Semiinterpenetrating Polymer Networks of Chitosan and L-Alanine for Monitoring the Release of Chlorpheniramine Maleate

Kamlesh Kumari, P. P. Kundu

Department of Chemical Technology, Sant Longowal Institute of Engineering and Technology, Longowal 148106, India

Received 8 July 2006; accepted 6 September 2006
DOI 10.1002/app.25432
Published online in Wiley InterScience (www.interscience.wiley.com).

ABSTRACT: *In vitro* studies have been carried on semiinterpenetrating polymer network (IPN) beads of chitosan–alanine as carrier for the controlled release of chlorpheniramine maleate (CPM) drug. A viscous solution of chitosan–alanine was prepared in 2% acetic acid solution, extruded as droplets by a syringe to NaOH–methanol solution and crosslinked using glutaraldehyde as a crosslinker. The swelling behavior of crosslinked beads in different pH solutions was measured at different time intervals. The swelling behavior was observed to be dependent on pH and degree of crosslinking. The structural and morphological studies of beads were carried out by using a scanning electron microscope. The drug release experiments of different drug loading capacity beads were performed in solutions of pH 2 and pH 7.4 using CPM as a model drug. The concentration of the released drug was evaluated using UV spectrophotometer. The results suggest that chitosan–alanine crosslinked beads are suitable for controlled release of drug. © 2006 Wiley Periodicals, Inc. J Appl Polym Sci 103: 3751–3757, 2007

Key words: chitosan; controlled drug release; interpenetrating polymer networks; alanine; beads; crosslinking; swelling

INTRODUCTION

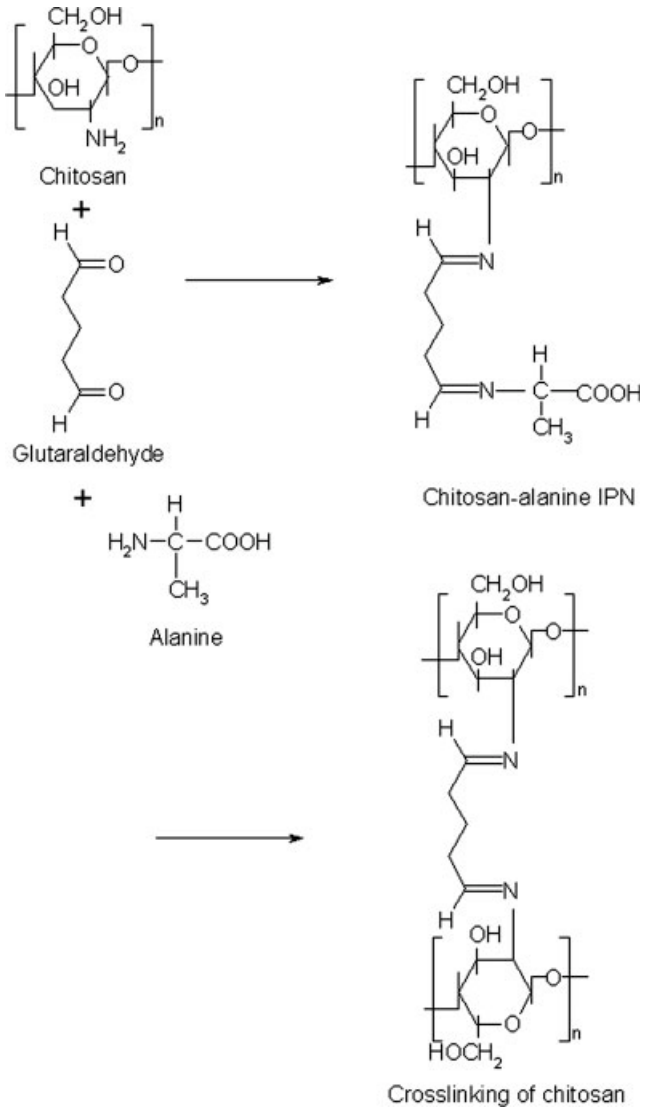
Over the past several decades, much interest has been focused on designing new drug dosage formulation to enhance the effectiveness of the existing medications. The meaning of ''controlled drug delivery'' expands to the targeting of drug at a specific body site or releasing drugs, when needed, as well as controlling the release rate of the drug. Stimuli-sensitive drug delivery has been required depending on the changes in physiological signals in the body. The advantages of using drug delivery system rely on maintaining the right concentration of the drug at the right period, the therapeutic level, and targeting the drug for the site of action, avoiding any side effect.

Generally, natural/semisynthetic polymers are preferred as a vehicle for drug delivery as they are biodegradable, nontoxic as well as biocompatible. Chitosan [(1 → 4) 2-amino-2-deoxy, β-D-glucan] is a semisynthetic polymer, normally obtained by alkaline deacetyation of chitin. Chitosan, a natural polycation, is nontoxic, biocompatible, and biodegradable, has excellent gel- and film-forming ability and structurally resembles glycans. Moreover, chitosan has antacid and antiulcer activities that prevent or weaken drug irritation in stomach.[1] Chitin is known to consist of 2-acetamido-2-deoxy-β-D-glucose through a β (1 → 4) linkage and occurs principally in animals of the phylum arthropoda. The necessary degree of deacetylation to obtain a soluble product must be 80 to 85% or higher; i.e., the acetyl content of the chitosan product must be <4–4.5%.[2–4] Practical grade chitosan obtained from crab shells has a minimum of 85% deacetylation and a viscosity >200 cps (Brookfield, 1% solution in 1% acetic acid); it may contain zsome foreign matters. High molecular weight (600,000 Daltons) chitosan is a coarsely ground polymer prepared from crab or shrimp shells with a viscosity of 800–2000 cps. Low molecular weight (150,000 Daltons) chitosan is 75–85% deacetylated and has a viscosity of 20–200 cps. Chitosan dissolves readily in dilute solutions of most organic acids, including formic, acetic, tartaric, and citric acids. Chitin and chitosan are of commercial interest because of their high percentage of nitrogen (6.89%) when compared with synthetically substituted cellulose (1.25%). Because chitosan is easily soluble in acid, crosslinking of chitosan to form a network is the only way to prepare chitosan IPNs or microspheres.

L-Alanine is an organic compound, one of the 20 amino acids, commonly found in animal proteins. It is hydrophobic, with nonpolar methyl group side chain, and is the second smallest of the 20 after gly-

Correspondence to: P. P. Kundu (ppk923@yahoo.com).

Journal of Applied Polymer Science, Vol. 103, 3751–3757 (2007)
© 2006 Wiley Periodicals, Inc.



Scheme 1 Crosslinking of chitosan and L-alanine by glutaraldehyde for the formation of Inter penetrating Network (IPN).

cine. It is only the levorotatory (l)—stereoisomer that participates in the biosynthesis of proteins. The α-carbon in alanine is substituted with a methyl group, making it one of the simplest amino acids with respect to the molecular structure and is one of the most widely used in protein construction.[5–7] The biocompatibility is the reason behind the use of L-alanine along with chitosan.

To form semiinterpenetrating polymer network (IPN), two chitosan polymer chains are crosslinked by glutaraldehyde. Amino groups of chitosan and alanine can react with glutaraldehyde resulting in the attachment of alanine in pendent form (Scheme 1). Hence, the resultant polymer system is characterized as semi-IPN (one phase consists of crosslinked chitosan and the other phase is made of alanine attached chitosan).

MATERIALS AND METHODS

Chitosan was purchased from Tokyo Kasei Kogyo, Japan and was used as received. Its percentage of deacetylation after drying was 80%; total nitrogen: 7% minimum, loss on drying < 15%, and ignition residue (sulfate): <2%. Chlorpheniramine maleate (CPM) [$C_{16}H_{19}ClN_2C_4H_4O_4$] was obtained as a gift sample from Japson Pharmaceuticals, Sangrur, India. Glutaraldehyde ($C_5H_8O_2$) (MW = 100.11), acetic acid, and L-alanine ($CH_3CH(NH_2)COOH$) (MW = 89.09) were procured from CDH, New Delhi.

Preparation of crosslinked chitosan–alanine beads

Purified chitosan and alanine were dissolved in 2% acetic acid solution by stirring conditions for 3 h at room temperature. The homogeneous mixture was extruded in the form of droplets using a 0.56 mm diameter syringe into NaOH–methnol solution (1 : 20 w/w) under stirring conditions. The beads were washed thrice with hot (50°C) and cold (25°C) water and it generally took 2–3 min, respectively. The resultant beads were allowed to react with glutaraldehyde solution at 50°C for about 10 min for crosslinking. The formation of crosslinked chitosan–alanine IPN is shown in Scheme 1. Finally, the crosslinked beads were successively washed with hot (50°C) and cold (25°C) water, respectively, and vacuum dried for 30 min at −700 mmHg and 55°C. The composition of the prepared beads is given in Table I.

For preparing drug-loaded beads, a known amount of CPM (75, 100, and 125 mg, respectively) was added to the chitosan–alanine mixture before extruding into the alkaline–methanol solution.

Swelling studies

Swelling behavior of chitosan beads (A1–A5) at different pH has been studied. The degree of swelling for each sample at time t was calculated using the relationship:

$$\text{Degree of swelling} = (W_t - W_0)/W_0$$

Where W_t and W_0 are the weights of the beads at time t and in the dry state, respectively.

TABLE I
Composition and Designation of IPN Beads

Sample no.	Chitosan (g)	Analine (g)	2% Acetic acid (mL)	Glutaraldehyde, 10 mL (%)
A1	0.5	0.5	20	25.00
A2	0.5	0.5	20	12.50
A3	0.5	0.5	20	06.25
A4	0.5	0.5	20	03.13
A5	0.4	0.5	20	12.50

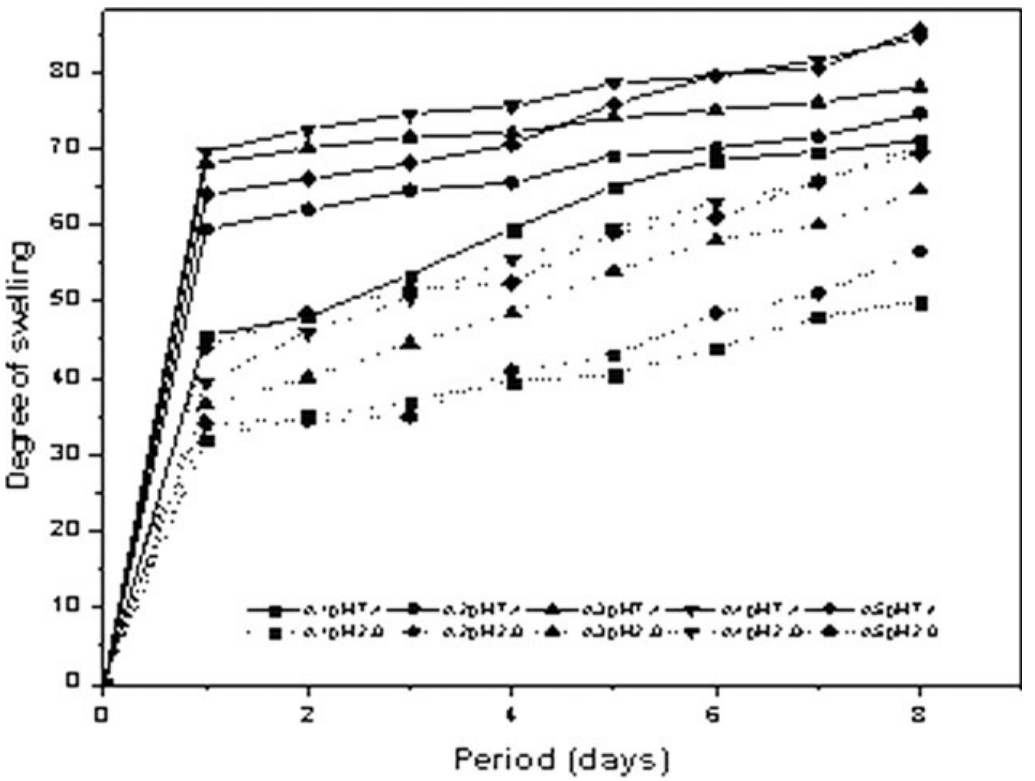


Figure 1 Swelling behavior of the crosslinked beads measured as a function of time in pH 2.0 and pH 7.4.

Scanning electron microscopy

The shapes and surface morphology of the beads were examined by using a scanning electron microscope (SEM). For SEM studies, the samples were mounted on metal stubs using double-sided adhesive tape and vacuum coated with gold.

Drug release studies

The release experiments were performed in a glass apparatus at 37°C under unstirred conditions in acidic (pH = 2) and basic (pH = 7.4) solution. Beads (0.2 g) containing known amounts of the drug were added to the release medium (30 mL). At predecided intervals, samples of 3 mL were withdrawn, filtered, and assessed by recording the absorbance at 193.5 nm. To maintain nearly constant release environment, the samples withdrawn for the record of absorbance were immediately added back to the release medium after recording the absorbance.

RESULTS AND DISCUSSION

Swelling studies

Swelling response of the glutaraldehyde-crosslinked chitosan–alanine beads in solutions of pH 2.0 and pH 7.4 is shown in Figure 1. The observed swelling rates of the crosslinked beads followed the order A5 > A4 > A3 > A2 > A1. Generally, the swelling process of the beads in pH < 6 involves the protonation of amino/imine groups in the beads and mechanical relaxation of the coiled polymeric chains. The process of protonation is expected to be completed in two stages. In the first stage, amino/imine groups of the bead surface were protonized, and that led to dissociation of the hydrogen bonding between amino/imine groups and other groups. The protonation resulted in solvent invading the polymer from the sample surface and forming a sharp boundary or moving front separating the unsolvated polymer region with that of the swollen portion of the beads. In the second stage, protons and counterions diffused into the bead to protonate the amino/imine groups inside the beads and dissociating the hydrogen bonds. This process of protonation continued until the whole structure of the beads was collapsed and solvated.[2,3] It has been observed that the swelling rates are directly proportional to the degree of crosslinking. In both cases, the degree of swelling is very high in solution of pH 7.4 compared to that of pH 2. Sample A5 has lesser composition of chitosan when compared with Sample A2. The lesser crosslinked density resulted because of the decrease in chitosan composition is the main reason of higher degree of swelling of A5 beads in both the mediums.

SEM studies

Crosslinked chitosan–alanine beads prepared by using different concentrations of glutaraldehyde have

rough and dense surfaces. The observed shape of the bead was like a droplet and is shown in Figure 2(a). The reason for nonspherical size may be due to higher viscosity of the chitosan–alanine solution. The approximate size of the beads is in the range of 0.8–1.2 mm. From the morphology of the beads shown in Figure 2(b), one can observe rough and folded surfaces of the beads. With the higher concentration of crosslinker, the chains come closer to each other and give a regular, fibrous structure, but with decreasing degree of crosslinking, the structural morphology changes to granular. The same pattern, however, was not observed in case of A3. This may be because of the higher time taken for the transfer of beads to the crosslinker solution leading to the phase separation of chitosan and L-alanine.

Drug release studies

The drug release experiments were performed with the 0.2 g of sample containing 15, 20, and 25 mg of CPM drug, respectively. Figures 3–5 show the dissolution profile of chlorpheniramine maleate from crosslinked chitosan–analine beads at various time intervals in acidic (pH 2.0) and basic (pH 7.4) medium. There is a burst release initially for the first hour in both media followed by a moderate release for next 5 h and finally an almost constant release of CPM from the matrix is observed for the studied period of 48 h. The amount and percentage of drug released were much higher in basic solution than in acidic solution, because the release rate depends on the swelling of the beads.

It has been observed from the dissolution profiles of CPM from the crosslinked chitosan–alanine beads at various time intervals that the release of drugs from the beads in basic medium is higher than that in acidic one. This can be explained by the fact that the release of drug, due to diffusion through the swollen beads, depends mainly on the degree of swelling of beads. At pH 2.0, there is less swelling; thus, drug entrapped in the beads cannot be released easily. However, at pH 7.4, the beads are swollen to a greater extent, leading to faster release of the drug.

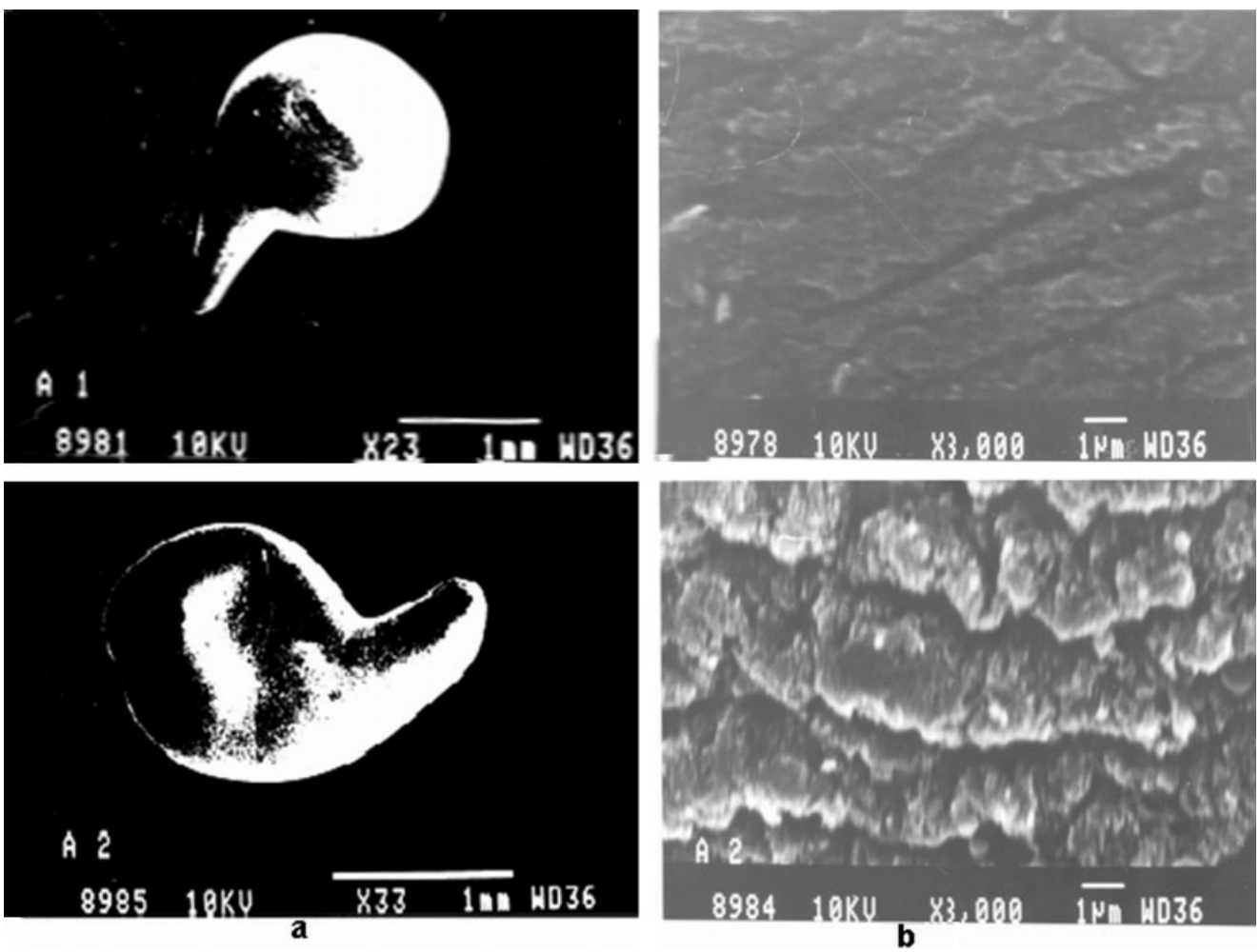


Figure 2 (a) SEM photographs of the crosslinked beads (A1–A5) and (b) their morphology.

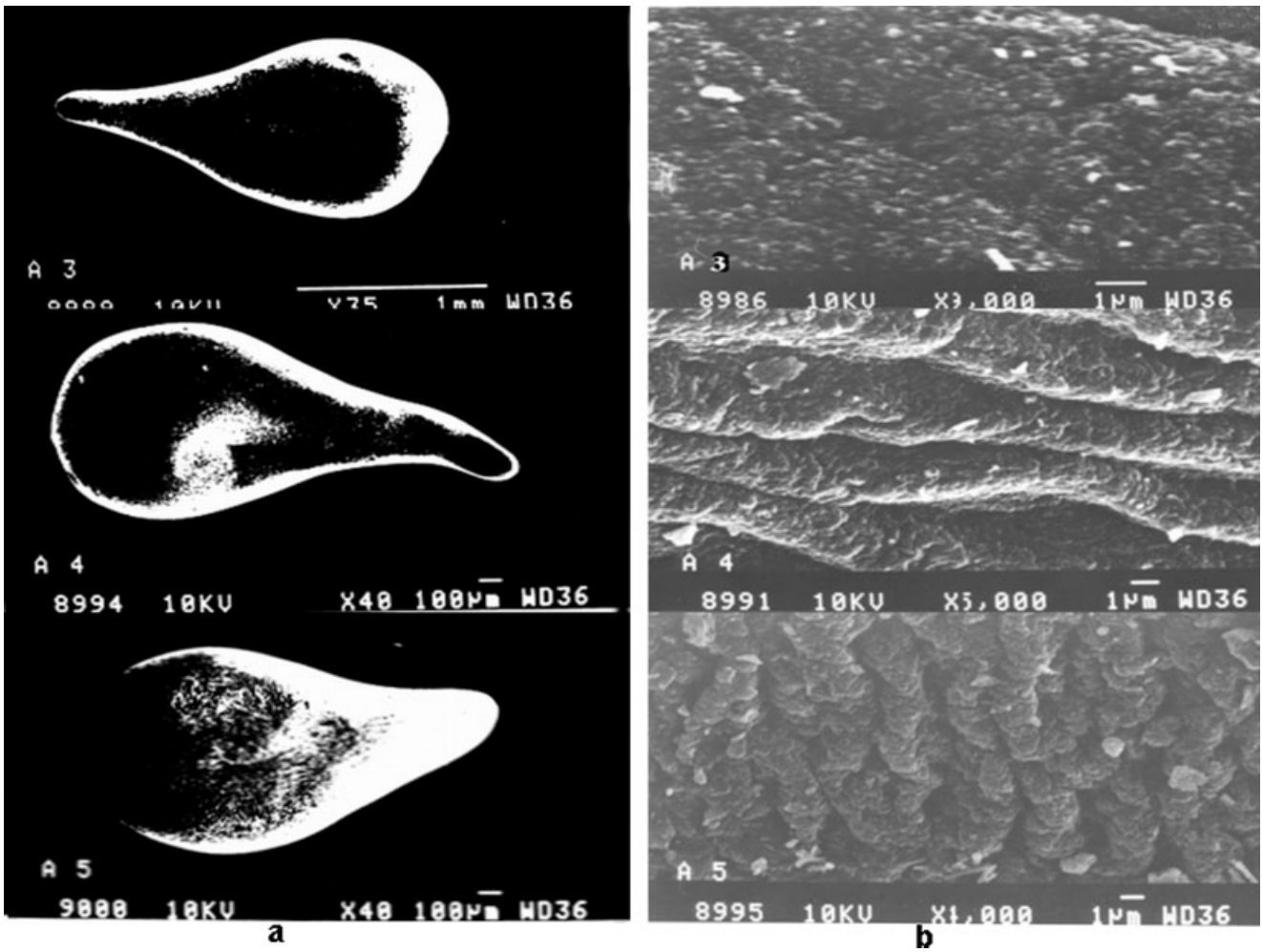


Figure 2 (*Continued from the previous page*)

The reason of higher drug release rate in the basic medium may be because of the presence of free carboxylic end of the crosslinked chitosan–alanine IPN (Scheme 1), which is more susceptible to be attacked by the basic solution. The drug release in the acidic medium is because of the interaction of acidic solution with the polar groups of IPN.

Similar studies[2] have been reported for chitosan–glycine system, but the release rates in the two systems are entirely different. According to the earlier report, the amount and percentage of drug released is higher in acidic media, whereas we observed that it is the basic medium in which the release rate is higher. This may be because of the different chemical structures of glycine and alanine. In case of alanine, steric hindrance is more when compared with glycine due to presence of an additional CH_3. As a result of which carboxylic group in alanine is free from such hindrance and alkaline group can easily penetrate into the system. In the case of glycine, because of the higher electron density, more electrons are available for the reaction with acid and they will form NH_3^+ (ammonium ions), but the same is not facilitated in case of alanine.

Our results are further supported by Peppas,[8] according to whom, many of the potentially most useful pH-sensitive polymers swell at high pH values and collapse at low pH values, and thus, the triggered drug delivery occurs upon an increase in the pH of the environment. Such materials are ideal for systems such as oral delivery, in which the drug is not released at low pH values in the stomach but rather at high pH values in the upper small intestine.

Further, the release pattern of the highly drug-loaded beads has been found to be similar to that of the beads loaded with a lower amount of drug. The percentage of release of drug from chitosan beads decreased with increased concentration of drug. However, the total amount of drug released from the beads loaded with a higher amount of drug was found to be higher in comparison to the beads loaded with a lower amount. It is because of the limited amount of drug encapsulation into the polymer metrics. It is further believed that the unentrapped drug

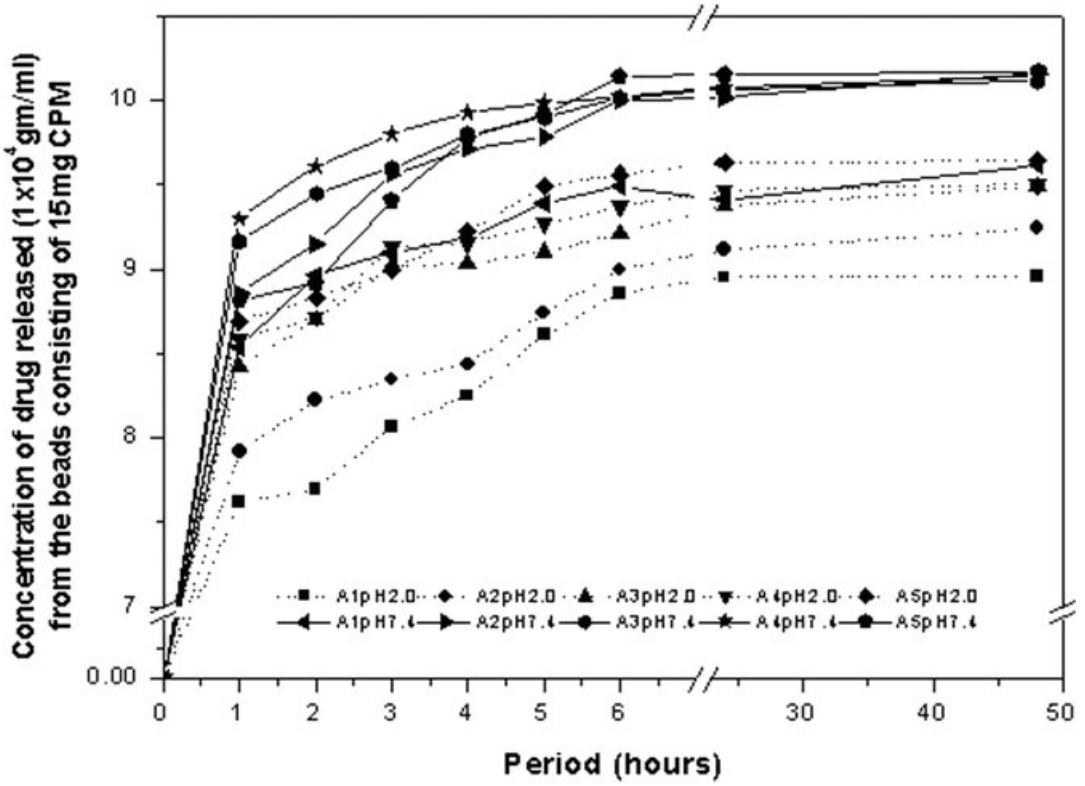


Figure 3 Release of CPM (15 mg) loaded beads versus time in solutions of pH 2.0 and pH 7.4.

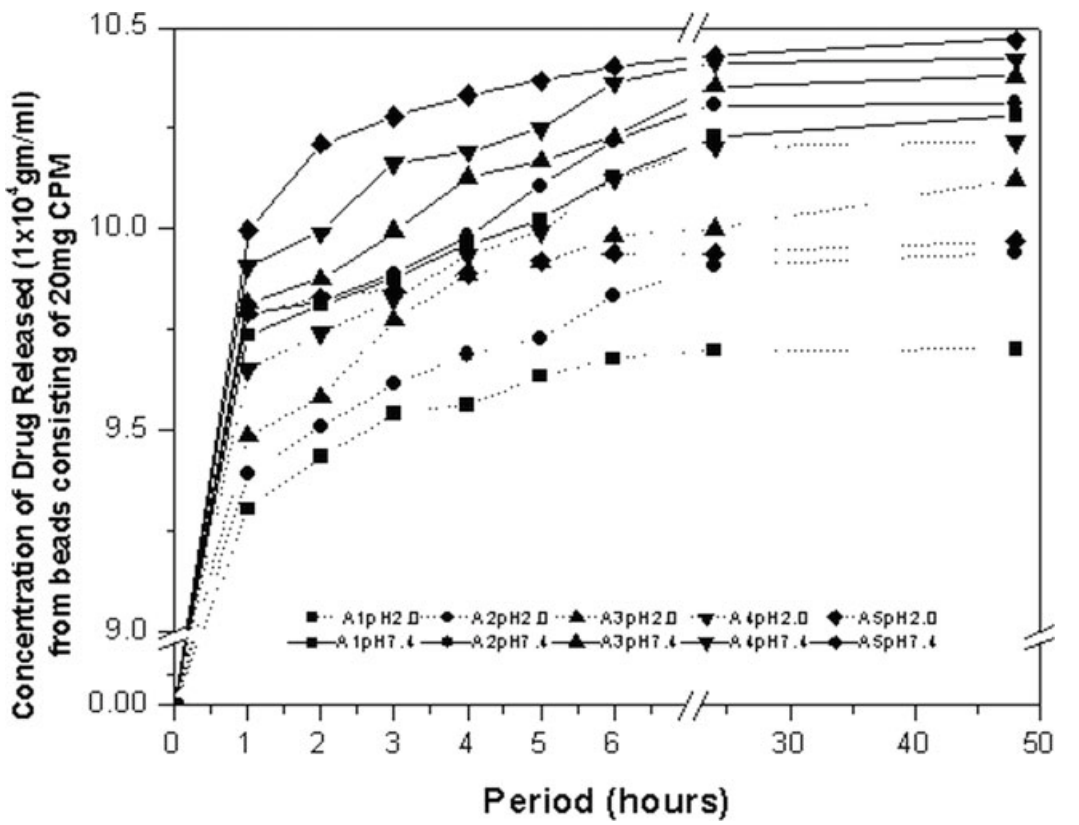


Figure 4 Release of CPM (20 mg) loaded beads versus time in solutions of pH 2.0 and pH 7.4.

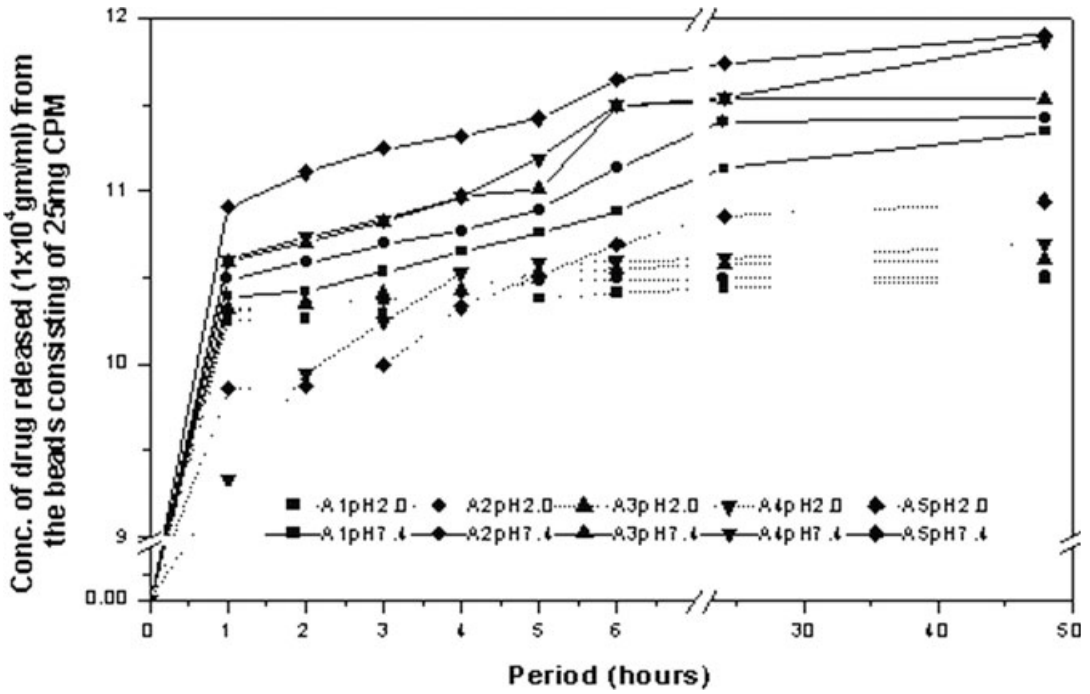


Figure 5 Release of CPM (25 mg) loaded beads versus time in solutions of pH 2.0 and pH 7.4.

is removed during the successive washing of beads and hence the percentage of drug release decreases with increase in drug concentration in the semi-IPN.

The maximum amount of drug released from 0.2 g of sample containing 15 mg CPM is 9.2 mg in basic and 8.7 mg in acidic solution, and the lower limit of drug released from the same composition is 8.6 mg in basic and 8.1 mg in acidic medium. The maximum amount of drug released increases to 10.8 mg in basic and 9.8 mg in acidic solutions. In case of 25 mg, CPM loaded sample of beads and the minimum amount of drug released is 10.2 mg and 9.4 mg in the basic and acidic mediums, respectively. The swelling experiments indicate that the solvent well penetrate into the drug loaded samples. From the thermodynamic point of view, the drug release is governed by the concentration gradient. Thus, the maximum amount of drug released approaches the maximum drug present in the sample with time (e.g., 9.2 mg from 15 mg drug-loaded beads).

CONCLUSIONS

Chitosan–alanine beads were crosslinked with varying concentrations of glutaradehyde to form IPNs with different degrees of crosslinkings. The morphological studies of the samples were carried out by using a scanning electron microscope. The swelling of beads and release rate of CPM in different pH solutions were studied. It was observed that the release of CPM is more in basic medium than in acidic medium due to higher degree of swelling and the chemical structure of alanine. From these investigations, it is evident that the rate of swelling of matrix and release of drugs is dependent on the degree of crosslinking and the solution pH. Therefore, by varying the crosslink densities, desired release rates can be achieved from diffusion-controlled chitosan–alanine semi-IPN. Further, it also helps in optimizing drug-entrapping capacity and its sustained release for an extended period of time.

References

1. Kumar, M. N. V. R. React Funct Polym 2000, 46, 1.
2. Kumar, M. N. V. R.; Gupta, K. C. J Appl Polym Sci 2000, 76, 672.
3. Gupta, K. C.; Kumar, M. N. V. R. J Appl Polym Sci 2001, 80, 639.
4. Gupta, K. C.; Kumar, M. N. V. R. Biomaterials 2000, 21, 1115.
5. Damrau, F. J Am Geriatr Soc 1962, 10, 426.
6. Feinblatt, H. M.; Gant, J. C. J Maine Med Assoc 1958, 49, 99.
7. Zello, G. A.; Wykes, L. F.; Ball, R. O. J Nutr 1995, 125, 2907.
8. Peppas, L. B. Polymers in Controlled Drug Delivery. Biomaterials 1997, 4, 34.