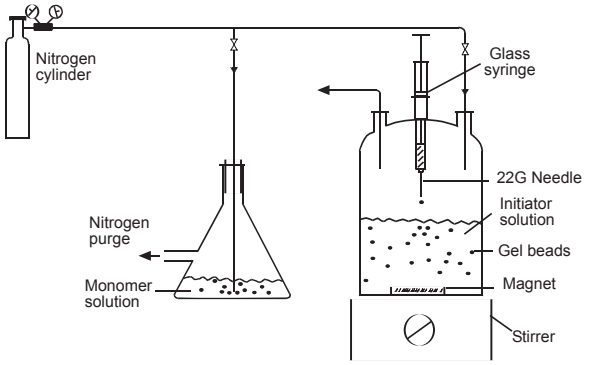

Figure 1. Apparatus for the synthesis of uniform, large size, spherical hydrogel beads.

experiments No. 3 and 4, molar ratios of monomer to comonomer were 95:5 and 90:10, respectively. Drops of monomer solution were introduced manually into the initiator solution by a glass syringe with a 22G stainless steel needle (Figure 1). The initiator solution was being stirred slowly under nitrogen atmosphere at room temperature (approximately 20°C). Small gel beads were being formed instantaneously after addition of each drop into the solution. Diffusion of calcium chloride and APS salts into the gel phase resulted in formation of poly(IPAAm) gel [or poly(IPAAm-*co*-SA)] and calcium alginate gel as IPN. After preparation of many beads in this way, dropping was stopped and reaction was continued for 8 h by stirring the solution under nitrogen atmosphere at room temperature. At the end of reaction time, gel beads were separated from solution by a nylon cloth filter and washed with distilled water. In order to allow the unreacted monomers, cross - linker and other impurities to leach out, the prepared beads were immersed in distilled water at room temperature for 48 h, and the water was refreshed every several hours. A portion of gel beads was poured into a solution of 0.1 M EDTA and phosphate buffer solution (PBS) to extract calcium alginate gel by calcium chelating in 48 h. The rest of hydrogel beads were stored for future experiments. Calcium alginate extracted gels were dried up under vacuum for 24 h and stored in desiccator.

Equilibrium Swelling

These experiments were conducted in a temperature range of 15 to 40°C for all synthesized gels. A weighed mass of dry samples was placed in PBS (pH 7) solution in a shaker bath at a fixed temperature. It was found that after about 2 h, the swollen gel beads reached a constant weight so that their equilibrium swelling ratio could be obtained by the following equation:

$$\text{Equilibrium swelling ratio} = \frac{W_{seq} - W_d}{W_d} \quad (1)$$

where, W_d and W_{seq} are the weights of dry and swollen gels at equilibrium state, respectively.

Table 1. Chemical composition of reactants for synthesis of poly (IPAAm) and poly (IPAAm-*co*-SA) gels.

Substances	Test 1	Test 2	Test 3	Test 4
Monomer solution				
IPAAm (g)	10.0	10.0	10.0	10.0
SA (g)	___	___	0.40	0.80
MBAAm (g)	0.45	0.45	0.45	0.45
Sodium alginate (g)	0.50	0.30	0.50	0.50
TEMED (mL)	0.40	0.40	0.40	0.40
Distilled water (mL)	100	100	100	100
Initiator solution				
APS (g)	1.50	1.50	1.50	1.50
Calcium chloride (g)	7.50	7.50	7.50	7.50
Distilled water (mL)	250	250	250	250

Solvent Uptake Kinetics

A weighed mass of dry sample was placed in phosphate buffer solution (PBS), (pH 7) in a shaker bath with a constant temperature of 20°C. Then the amount of water sorbed by bulk sample was measured at different times. The percentage of solvent uptake at each time is given by:

$$\text{Solvent uptake}\,(\%) = \frac{W_s - W_d}{W_{seq} - W_d} \times 100 \quad (2)$$

where, W_s is the swollen gel weight at time t.

Drug Loading

Gel beads were loaded with dilthiazem hydrochloride [22] by means of solution loading method. A weighed mass of dried gel sample was placed in a 10 wt% drug

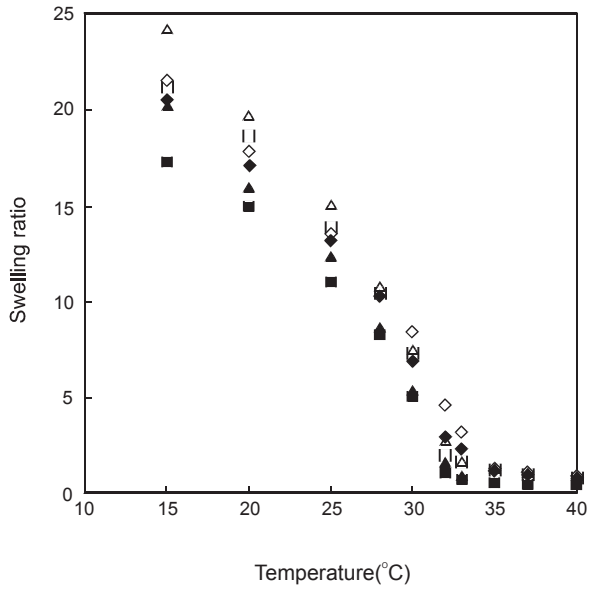

Figure 2. Effect of temperature on swelling behaviour of hydrogels; poly (IPAAm) & Ca alginate (Na-alginate conc. ■ 0.5% w/v, ▲ 0.3% w/v), poly (IPAAm) after extraction of Ca alginate (Na-alginate conc. □ 0.5% w/v, △ 0.3% w/v), ◆ poly (IPAAm-*co*-SA) & Ca-alginate, and ◇ poly (IPAAm-*co*-SA).

solution at room temperature (20°C) for three days. Then swollen gel beads were removed from the solution and dried under vacuum for 24 h at room temperature. The dry weight of drug loaded gels, W_D, was measured and the percentage of drug loading was calculated by the following equation:

$$\text{Drug loading}\ (\%) = \frac{W_D - W_d}{W_d} \times 100 \qquad (3)$$

Drug Release Kinetics

Several 25 mL-flasks were filled with a certain amount of PBS solution and placed in a shaker bath at 37°C. After reaching equilibrium temperature, a predetermined amount of loaded gel beads was introduced successively into the each flask for a certain time to meet the sink condition. At the end, gel beads were removed and drug concentration in each flask was measured by a spectrophotometer at 240 nm.

RESULTS AND DISCUSSION

Characteristics of Hydrogel Beads

Poly(IPAAm) and poly(IPAAm-*co*-SA) hydrogels were synthesized as uniform spherical beads. Gel beads were transparent with a size range of 1.5-3.0 mm in diameter. Hoffman and Park [19] synthesized poly(IPAAm) gel beads with the same procedure having an average size of 2 mm which is comparable with these results.

In preliminary experiments sodium metabisulphite (SMB) was used as an accelerator, but the resulting polymeric beads were non-uniform and opaque. Their apparent structure collapsed after extraction of calcium alginate. It can be postulated that there was an ionic interaction between carboxylic groups of alginate and metabisulphite anions. This ionic interaction forces SMB out of the gel beads and decreases the rate of redox reaction and free radical polymerization, correspondingly. This effect also decreases the rate of formation of calcium alginate gel and consequently results in non-uniform beads. Besides, due to decreased rate of polymerization reaction, calcium alginate proportion of the interpenetrating polymer networks might be more than poly(IPAAm). Therefore, extraction of calcium alginate by chelating agent would result in a loose hydrogel with collapsible structure. Based on this result the rest of experiments were carried out using TEMED as accelerator.

Equilibrium Swelling

Equilibrium swelling behaviour of poly(IPAAm) hydrogels at different temperatures is shown in Figure 2. As it was expected, the equilibrium swelling degree of alginate extracted homopolymer gel was greater than composite gel. However, the lower critical solution temperature (LCST) of poly(IPAAm) gels was not affected by the presence of Ca-alginate in the network. Comparison of swelling degrees of hydrogels prepared from monomer solutions containing 0.3% and 0.5% (w/v) sodium alginate indicates that increasing of sodium alginate results in increased hydrophobicity of composite gels. These phenomena can be explained by Lin [23] observations. Increasing Na-alginate concentration in monomer solution not only increases the Ca-alginate proportion of the resulting network and rigidity of the Ca-alginate gel shell, but also affects diffusion of calcium chloride and ammonium persulphate (APS) across this shell. During preparation of composite thermosensitive hydrogels, calcium chloride and APS molecules of the external solution diffuse inward through the developing gel shell, which advances towards the core. Since APS molecule is larger than calcium chlo-

ride molecules, its diffusion across outer gel shell reduces by increasing Na-alginate concentration in monomer solution. This in turn results in decreased rate of poly(IPAAm) gel formation.

Swelling behaviour of Ca-alginate extracted hydrogels indicate that extraction process is not complete because swelling degree of hydrogel prepared from monomer solution containing 0.5% (w/v) Na-alginate is less than that of hydrogel prepared from monomer solution containing 0.3% (w/v) Na-alginate.

Figure 2 shows equilibrium swelling behaviour of IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hydrogels prepared from monomer solution containing 0.5% (w/v). It is observed that the equilibrium swelling degree of ionic copolymer gels of poly(IPAAm-*co*-SA) is greater than that of poly(IPAAm) hydrogel due to the presence of ionic species within ionic network which contributes to higher swelling pressure.

It is interesting to note that the equilibrium swelling degree of IPNs of poly(IPAAm-*co*-SA) and calcium alginate gel is greater than that of Ca-alginate extracted copolymer gels below 35°C. This behaviour can be attributed to the osmotic coefficient of composite ionic network, which is neutralized by monovalent Na^+ ions and divalent Ca^{2+} ions. Dolar and Peterlin [24], theoretically, and then Dolar and Kozak [25] with quantitative experiments reasoned that whenever a polyelectrolyte solution is neutralized by mono- and divalent counterions, the osmotic coefficient of polyelectrolyte solution reaches maximum at a definite value of two - ions ratio. This theory has been proved quantitatively by Vasheghani-Farahani et al. [26] for osmotic pressure of anionic copolymer gels of acrylamide and sodium acrylate, which were neutralized by Na^+ and Ca^{2+} counterions. Accordingly, for the IPNs of poly(IPAAm-*co*-SA) and calcium alginate it is supposed that the osmotic coefficient of gel network due to the presence of Na^+ and Ca^{2+} counterions is at maximum value so that the equilibrium swelling degree of IPNs is greater than that of alginate extracted gel.

It is also observed that there is not a sharp phase transition temperature for ionic copolymer gel. This result can be explained by the presence of excessive amount of hydrophilic sodium acrylate, which resulted in omission of LCST. In other words, increased hydrophilic effect due to high concentration of SA prevails hydrophobic effect of substituted isopropyl group

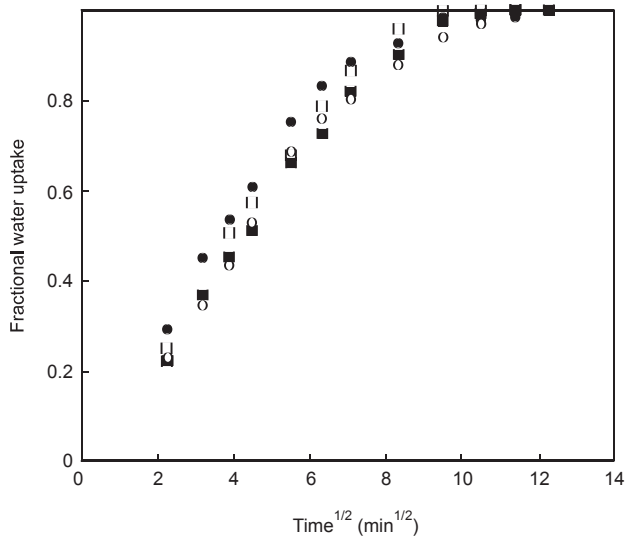

Figure 3. Fractional approach to eqilibrium swelling for IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hydrogels: ■ poly(IPAAm) & Ca-alginate, ● poly(IPPAm-*co*-SA) & Ca-alginate, □ poly(IPAAm), and ○ poly(IPAAm-*co*-SA).

in IPAAm structure and hence equilibrium swelling degree decreases with increasing temperature without any abrupt phase transition. This result is in agreement with those of Seida and Nakano [27].

Kinetics of Gel Swelling

The rate of solvent uptake by IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hydrogels prepared from monomer solution containing 0.5% (w/v) Na-alginate are shown in Figure 3. It can be seen that water absorption rate of alginate extracted poly(IPAAm) gel is greater than that of IPNs of poly(IPAAm) and calcium alginate gels. In fact, the void space left within the alginate extracted network has increased mass transfer area and hence resulted in faster approach to final degree of swelling.

In case of copolymer gels, water absorption rate for composite gel is more than that for non-composite gel. Increasing of osmotic coefficient of composite network, as mentioned earlier, has resulted in increase of chemical potential difference between polymer network and external solution. This effect has increased diffusion coefficient or mass transfer rate in turn.

These results also indicated that the process of water diffusion through these hydrogels is Fickian type because of linear relationship between solvent uptake and square root of time up to 60% of equilibrium state

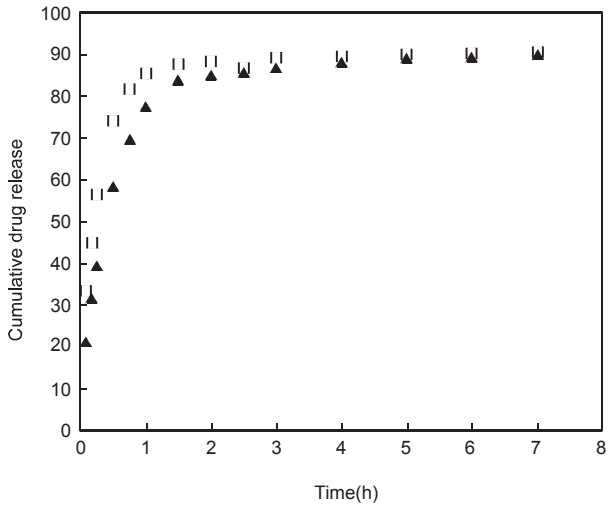

Figure 4. Cumulative release of dilthiazem hydrochloride as a function of time from IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hyrogels at pH 7: □ poly(IPAAm), and ▲ poly(IPAAm-*co*-SA).

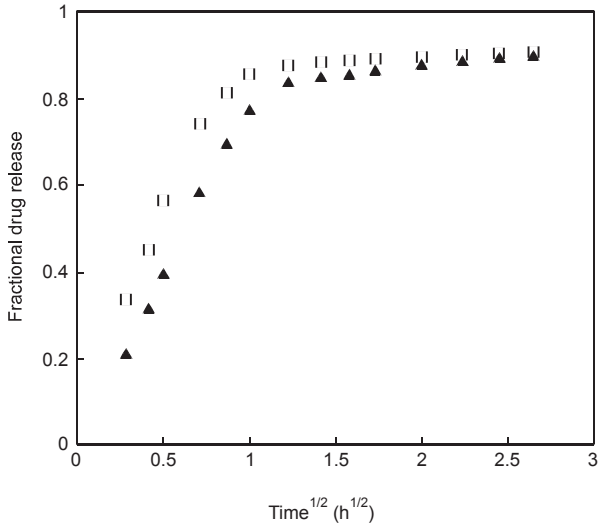

Figure 5. Fractional approach to eqilibrium release of dilthiazem hydrochloride from IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hydrogels: □ poly(IPAAm) and ▲ poly(IPAAm-*co*-SA).

(Figure 3). According to Fickian diffusion equation, at short periods, percent of solvent uptake is approximately proportional to the square root of time:

$$\text{Fractional solvent uptake} = 4\times\left[\frac{D.t}{\pi.l^2}\right]^{1/2} \quad (4)$$

Where, l is the characteristic length of device, D is diffusivity and t is time. In this case, short period approximation is valid for totally 60% of swelling process.

Drug Loading and Release Kinetics

The loaded amount of dilthiazem hydrochloride was, respectively, 65% and 66% for alginate extracted IPAAm and IPAAm-*co*-SA (90:10 molar ratio) hydrogels prepared from monomer solution containing 0.5% (w/v) Na-alginate.

Figure 4 shows the amount of drug release from these hydrogels as a function of time. It can be seen that almost 80% of entrapped drug released within an hour from polymer matrix due to fast penetration of water into the gel network. Thus, these hydrogels are suitable as fast responsive drug delivery systems if necessary. To determine the amount of drug, which is left within carrier, hydrogel beads were removed from solution and kept in distilled water overnight at 6°C. Concentration of drug released into distilled water indicated that 5.5% and 6.8% of initially loaded amount of drug was left within poly(IPAAm) and poly(IPAAm-*co*-SA) gels, respectively.

Fractional approach to the equilibrium release of drug vs. square root of time (Figure 5) indicates that dilthiazem hydrochloride release from the above mentioned hydrogels is Fickian. According to Fickian law, at short periods, fractional release is pproximately proportional to the square root of time:

$$\frac{M_t}{M_\infty} = 4\times\left[\frac{D.t}{\pi.l^2}\right]^{1/2} \quad (5)$$

where, l is thickness of slab, D is diffusivity, M_t is the amount of drug released by time t and M_∞ is the amount of drug released at infinite time. In this case, short period approximation is valid for totally 60% of release process.

CONCLUSION

The results of present study can be summarized as follows:

It was possible to synthesize *N*-isopropylacrylamide based hydrogels as single or IPNs in form of uniform, large size, spherical beads using sodium alginate without applying any emulsifier.

Equilibrium swelling degree of homopolymer gel

increased after extraction of calcium alginate due to increased hydrophilicity. Calcium alginate had no effect on LCST of homopolymer gel but equilibrium swelling degree decreased with the increase of sodium alginate concentration in monomer solution used for gel formation.

Because of augmented osmotic coefficient of ionic network neutralized by a mixture of mono- and divalent counterions, equilibrium swelling degree of copolymer composite hydrogel containing Na^+ and Ca^{2+} cations was greater than that of extracted hydrogel containing only monovalent cations.

Water absorption rate of porous alginate extracted homopolymer gel was greater than that of composite gel due to increased mass transfer area. Because of increased chemical potential, resulted from increased osmotic coefficient of ionic networks neutralized by a mixture of mono and bivalent counterions, the rate of water absorption by composite copolymer gels was more than that of alginate extracted gels. Kinetics of swelling indicated a Fickian type diffusion of water through these hydrogels.

Dilthiazem hydrochloride was easily loaded in alginate extracted homo- and copolymer gel beads by solution loading method. Drug release from these hydrogels was fast and in Fickian manner rendering them suitable as fast responsive drug delivery systems.

ACKNOWLEDGEMENTS

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REFERENCES

1. Huang X. and Brazel C.S., On the importance and mechanisms of burst release in matrix-controlled drug delivery systems, *J. Cont. Rel.,* **73**, 121-136 (2001).
2. Yaung J.F. and Kwei T.K., pH-Sensitive hydrogels based on polyvinylpyrrolidone-poly acrylic acid (PVP-PAA) semi-interpenetrating networks (semi-IPN): swelling and controlled release, *J. Appl. Polym. Sci.,* **69**, 921-930 (1998).
3. Mika A.M., Childs R.F., and Dickson M., Chemical valves based on poly(4-vinylpyridine)-filled microporous membrane, *J. Memb. Sci.,* **153**, 45-56 (1999).
4. Sershan S.R., Westocott S.L., Halas N.J., and West J.L., Temperature sensitive polymer nano shell composites for photothermally modulated drug delivery, *J. Biomed. Mater. Res.,* **51**, 293-298 (2000).
5. Chen J., Park H., and Park K., Synthesis of super porous Hydrogels: hydrogels with fast swelling and super absorbent properties, *J. Biomed. Mat. Res.,* **44**, 53-62 (1999).
6. Amsden B. and Turner N., Diffusion characteristic of calcium alginate gels, *Biotech. Bioeng.,* **65**, 605-610 (1999).
7. Chiu H.C., Wu A.T., and Lin Y.F., Synthesis and characterization of acrylic acid-containing dextran hydrogels, *Polymer,* **42**, 1471-1479 (2001).
8. Kim S.Y. and Lee Y.M., Drug release behavior of electrical responsive poly(vinyl alcohol)/poly(acrylic acid) IPN hydrogels under an electric stimulus, *J. Appl. Polym. Sci.,* **74**, 1752-1761 (1999).
9. Kim S.Y., Shin H.S., Lee Y.M., and Jeong C.N., Properties of electro-responsive poly (vinyl alcohol)/ poly(acrylic acid) IPN hydrogel under an electric stimulus, *J. Appl. Polym. Sci.,* **73**, 1675-1683 (1999).
10. Otake K., Inomata H., Konno M., and Saito S., Thermal analysis of the volume phase transition with *N*-isopropylacrylamide gels, *Macromolecules,* **23**, 283-289 (1990).
11. Hirokawa Y. and Tanaka T.J., Volume phase transition in a non-ionic gel, *J. Chem. Phys.,* **81**, 6379-6380 (1984).
12. Vasheghani-Farahani E., Cooper D.G., Vera J.H., and Weber M.E., Concentration of large biomolecules with hydrogels, *Chem. Eng. Sci.,* **47**, 31-40 (1992).
13. Zhang X-Z and Zhuo R.X., Preparation of fast responsive, temperature sensitive poly(*N*-isopropylacrylamide) hydrogel, *Macromol. Chem. Phys.,* **200**, 2602-2605 (1999).
14. Zhuang Y., Chen L., Zhu Z., and Yang H., Preparation and separation function of *N*-isopropylacrylamide copolymer hydrogels, *Polym. Adv. Tech.;* **11**, 192-197 (2000).
15. Lee W.F. and Yen S-H., Thermoreversible hydrogels. XI. Effect of the polymerization conditions on the swelling behavior of the *N*-isopropylacrylamide gel, *J. Appl. Polym. Sci.,* **78**, 1604-1611 (2000).
16. Champ S., Xie W., and Huglin M.B., Thermal effects in the synthesis of thermo-responsive hydrogels of poly(*N*-isopropylacrylamide-co-acrylic acid), *Macromol. Mater. Eng.,* **282**, 3462-3466 (2000).
17. Zhang X-Z and Zhuo R-X., A novel method to prepare a fast responsive, thermo-sensitive poly(*N*-isopropylacrylamide) hydrogel, *Macromol. Rapid Commun.,* **20**, 229-231 (1999).
18. Liu Y., Velada J.L., and Huglin M.B., Thermo-reversible

swelling behavior of hydrogels based on *N*-Isopropylacrylamide with sodium acrylate and sodium methacrylate, *Polymer,* **40**, 4299-4306 (1999).
19. Hoffman A.S. and Park T.G., Preparation of large, uniform-size temperature sensitive hydrogel beads, *J. Polym. Sci., Polym. Chem. Ed.,* **30**, 505-507 (1992).
20. Bromberg L.E. and Ron E.S., Temperature-responsive gels and thermogelling polymer matrices for protein and peptide delivery, *Adv. Drug Deliv. Rev.,* **31**, 197-221 (1998).
21. Rudin A., *The Elements of Polymer Science and Engineering,* Academic, Orlando; 246-247 (1982).
22. Katzung B.G., Ed., *Basic and Clinical Pharmacology,* Printice-Hall International, London; Ch. 12 (1989).
23. Lin S.H., Gellation of sodium alginate in a batch process; *Chem. Eng. Sci.,* **46**, 651-655 (1991).
24. Dolar D. and Peterlin A., Rodlike model for a polyelctrolyte solution with mono- and divalent counterions, *J. Chem. Phys.,* **50**, 3011-3015 (1969).
25. Dolar D. and Kozak D., Osmotic coefficients of polyelectrolyte solutions with mono- and divalent counterions, *Proc. Leiden Symp.,* **11**, 363-366 (1970).
26. Vasheghani-Farahani E., Vera J.H., Cooper D.G., and Weber M.E., Swelling of ionic gels in electrolyte solutions, *Ind. Eng. Chem. Res.,* **29**, 554-556 (1990).
27. Seida Y. and Nakano Y., Effect of pH on the phase transition of *N*-isopropylacrylamide-sodium acrylate copolymer gel, *J. Chem. Eng. Japan,* **26**, 328-330 (1993).