VERIZONA PUBLISHER
Publishing Your Ideas

www.verizonaonlinepublishing.com
Vol: 1, Issue: 3

Journal of Drug

Temperature-sensitive Ternary Interpenetrating Polymeric Networks for Potential Gastrointestinal Drug Release

Sema Ekici[1*] and Dursun Saraydin[2]

[1]Faculty of Sciences and Arts, Department of Chemistry Hydrogel Research Laboratory Terzioglu Campus, Canakkale Onsekiz Mart University, 17020 Canakkale, Turkey

[2]Faculty of Sciences, Department of Chemistry, Hydrogel Research Laboratory, Cumhuriyet University, Sivas, Turkey

***Corresponding author:** Sema Ekici, Assoc Prof Dr, Canakkale Onsekiz Mart University, Faculty of Sciences and Arts, Department of Chemistry Hydrogel Research Laboratory Terzioglu Campus, 17020 Canakkale, Turkey; E mail: sekici@comu.edu.tr

Article Type: Research, **Submission Date**: 30 March 2017, **Accepted Date:** 09 May 2017, **Published Date:** 12 September 2017.

Citation: Sema Ekici and Dursun Saraydin (2017) Temperature-sensitive Ternary Interpenetrating Polymeric Networks for Potential Gastrointestinal Drug Release. J. Drug 1(3): 1-6.

Abstract

Interpenetrating polymeric network (IPN) structures have been studied extensively as drug carriers to enhance the required therapeutic effect. In this study, IPNs were prepared in the form of cylindrical by using chitosan known to be biocompatible with the body, poly(N-isopropyl acrylamide) (PNIPAM) which has temperature-sensitive nature, and poly(N-vinyl pyrrolidone) (PVP). N,N'-methylenebisacrylamide (MBA) and glutaraldehyde (GA) were selected as crosslinkers to clamp polymer chains. GA were used in different concentration to control the network porous of the IPNs. Dynamic swelling studies were carried out at pH 1.1 and pH 7.4. Pulsatile swelling studies were performed on all IPN hydrogels to determine to what extent the hydrogels would respond to changes in environmental pH and temperature and how fast that response would be. Amoxicillin was loaded into the hydrogels, and released into buffered solutions as a function of pulsatile changes in temperature and pH. Amoxicillin was loaded into the hydrogels, and released into buffered solutions as a function of pulsatile changes in temperature and pH. The final percentage of amoxicillin released is noted to decrease as the content of GA in the hydrogel increases. Data clearly demonstrate that the use of thermosensitive IPN hydrogels can increase the release of drug molecules from hydrophilic IPNs based on PNIPAM. It can be said from these values that IPNs are intelligent hydrogels and can be thought of as potential devices for pH- and temperature-stimulated sustained delivery of drugs into physiological solutions or for other biomedical applications.

Keywords: Drug release, Chitosan, Hydrogel networks, Amoxicillin, Crosslinking, Glutaraldehyde, Poly(N-isopropyl acrylamide), Interpenetrating network.

Introduction

Interpenetrating polymeric networks (IPNs) comprise two or more independent polymer networks that are formed in presence of one another. Nowadays many natural hydrogels such as chitosan, chitin, gelatin, hyaluronic acid, dextran, etc., and synthetic hydrogels such as polyacrylamide, poly(acrylic acid), poly(N-isopropyl acrylamide), and poly(N-vinyl pyrrolidone) have been used extensively for preparing IPNs [1-4]. IPNs share properties characteristic of each network [5,6]. In our previous papers [7,8] we have synthesized new IPNs composed of chitosan, (PVP), poly(acrylic acid), and polyacrylamide hydrogels. These IPNs were included in the class of the environmental-sensitive drug release devices. In this investigation we prepared ternary IPN systems in a single step solution polymerization and carried out for gastrointestinal antibiotic release. With this aim, chitosan, PVP, and PNIPAM were chosen as the components of the IPNs.

Chitosan is a deacetylated derivative of chitin, a naturally occurring polymer. It is widely studied in pharmaceutical and biomedical fields because of biodegradability, biocompatability and interesting structural properties (presence of amino and hydroxyl groups). Chitosan (polysaccharide) and cyclodextrins (oligosaccharides) can be used as carriers for release of drugs and bioactive molecules [9-12]. On the other hand, the lack of mechanical strength, required chemical functionality and physical properties necessitates the enhancement of characteristics in polymeric biomaterials with novel properties. PVP is a polymeric compound that is widely used for textiles, cosmetics and toiletries, pharmaceuticals, and medical applications [13,14]. PNIPAM hydrogels possessing a lower critical solution temperature (LCST, about 34 °C) in the range between room and body temperatures are one of the most widely

studied gels. Crosslinked PNIPAM gels swell at temperatures lower than about 34 °C and collapse at temperatures higher than this value [15,16]. This type of thermo-responsive behaviour has demonstrated successful drug release. PNIPAM hydrogel, after phase separation, regulates the release of the drug from gel to medium.

The localized treatment of H. pylori infections of the stomach could be scientifically improved if a site-specific antibiotic drug delivery system could be developed. H. pylori is closely associated with the adherent mucus layer, living both within and beneath it and adhering to the gastric epithelial cells. The failure of conventional treatments could be due to poor permeability of the antibiotics across the mucus layer or from subtherapeutic antibiotic concentrations at the site of infection after administration from conventional tablets or capsules. pH- or temperature-sensitive swelling hydrogels seem to be useful for localized antibiotic delivery in acidic environment of gastric fluids. One of the most important advantage of these hydrogels is that formulations remain on the targeted site more time than conventional ones [17-22].

Here we developed new chitosan-poly (N-vinyl pyrrolidone)-poly(N-isopropyl acrylamide) IPNs with different glutaraldehyde concentration to take advantage of the desirous properties of all polymers. Because chitosan is not an easy polymer to use, we have chosen to work with other hydrophilic polymers with chitosan. Although chitosan is a biocompatible and naturally occurring polymer, it has poor mechanical properties, i.e. chitosan beads and films are very brittle. These undesirable properties of a hydrogel can limit its bio-applications. Besides, PNIPAM hydrogel was selected as temperature-sensitive component of the IPNs. Cylindrical shaped IPN hydrogels were prepared for oral administration and characterized. Two different crosslinkers (GA and MBA) were used to control the network properties. These IPNs were named as IPN-1, IPN-2, and IPN-3. IPNs were loaded with a model drug, amoxicillin, which used in treatment of diseases caused by H. pylori. pH- (1.1 and 7.4) and temperature- (25 °C and 37 °C) sensitive swelling and release studies were realized to calculate swelling and network parameters of the IPNs and to optimize the IPN carrier for possible oral administration.

Experimental

Chemicals

Chitosan (M_w~600.000 g mol^{-1}) was purchased from Fluka (Steinheim, Switzerland) and PVP (M_w=40.000 g mol^{-1}) was obtained from Calbiochem (Darmstadt, Germany). NIPAM is a product of Aldrich (Milwaukee, USA). Hydrochloric acid, potassium chloride, disodium hydrogen phosphate and potassium dihydrogen phosphate were purchased from Merck (Darmstadt, Germany). MBA and GA (25% w/w) were used as crosslinkers and ammonium persulfate (APS) as redox initiator were obtained from Merck (Schuchardt, Germany). The model drug, amoxicillin trihydrate, was also a product of Fluka (Steinheim, Switzerland). All chemicals were of analytical grade and were used as received. Double-distilled water was used for all the experiments.

Preparation of the IPNs

PVP, crosslinkers and NIPAM were added to solution of chitosan (10 mL, 1 wt% in 0.8 wt% acetic acid) under continuous mixing. Chitosan, PVP and NIPAM were used in 4.0:4.0:90 weight ratio. The ratios (w/w) between GA and chitosan are 0.5, 1 and 2 for IPN-1, IPN-2, and IPN-3, respectively. The weight ratio of MBA to NIPAM was 2:100. APS solution (25 μL, 5 wt%) was added to this mixture. The mixture was placed in PVC straws of 3 mm diameter. A gel formed after 10 h of reaction time at ambient temperature. After 24 h, the IPNs were obtained in long cylindrical shapes, and were cut into pieces of 4-5 mm in length and washed with distilled water. Then, IPNs were dried in air and vacuum, and stored for further use.

Swelling studies

Swelling studies of IPNs were done as dynamic and pulsatile equilibrium swelling experiments. For dynamic swelling experiments, IPNs were placed in solution at pH 1.1 (KCl–HCl) and pH 7.4 (Na_2HPO_4–KH_2PO_4) at 37 °C. During swelling, gels were removed from the water bath at regular time intervals and after drying superficially with filter paper, and weighing, they were returned into the same swelling bath. The increase in radii of the cylindrical swollen gels was measured with a micrometer, and the swelling parameters and diffusion types were determined. The effects of temperature and pH on swelling behaviours of the IPNs were investigated by cycled equilibrium swelling, in which the gels were alternately swollen to their equilibrium swelling values for 24 h at 25 °C, and at 37 °C for 30 days.

Drug loading and release studies

Drug loading was carried out by adding 50 mg amoxicillin per gram of precursor IPN before polymerization/crosslinking reactions. After polymerization, the hydrogels were optically transparent indicating complete solubility of the drug in the polymer matrix. The in vitro release studies of the entrapped drug were carried out by placing the drug loaded IPN samples into 10 mL of pH 1.1 solution at 37 °C in a shaker. After 30 min, 3 mL of the drug-containing solution were taken out and the concentration was measured at λ_{max}=271 nm using a Shimadzu 160A UV-visible spectrophotometer. The release media were replenished periodically with fresh KCl–HCl solutions (10 mL). The dilution effects were taken into account for calculations of the released drug. The release studies were continued until no more change was observed in the absorbance value of the medium. The same release experiments were also carried out for pH 7.4 solution (Na_2HPO_4–KH_2PO_4) at 37 °C. The amount of released amoxicillin was quantified and its corresponding release graphs were constructed.

Results

Preparation of the IPNs

The formation of IPN hydrogels is schematized and presented in Scheme. Amine groups (-NH_2) of chitosan chains in the acidic environment react with the GA by iminization reaction and chitosan chains are crosslinked. PNIPAM chains are also crosslinked with bifunctional crosslinker (MBA) via conventional free radical polymerization. In this way, cylindrically shaped full-IPN structure was constructed (Scheme).

Kinetics swelling studies

It is important that the nature of the swelling behaviours of IPNs in body fluids be known for release studies of active drug substances from polymeric devices. With this in mind, time-dependent swelling studies of prepared IPNs were accomplished in synthetic physiological solutions by using HCl–NaOH and Na_2HPO_4–KH_2PO_4 in vitro at 25 °C. The swelling degree, *S*% is calculated from the following equation:

$$\%S = \frac{M_t - M_o}{M_o}\ x100 \qquad (1)$$

where M_o is the mass of dry gel at time 0, M_t is the mass of swollen gel at time t. Swelling curves of the hydrogels in simulated body solutions are shown in Figure 1 (a).

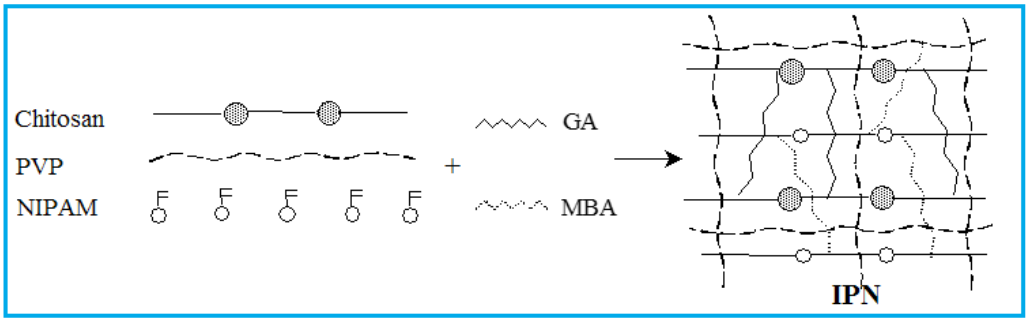
If Figure 1 (a) is investigated, it is shown that swelling is increased by time, but then it levels off. This value may be named equilibrium swelling (S_e%). A simple kinetic analysis is a second-order equation of the form;

$$\frac{t}{S} = A + Bt \qquad (2)$$

where *A* is reciprocal of initial swelling rate r_o or $1=S_{max}^2 k_s$ and *B* is inverse of the weight-swelling ratio at equilibrium. To test the kinetic model, *t/S* vs. *t* graphs were plotted and the graphs are shown in Figure 1 (b). The calculated kinetic parameters are tabulated in Table. The following equation is used to determine the nature of diffusion of water and drug molecules into the hydrogels;

$$F = \frac{M_t}{M_d} = kt^n \qquad (3)$$

where *F* is the fractional uptake at time *t*, *k* is a constant incorporating characteristics of the macromolecular network



Scheme: Schematic presentation of cylindrical shaped full-IPN preparation

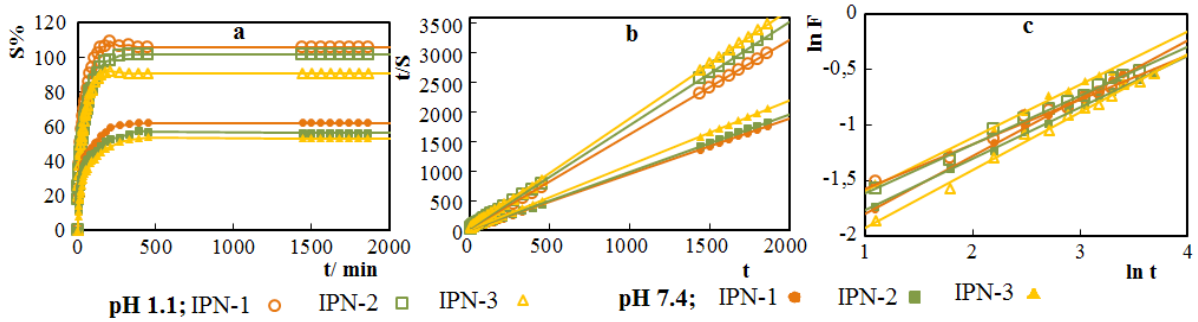


Figure 1: (a) Dynamic swelling curves, **(b)** t/S-t graphs and **(c)** ln F-ln t graphs of the IPNs

Table: Swelling and diffusion parameters of the hydrogels

Medium	Hydrogel	S_e%	S_{max}%	$k_s x10^5$	n	$kx10^5$	$Dx10^6$
pH 1.1	*IPN-1*	110	110	105	0.52	160	2.1
	IPN-2	100	100	76	0.46	180	0.7
	IPN-3	90	92	150	0.48	210	1.3
pH 7.4	*IPN-1*	60	60	125	0.40	230	0.4
	IPN-2	55	55	140	0.44	210	0.6
	IPN-3	50	50	120	0.52	145	0.8

system and the penetrant, and *n* is the diffusional exponent, which is indicative of the transport mechanism. Eq. (3) is valid for the first 60% of the fractional uptake. Fickian diffusion and Case II transport are defined by *n* at values of 0.5 and 1, respectively. Anomalous transport behavior (non-Fickian diffusion) is intermediate between Fickian and Case II. That is reflected at *n* between 0.5 and 1. When n=1, chain relaxation is slower than the liquid diffusion. A typical value of n is 0.5 for purely Fickian processes [23]. For the hydrogels, *lnF* vs. *lnt* graphs were plotted (Figure 1(c)). The *n* exponents and *k* parameters were calculated from the slopes and intercepts of the lines, respectively, and were listed in Table. The diffusion coefficients, *D* of the cylindrical hydrogels were found using the following equation:

$$k=4(D/\pi a^2)^n \quad (4)$$

where *D* is in cm^2 sec^{-1} and *r* is the radius of the cylindrical hydrogel sample. The diffusion coefficients were calculated from Eq. (4) and were listed in Table.

Table shows, the number determining type of diffusion (*n*), is genarally changed between 0.40 and 0.52 in both solutions. The mode of transport is near Fickian due to n values of the IPNs fall in the range 0.40–0.55. For the Fickian type of transport, the rate of diffusion of solvent molecules are much less than the relaxation rate of the polymer chains. It was observed that experimental S_e% values were close to calculated S_{max}% values as expected. IPNs exhibited greater swelling at pH 1.1 when compared with swelling at pH 7.4. This can be explained due to the protonation of $-NH_2$ groups of chitosan in stomach pH conditions. Strong electrostatic repulsion occurs among the polymer chains and the gel swells. Besides, while the concentration of crosslinker in the IPNs increased, the swelling degree of IPNs decreased. Also, network parameter, k, raised at pH 1.1. Diffusion coefficients, which means the gel area that solvent molecules cross per time unit, changed with increasing network density in the hydrogels and were not generally proportional to equilibrium swelling values (S_e%).

Enviromentally sensitive swelling studies of the IPNs

It is well known that PNIPAM in aqueous solution exhibits a sharp phase transition called the lower critical solution temperature (LCST) at temperature in the range of 25-32 °C depending on composition of solution. Below the LCST, this temperature-sensitive hydrogel are hydrated and hydrophilic in nature. Above the LCST, the chains become hydrophobic and weakly hydrogen-bonded with water molecules [24]. As shown in Figure 2, IPN-1 hydrogels exhibited phase transitions at both pHs with increasing temperature. Besides, graphs illustrate the classical temperature-dependent swelling behaviours of PNIPAM hydrogels at pH 1.1 and pH 7.4 when the temperature increased from 25 to 40 °C. It can be concluded that temperature affects the swelling degree of the IPN-1 with changes in pH. The shift of the LCST at pH 7.4 is due to less swelling of the IPN-1 as discussed before.

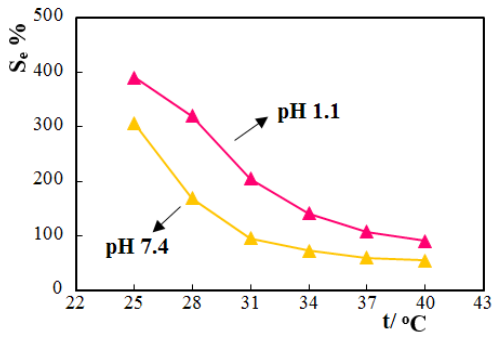


Figure 2: Phase transitions of IPN-1 hydrogels varying pH

In order to investigate the pulsatile swelling behavior of the IPNs, the equilibrium swelling studies were studied at 25 °C and 37 °C during 30 days (Figure 3). Mass measurements of the wet gels and changing of temperature were realized every day. All IPNs retained its shape and integrity during the whole period of the experiments. The graphs in Figure 3 showed that respond interval of the IPN hydrogels at pH 7.4 is much smaller than that of at pH 1.1. The swelling-deswelling cycle is reversible at both pHs and its profile not affected by crosslinker concentration except less declining of S_e% values. Similiar profiles for all IPN samples were observed at 25 °C and 37 °C.

Antibiotic release studies

The potential application of IPNs for drug delivery was investigated using amoxicillin trihydrate as a model drug molecule. The pH values were selected by considering an acid environment similar to stomach and a neutral environment similar to thin intestine in which amoxicillin is mostly absorbed. HCl-KCl (pH 1.1) and Na_2HPO_4-KH_2PO_4 (pH 7.4) solutions were used as simulated body fluids. The percentage cumulative release (*R%)* of amoxicillin from the IPNs was calculated using equation;

$$R\% = \frac{m_t}{m_o} x100 \quad (5)$$

where m_t is the amount of drug released at time t and m_o is the initial loaded drug amount. *R%-t* graphs were constructed and presented in Figure 4. Graphs show the second-order release profiles of model drug through the IPN hydrogels. Besides, it has been concluded that the amoxicillin trihydrate release from the IPNs is dependent on pH and temperature. IPN-1, IPN-2, and IPN-3 showed a final percentage of amoxicillin released at pH 1.1 of 57, 51, and 45, respectively. Whereas, in the studies conducted at pH 7.4 and 25 °C the final values were 39, 38, and 38. The lower R% values at pH 7.4 are probably largely contributed by the pore size decrease. These patterns bear a striking similarity to that for the IPNs shown in Figure 1(a) and suggests that the increase in drug mobility is directly related the increase in hydrogel pore size as pH decreases [25].

Meanwhile, the percentage of drug released increased when temperature was increased. For example, the release of drug from IPN-1 hydrogels was 82% at 37 °C for pH 1.1 while the

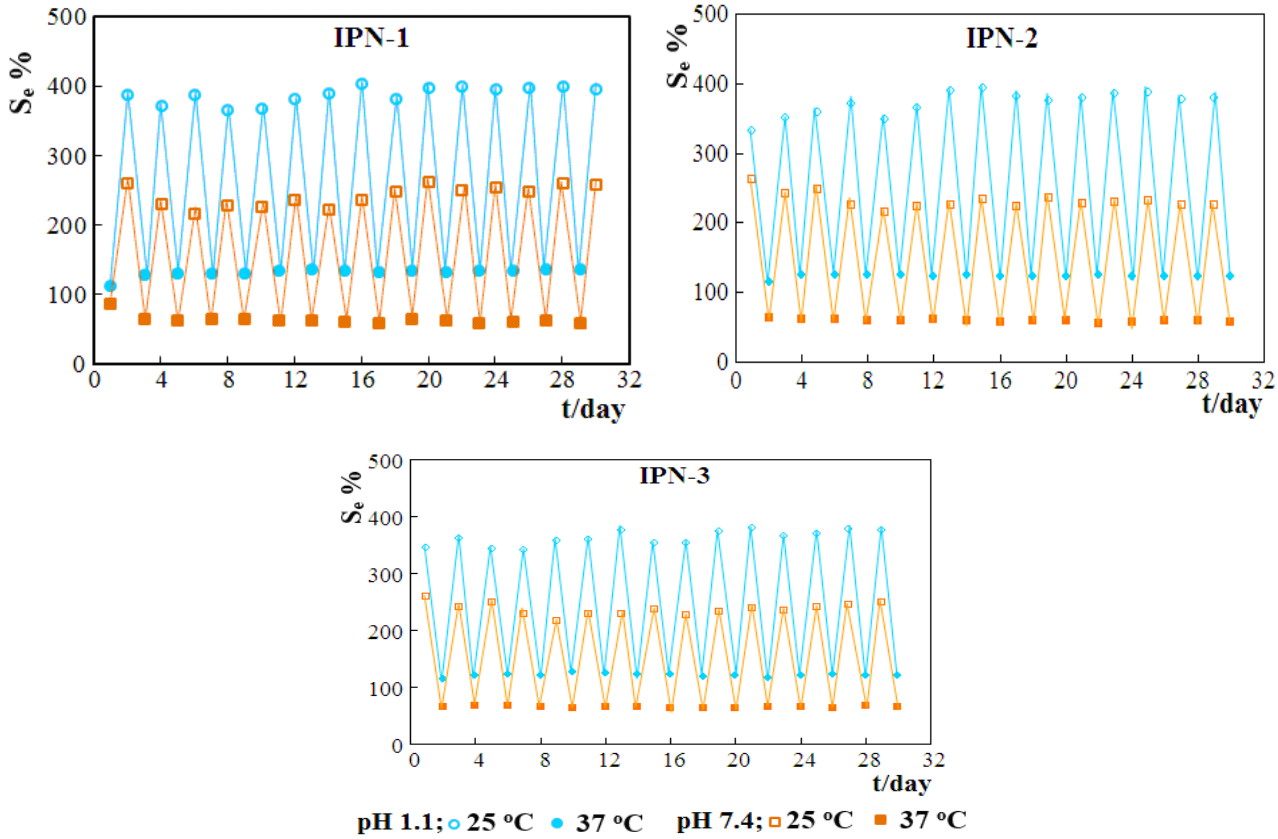


Figure 3: Pulsatile temperature-dependent swelling behavior of the IPNs

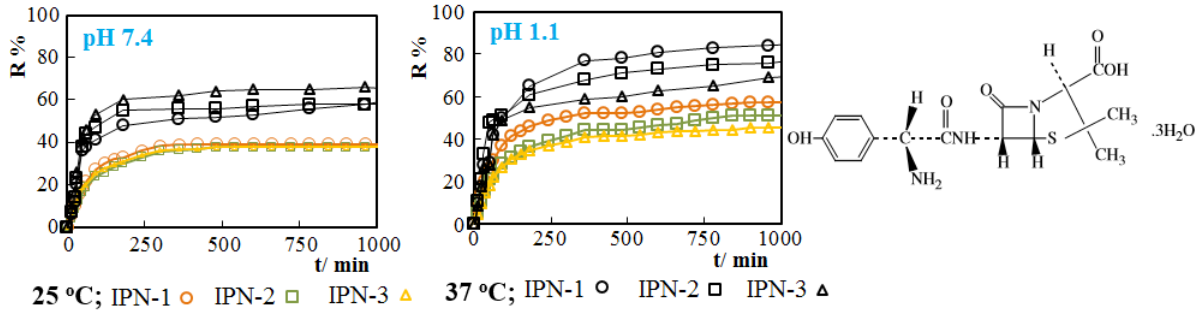


Figure 4: Release profiles of IPNs at different temperatures and pHs, and chemical structure of amoxicillin

R% value of the same sample was 60% at 25 °C. This behavior can be attributed to temperature-sensitive behaviour of PNIPAM in the IPNs which causes a decrease in the swelling of IPN hydrogel, which in turn favors the solute release. Also, the higher temperature makes the higher kinetic energy of amoxicillin molecules, which also favors its release from the hydrogel matrix. The drug release is further decreased as crosslinking increases with the consequence that the pore size of the hydrogel is further decreased as seen from Figure 4.

Conclusion

Ternary IPN systems were developed in the form of cylindrical by using three different hydrogel and crosslinker concentration. IPNs displayed remarkable good mechanical strength during all experiments, i. e., swelling and release studies. It was observed that the hydrophilic components, chitosan and PVP, did not change the LCST character of the PNIPAM. Deswelling temperature of the IPNs was determined as 37 °C, which is body temperature, and this behaviour facilitated the releasing of drug molecules from the hydrogels. IPNs showed second-order swelling and release kinetics. The swelling and release values were found higher in acidic solution than neutral solution due to the repulsion forces between amino groups of chitosan and hydronium ions in stomach pH conditions. Also, the release time of amoxicillin from the IPNs was about 7–11 h, signaling gastrointestinal transit time of oral dosage forms in human. Consequently, prepared IPNs may be suggested as model devices for gastrointestinal drug release studies.

References

1. Ranjha NM, Hanif M, Naz A, Shah MS, Abbas G, Afzal Z. J.Appl. Polym.Sci. 2016;43407.
2. Priya P, Raja A, Raj V. Cellulose. 2016; 23:699.
3. Aminabhavi TM, Nadagouda MN, More UA, et al. Expert Opin. Drug Del. 2015; 12:669.
4. Anirudhan TS, Rejeena SR. J.Appl.Polym.Sci. 2014; 131:40699.
5. Kosmala JD, Henthorn DB, Peppas LB. Biomaterials. 2000; 21:2019.
6. Chakrabarty D. Polym. Gels Networks. 1998; 6:191.
7. Ekici S, Saraydin D. Drug Deliv. 2004; 11:381.
8. Ekici S, Saraydin D. Polym. Int. 2007; 56:1371.
9. Khalid MN, Agnely F, Yagoubi N, Grossiord JL, Couarraze G. Eur. J. Pharm. Sci. 2002; 15:425.
10. Shityakov S, Puskas I, Papai K, Salvador E, Roewer N, Forster C, et al. Molecules. 2015; 20:10264.
11. Shityakov S, Salmas RE, Durdagi S, Roewer N, Forster C, Broscheit J. J. Mol. Struct. 2017; 1134:91.
12. Shityakov S, Puskas I, Papai K, Salvador E, Roewer N, Forster C, et al. Molecules. 2015; 20:10264.
13. Peppas NA, Bures P, Leobandung W, Ichikawa H. Eur. J. Pharm. Biopharm. 2000; 50:27.
14. Quimet MA, Fogaça R, Synder SS, Sathaye S. Macromol. Biosci. 2015; 15:342.
15. Zhang S, Alvarez DJ, Sofroniew MV, Deming TJ. Biomacromolecules. 2015; 16:1331.
16. Cross MC, Toomey RG, Gallant ND. Biomed. Mater. 2016; 11:022002.
17. Torre PM, Enobakhare Y, Torrado G, Torrado S. Biomaterials. 2003; 24:1499.
18. Baptista JGC, Rodrigues SPJ, Matsushita AFY, et al. J. Mol. Liq. 2016; 222:287.
19. Singh B, Sharma V. Polymer. 2016; 91:50.
20. Sabbagh F, Muhamad II. J. Taiwan Inst. Chem. Eng. 2017; 72: 182.
21. Gerola AP, Silva DC, Jesus S, et al. RSC Advan. 2015; 5:94519.
22. Singh B, Sharma V. Langmuir. 2014; 30:8580.
23. Peppas NA, Franson NM. J. Polym. Sci. 1983; 21:983.
24. Abdelghani H, Ravin N. Eur. Polym. J. 2007; 43:4344.
25. Das D, Das R, Ghosh P, Dhara S, Panda AB. Pal S. RSC Adv. 2013; 3:25340.