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Preparation and in vitro evaluation of xanthan gum facilitated superabsorbent polymeric microspheres

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ABSTRACT

Interpenetrating polymer network (IPN) hydrogel microspheres of xanthan gum (XG) based superabsorbent polymer (SAP) and poly(vinyl alcohol) (PVA) were prepared by water-in-oil (w/o) emulsion crosslinking method for sustained release of ciprofloxacin hydrochloride (CIPRO). The microspheres were prepared with various ratios of hydrolyzed SAP to PVA and extent of crosslinking density. The prepared microspheres with loose and rigid surfaces were evidenced by scanning electron microscope (SEM). Fourier transform infrared spectroscopy (FTIR) and X-ray diffraction (XRD) analysis confirmed the IPN formation. Differential scanning calorimetry (DSC) study was performed to understand the dispersion nature of drug after encapsulation. The in vitro drug release study was extensively evaluated depending on the process variables in both acidic and alkaline media. All the formulations exhibited satisfactory physicochemical and in vitro release characteristics. Release data indicated a non-Fickian trend of drug release from the formulations. Based on the results, this study suggest that CIPRO loaded IPN microspheres were suitable for sustained release application.

1. Introduction

Over the past decades, blends have been investigated to satisfy the need of specific sectors of polymer industry. Such polymeric blends showed superior performances over the conventional individual polymers and consequently, the range of applications have grown rapidly for such class of materials. In recent years, carbohydrate and biodegradable hydrophilic polymers have been extensively used to develop controlled release formulations of drugs having short plasma life. Among the various polymers employed, hydrophilic biopolymers are quite suitable because they are non-toxic and acceptable by the regulating authorities (Banerjee et al., 2013; Kulkarni & Sa, 2008a, 2008b). The importance of biocompatible and biodegradable hydrophilic polymers have wide applications in different fields such as polymer engineering, chemical engineering, pharmaceuticals, food and agriculture (Dong & Hoffman, 1991; Peppas, Bures, Leobandung, & Ichikawa, 2000; Siegel & Firestone, 1990) because of their propensity to combine with other polymers to form crosslinked three-dimensional interpenetrating polymer network (IPN) hydrogels that tend to swell in water or biological fluids. Such IPN hydrogel systems have been considered as a potential candidate to deliver bioactive molecules, particularly in controlled release applications to deliver the drugs at constant rate over an extended period of time. Therefore formation of IPN hydrogel appears to be a better approach (Kulkarni & Sa, 2008a, 2008b; Pan and Ragauskas, 2012). IPN hydrogel has more complicated network structures and possesses improved mechanical properties; in such systems, the extent of crosslinking can be monitored to control the drug release (Rokhade et al., 2006; Rudzinski et al., 2002).

Recently, lot of research work has been carried out to develop oral controlled release multiple unit dosage forms using hydrophilic polymers in the form of microspheres because they are becoming more popular than single unit dosage forms due to its several inherent advantages. The administration of drugs in the form of microspheres has received much attention because they avoid vagaries of gastric emptying and different transit rates through the gastro-intestinal tract, thereby releasing drugs more uniformly (Tamilvanan & Sa, 2000). This fact coupled with their ability to prolong the release of drugs has given impetus to the

Abbreviations: IPN, interpenetrating polymer network; SAP, superabsorbent polymer; PVA, poly (vinyl alcohol); w/o, water-in-oil; CIPRO, ciprofloxacin hydrochloride; FTIR, fourier transform infra-red; XRD, X-ray diffraction; DSC, differential scanning calorimetry; SEM, scanning electron microscopy; DEE, drug encapsulation efficiency; KBr, potassium bromide; UV–Vis, ultraviolet–visible; AA, acrylic acid; XG, xanthan gum; MBA, N,N′-methylenebisacrylamide; GA, glutaraldehyde; APS, ammonium persulfate.

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development of oral micro-particulate systems for drug delivery.

Xanthan gum (XG) is a high molecular weight, anionic extracellular polysaccharide that is produced by gram-negative bacterium *Xanthomonas campestris*. It is widely used in food, cosmetics and pharmaceuticals because of its encouraging reports on safety (Bhattacharya et al., 2012a; Bhattacharya, Ghosh, Banerjee, Chattopadhyay, & Ghosh, 2012). On the basis of short-term and long-term feeding studies, XG was cleared by the US Food and Drug Administration (FDA) in 1969 permitting its use in food products without any specific quantity limitations

Superabsorbent polymers (SAPs) are crosslinked hydrophilic networks that can absorb water in the amount from 10% up to thousands of times their dry weight. However high water solubility has limited their use as a drug carrier to a certain extent because of dissolution before the drug can be delivered. In order to overcome the above drawback, in our present work we have polymerized acrylic acid (AA) with hydrophilic polymer in presence of organic crosslinkers to form copolymers of tunable physicochemical properties (Kawaguchi, 2000). Superabsorbent polymer (SAP) was being prepared by introducing modified bentonite in poly (acrylic acid) solution through chemical crosslinking by polymerization technique using N,N′-methylene bisacrylamide as a crosslinker and ammonium persulfate as an initiator in a complete aqueous environment. Poly(vinyl alcohol) (PVA) is widely used as hydrophilic bio-polymer in controlled release drug delivery system because of its processibility, strength, and pH, as well as temperature stability and semi-crystalline nature. As it is biodegradable, biocompatible and non-toxic, it has a wide variety of pharmaceutical applications (More, Kulkarni, Sa, & Kayane, 2010).

Earlier, we have developed the poly (acrylic acid) based SAP (Bhattacharya, Ghosh et al., 2012). So as a part of our ongoing research work on the application of hydrophillic biopolymers, the present work describes the development and evaluation of IPN hydrogel microspheres composed of xanthan gum (XG) based superabsorbent polymer (SAP) and PVA to encapsulate ciprofloxacin hydrochloride (CIPRO), which is used as a model drug. SAP was hydrolyzed to increase its water solubility property so that the yield of microsphere can be enhanced. The microspheres prepared have been characterized by different analytical techniques to understand their various physicochemical behavior and in vitro drug release characteristics.

2. Experimental

2.1. Materials

Xanthan gum (XG, food grade) was obtained from Loba Chemie Private Limited, Mumbai, India. N,N′-methylenebisacrylamide (MBA, chemically pure) and Glutaraldehyde (GA: 25%, v/v) was purchased from Loba Chemie Private Limited, Mumbai, India. Acrylic acid (AA, analytical grade) was distilled under reduced pressure before use and was commercially purchased from Merck Specialties Private Limited, Mumbai, India. Bentonite and ciprofloxacin hydrochloride (CIPRO) was supplied by Central Drug House Private Limited, New Delhi, India. Ammonium persulfate (APS) was recrystallised twice by distilled water and supplied by Qualigens Fine Chemicals Private Limited, Mumbai, India. Sodium hydroxide (NaOH, analytical grade) was supplied from BDH chemicals, India. Poly(vinyl alcohol) (PVA: 98% hydrolyzed, average molecular weight 125,000), & light liquid paraffin (LLP, viscosity 25–80 mPa at 20 °C) were procured from Hi-Media Lab-oratories Private Limited, Mumbai, India. Span 80 was procured from Pioneer in-organics, Delhi, India. Acetone was bought from Qualigens fine chemicals, Mumbai, India. The water used was a high-purity grade after double distillation and deionization. All other reagents were of analytical grade and used as received without further purification.

2.2. Preparation of XG modified bentonite

0.5 g bentonite was added to 50 ml distilled water and mechanically stirred (Remi Equipments Private Limited, Mumbai, India) followed by ultrasonic treatment (FS-600, Frontline Electronics and Machinery Pvt. Ltd., Gujarat, India.) for 15 min, which resulted in good bentonite dispersion. The XG solution obtained by dissolving 1 g XG into 50 ml distilled water under vigorous stirring was added to the bentonite suspension and the mixture was stirred vigorously for 2 h at 70 °C temperature. The modified bentonite suspension was dried in an oven (Lunar Amalgamated Suppliers, Kolkata, India) at 70 °C temperature to constant weight and then milled through a 120 mesh screen.

2.3. Preparation of poly (acrylic acid)/modified bentonite superabsorbent base

In brief, AA 10 ml was dissolved in 10 ml distilled water and then neutralized in an ice bath with a predetermined amount of aqueous sodium hydroxide solution until the neutralization was completed in a 250 ml three-necked flask equipped with a stirrer, an efficient reflux condenser, and a thermometer. Then 1 g of modified bentonite was added to the above neutralized solution and stirred vigorously until the modified bentonite was well dispersed. Thereafter 20 mg of APS in 2 ml water was charged as initiator followed by 20 mg of MBA in 2 ml water as crosslinker into the reaction mixture dropwise. The mixture was stirred and heated to 70 °C in a heating mantle (Lunar amalgamated suppliers, Kolkata, India) for 4 h. After complete polymerization, the resulting product was washed with double distilled water and dried in an oven at 70 °C temperature for 24 h. The dried material was pulverized into particles and screened through 120 mesh screen.

2.4. Preparation of hydrolyzed superabsorbent polymer

1 g powdered SAP polymer was added to 100 ml of 0.1 N NaOH solution in a beaker and was heated to 100 °C in a heating mantle (Lunar amalgamated suppliers, Kolkata, India) for 1 h with occasional stirring. Then this hydrolyzed polymer was allowed to cool at room temperature and neutralized with 10% HCL solution. This neutralized polymer was cut into small pieces and was dried in an oven (Lunar amalgamated suppliers, Kolkata, India) at 60°C temperature to constant weight and then milled through a 100 mesh screen.

2.5. Development of IPN hydrogel microspheres

Microspheres of hydrolyzed SAP and PVA containing ciprofloxacin hydrochloride (CIPRO) were prepared by water-in-oil (w/o) emulsion-crosslinking method (Banerjee et al., 2012). In brief, PVA was dissolved in double distilled water heated at 80 °C by continuously stirring until a homogeneous solution was obtained. After that, hydrolyzed SAP was dispersed in PVA solution and stirred overnight with help of magnetic stirrer to obtain a homogeneous polymeric mass. Then, CIPRO was dissolved in water and dispersed in the above polymeric mass. This drug polymer mixture was added slowly to a light liquid paraffin (100 g, w/w) containing 1% (w/w) Span-80 under constant mechanical stirring at 800 rpm speed for 2 h to form water in oil (w/o) emulsion. To this w/o emulsion, 5 ml of concentrated H_2SO_4 was added slowly followed by GA and stirring was continued for 5 h in order to produce hardened microspheres. The hardened microspheres were then separated by filtration process and washed with acetone,

Table 1
Composition of drug loaded IPN hydrogel microspheres.

Sample code	PVA (g)	SAP (g)	Drug Loading (% w/w of total polymer)	GA (ml)
F_1	3	1	50	10
F_2	3	1	50	15
F_3	3	2	50	10
F_4	3	2	50	15
F_5	3	3	50	10
F_6	3	3	50	15

glycine and double distilled water to remove excess amount of oil droplets, unreacted GA and surfactant respectively. The complete removal of unreacted GA was confirmed by the aldehyde negative test of the washings with Fehling's reagent. Then the microspheres were dried at 40 °C for overnight and stored in desiccators until further use.

The similar method was adopted for the preparation of blank microspheres without the addition of pure drug.

2.6. Experimental design

In total, six formulations were prepared by varying two parameters, i.e., extent of crosslinkers concentration and ratios of hydrolyzed SAP and PVA. The assigned formulation codes were given in (Table 1).

2.7. Surface morphology analysis

The microspheres were deposited onto stubs using one side of a double-sided adhesive dried carbon tape (NEM Tape, Nissin Co., Tokyo, Japan) and sputter coated with platinum using a sputter coater (Edward S 150, UK). The coated microspheres were observed under SEM (JEOL, JSM-6360, Kyoto, Japan) at the required magnification at room temperature. The acceleration voltage used was 17 kV with the secondary electron image (SEI) as a detector.

2.8. Particle size analysis

The size of dried drug loaded IPN hydrogel microspheres was measured using an optical microscope (Olympus Model HB, India). A standard stage micrometer was used to calibrate the eye-piece micrometer. The mean diameter was calculated using 50 microspheres, randomly selected from each formulation.

2.9. Drug–polymer interaction study

FTIR spectral measurements were performed using FTIR Spectrophotometer (Model FTIR-8400s, Shimadzu, Japan) to confirm the formation of IPN structure, the presence of crosslinking agent in hydrolyzed SAP and PVA and also to find the chemical stability of the drug in the IPN hydrogel microspheres. FTIR spectra of the pristine drug, PVA, hydrolyzed SAP, placebo microspheres and drug-loaded microspheres were obtained. Each sample was grounded thoroughly with potassium bromide powder in a weight ratio of 1:19 under a hydraulic pressure (100 kg/cm^2) for 10 min. The pellet was placed in the sample holder and spectral scanning was taken in the wavelength region between 4000 and 400 cm^{-1} at a resolution of 4 cm^{-1} with scan speed of 1 cm/s.

2.10. Differential scanning calorimetric (DSC) study

DSC (Pyris Diamond, Perkin Elmer, Osaka, Japan) was performed on pristine drug, hydrolyzed SAP, PVA, placebo microspheres and drug-loaded microspheres. Samples were heated from 25 to 500 °C at the heating rate of 10 °C min^{-1} in nitrogen atmosphere (flow rate, 20 ml/min).

2.11. X-ray diffraction (X-RD) studies

Crystallinity of the drug after encapsulation was evaluated by X-ray diffraction (X-RD) measurements recorded for pristine drug, placebo microspheres and drug-loaded microspheres using X-ray diffractometer (X-Pert, Philips, UK). Scanning was done up to 2θ range of 0–50° using CuK– radiation source.

2.12. Estimation of percent yield values

The percent yield of microspheres calculated by considering total amounts of microspheres obtained. Theoretical weight was calculated while considering the weight of the drug and polymers employed during the preparation. The percent yield was calculated as follows (Banerjee et al., 2010):

$$\text{Yield } \% = \left[\frac{\text{amount of microspheres}}{\text{amount of drug} + \text{amount of polymer}}\right] \times 100 \quad (1)$$

2.13. Estimation of drug entrapment efficiency

The actual amount of CIPRO present in different formulations was estimated by crushing 10 mg of swollen microspheres in phosphate buffer (100 ml; pH 7.4) at 50 °C to extract the drug from formulations in water bath. The whole system was kept for 24 h. Then the solution was centrifuged to remove the suspended polymeric debris and the clear supernatant liquid was taken for the determination of drug content spectrophotometrically by using UV–vis spectrophotometer (UV-1800, Shimadzu, Japan) at a wavelength of 276 nm against appropriate blank. In order to maintain the accuracy, experiments were carried out in triplicate for all formulations to check its reproducibility. The percentage drug entrapment efficiency in microspheres was calculated as follows (Banerjee et al., 2012; Kajjri, Manjeshwar, & Aminabhavi, 2011):

$$\text{Drug encapsulation efficiency } (\%) = \left[\frac{\text{experimental drug content}}{\text{theoretical drug content}}\right] \times 100 \quad (2)$$

2.14. Percent equilibrium water uptake study

The pH-dependent equilibrium water uptake of blank microspheres was measured by immersing samples (10 mg) into both acidic and alkaline pH conditions (50 ml). In order to ensure complete equilibration, microspheres were allowed to swell completely for about 24 h to attain equilibrium at 37 °C temperature. The excess surface adhered liquid droplets of particles were removed by blotting with soft tissue papers without pressing hard and the swollen microspheres were weighed using single pan balance. The percentage equilibrium water uptake was calculated as follows (Banerjee et al., 2012):

$$\text{Equilibrium water uptake } (\%\ Q_{eq}) = \left[\frac{\text{mass of swollen microspheres} - \text{mass of dry microspheres}}{\text{mass of dry microspheres}}\right] \times 100 \quad (3)$$

2.15. In vitro drug release studies

The in vitro drug release from the IPN hydrogel microspheres were investigated both in acidic and alkaline dissolution media. These experiments were performed using a USP-II rotating paddle type dissolution test apparatus (Electro Lab TDT-68L, India) under sink conditions. A weighed quantity of each sample for all formulations were accurately weighed and added to different dissolution medium. At regular intervals of time, aliquot samples were withdrawn and replaced by an equal volume of fresh stock solution. The amount of drug released was analyzed using a double beam UV–vis spectrophotometer (UV-1800, Shimadzu) at a wavelength of 276 nm. From this, cumulative percentage drug release was calculated and plotted against the function of time to study the pattern of drug release (Ray et al., 2010).

2.16. Kinetics analysis of drug release

In order to understand the mechanism of drug release behavior of the crosslinked microspheres, the power law equation was fitted into the kinetic data up to 55% of drug release. Power law model known as Korsmeyer–Peppas equation (Korsmeyer & Peppas, 1981) can be expressed as:

$$\frac{M_t}{M_\infty} = Kt^n \quad (4)$$

Where M_t and M_∞ were respectively, the amount of drug released at time t and at infinite time, K represents a constant incorporating structural and geometrical character of the dosage form, and n values denote the diffusion exponent indicative of the mechanism of drug release (Ritger & Peppas, 1987).

3. Results and discussion

The various blend compositions of xanthan gum based hydrolyzed superabsorbent polymer with poly vinyl alcohol were crosslinked by GA to produce IPNs having a three dimensional structure to facilitate the entrapment of drug for oral controlled release application were prepared. When hydrolyzed SAP–PVA blend were cross-linked with GA, a bi-functional cross-linking agent, it forms an acetal ring between the hydroxyl groups of hydrolyzed SAP–PVA polymer strands and aldehyde groups of GA to produce IPN and making the network rigid and insoluble.

3.1. Surface morphology analysis

The microspheres produced were all spherical in nature, with smooth surfaces as revealed by SEM images shown in Fig. 1. The SEM micrograph of drug loaded microspheres showed a dependence on crosslinking density. The surface of a drug loaded microspheres matrix structure of F_1 (Fig. 1(a) and (b)) were held by loose polymeric network with undulant porous matrix structure; whereas the surface of a drug loaded microspheres matrix structure of F_2 (Fig. 1(c) and (d)) were held by dense rigid polymeric network, without showing any porous structure. This may be due to the shrinking of the matrix at higher extent of crosslinking density which influences rigid matrix structure with almost non-porous surface. Again SEM micrographs of the microspheres containing various amounts of hydrolyzed SAP exhibit different surface morphology. The surface morphology of F_1 differs from F_3. F_3 exhibited a smooth, dense and tight surface (Fig. 1(e)); however, the superabsorbent composites F_1 incorporated lower amounts of hydrolyzed SAP showed a relatively undulant, porous and coarse surface (Fig. 1(b)). This surface is convenient for the penetration of water into the polymeric network, which may be of benefit to the water absorbency of the corresponding microsphere.

3.2. Particle size analysis

Results of mean particle size of IPN microspheres were presented in Table 2. The arithmetic mean diameter of the microspheres ranges from 325 μm to 607 μm. These data showed a systematic dependence on the extent of crosslinker employed and amount of hydrolyzed SAP utilized. It was observed that particle size decreased with an increasing extent of crosslinking density. Particle size of F_1 was higher than that of F_2. This suggests that during crosslinking, the polymeric network might have undergone rapid shrinking leading to the formation of smaller and rigid matrix at higher crosslinking densities. Again by increasing hydrolyzed SAP content, size of the microspheres increased from 475 to 607 μm when drug loading was kept constant. This can be explained on the basis of hydrodynamic viscosity concept, i.e., as the amount of hydrolyzed SAP increases, interfacial viscosity of the polymer droplets in the emulsion also increases and the number of free sites available for cross-linking was less so that size of the microsphere will also increase.

3.3. Drug–polymer interaction study

FTIR spectral analysis was studied to investigate the formation of IPN structure, presence of crosslinking agent and chemical stability of the drug after encapsulation into the IPN matrix. FTIR-spectral analysis of pure drug (CIPRO), PVA, hydrolyzed SAP, blank microspheres and drug-loaded microspheres were shown in Fig. 2(a)–(e), respectively. In the FTIR spectral analysis of pure drug peak at 3375 cm^{-1} was due to N—H stretching mode. The peak at 3050 cm^{-1} was attributed to the presence of C-H alkene functional group. A characteristic peak due to the presence of cyclopropyl group was found at 2900 cm^{-1}. Band at 1500–1600 cm^{-1} was contributed due to the presence of C=C aromatic ring. C—F stretching and OH bending was observed at 1025 cm^{-1} and 945 cm^{-1} respectively. The FTIR spectra of PVA showed a broad peak around 2350 cm^{-1}, indicating stretching of hydroxyl groups and peaks at 2925 cm^{-1} is attributed to the stretching vibration of $-CH_2$. The band at 1097 cm^{-1} indicates the C—O stretching vibration. The FTIR spectra of hydrolyzed SAP revealed C—H asymmetrical stretching and C—H symmetrical stretching at around 2926 cm^{-1} and 2853 cm^{-1} respectively. C—O stretching was confirmed by the presence of peak at 1050 cm^{-1} and the peak at 650 cm^{-1} was attributed due to the presence of C—Cl stretching. Crosslinked polymeric IPNs of hydrolyzed SAP and PVA were washed repeatedly with acetone followed by water to remove the unbound PVA from the matrix. In blank microsphere peaks due to stretching of hydroxyl groups of PVA in the IPN network confirms the incorporation of PVA in the matrix structure. All peaks appeared in CIPRO were observed in the drug loaded microsphere, indicating the chemical stability of drug after encapsulation in the polymeric matrix. During crosslinking GA might have reacted with the hydroxyl groups of polymers through the formation of ether linkages. FTIR spectra of the cross linked blank microspheres and drug-loaded microspheres showed sharp peak intensity near about at 1151.42 cm^{-1} and 1045.35 cm^{-1} respectively in the spectra confirming the presence of an ether of acetal group, which was formed due to the reaction of GA with hydroxyl groups present in both hydrolyzed SAP and PVA. Thus, FTIR confirms the crosslinking reaction of GA with hydrolyzed SAP and PVA.

3.4. Differential scanning calorimetric (DSC) study

DSC thermograms of pristine drug, hydrolyzed SAP, PVA, blank microsphere, drug loaded microspheres were displayed in Fig. 3(a)–(e) respectively. The thermal behavior of CIPRO exhibits two endothermic peaks (Fig. 3(a)). First peak is at 140 °C, which shows water evaporation and the sharp endothermic

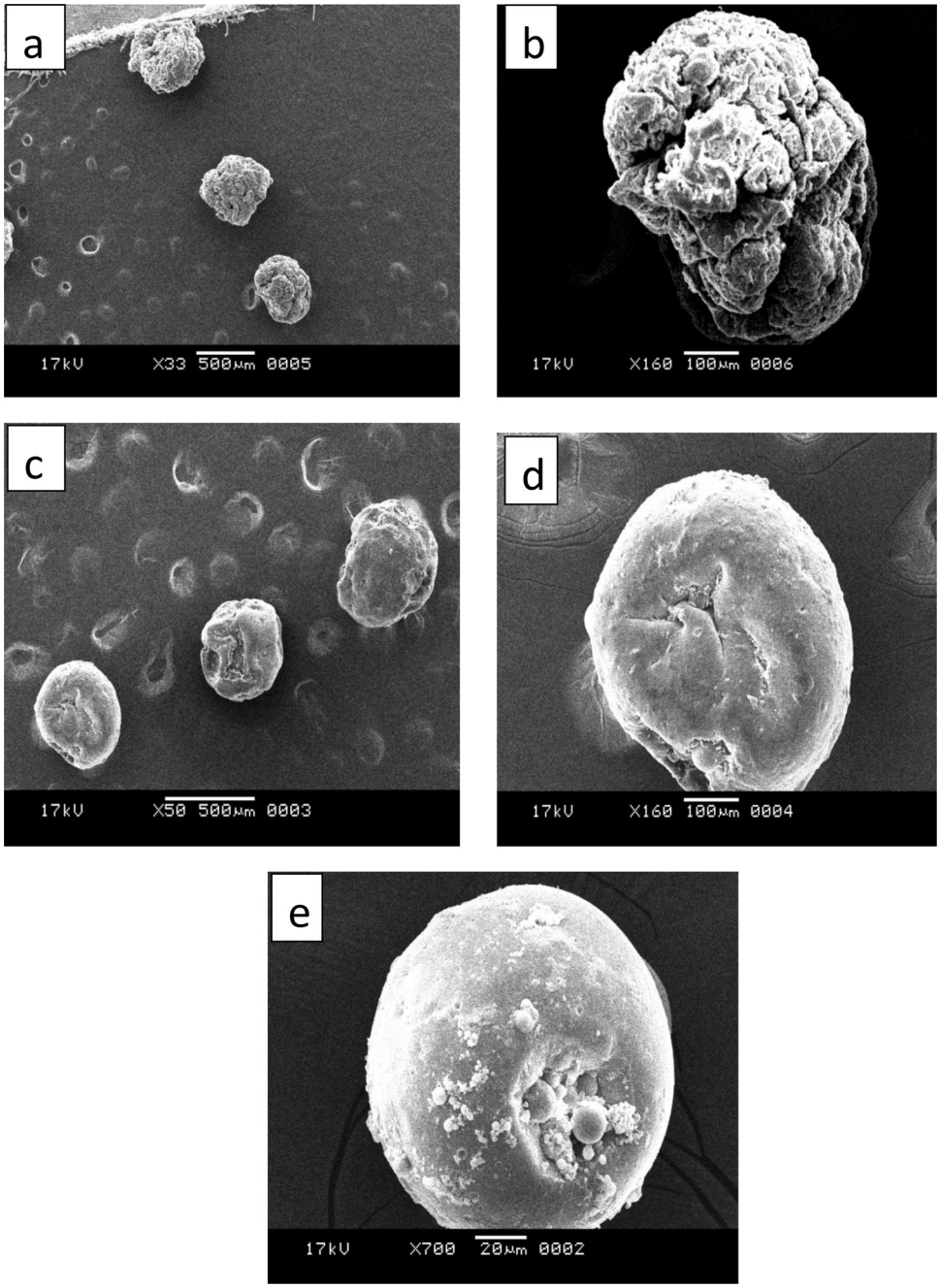

Fig. 1. Scanning electron micrographs of drug-loaded IPN microspheres of F_1 (a and b), F_2 (c and d), F_3 (e).

Table 2
Effect of processing variable on % yield, % drug entrapment efficiency (DEE), particle size, % equilibrium water uptake and n value of drug loaded IPN hydrogel microspheres.

Assigned formulation code	Mean particle size (μm)	% Yield value	%DEE (±S.D, $n = 3$)	% Equillibrium water uptake		Koresmeyer-peppas equation		
				pH 1.2	pH 7.4	n	k	r^2
F_1	510 ± 7	51.49	60.5 ± 2.2	175.28	221.47	0.357	0.352	0.988
F_2	325 ± 6	54.28	65.7 ± 1.8	155.22	207.81	0.368	0.385	0.985
F_3	560 ± 6	58.21	51.9 ± 1.2	212.39	307.78	0.344	0.361	0.992
F_4	370 ± 5	64.07	57.4 ± 2.4	190.72	272.89	0.362	0.372	0.997
F_5	607 ± 4	68.15	37.5 ± 2.1	290.17	380.27	0.332	0.337	0.979
F_6	475 ± 8	75.27	40.6 ± 1.5	235.46	342.51	0.339	0.325	0.981

Note: n values indicate the release mechanism; r^2 values indicate correlation coefficient.

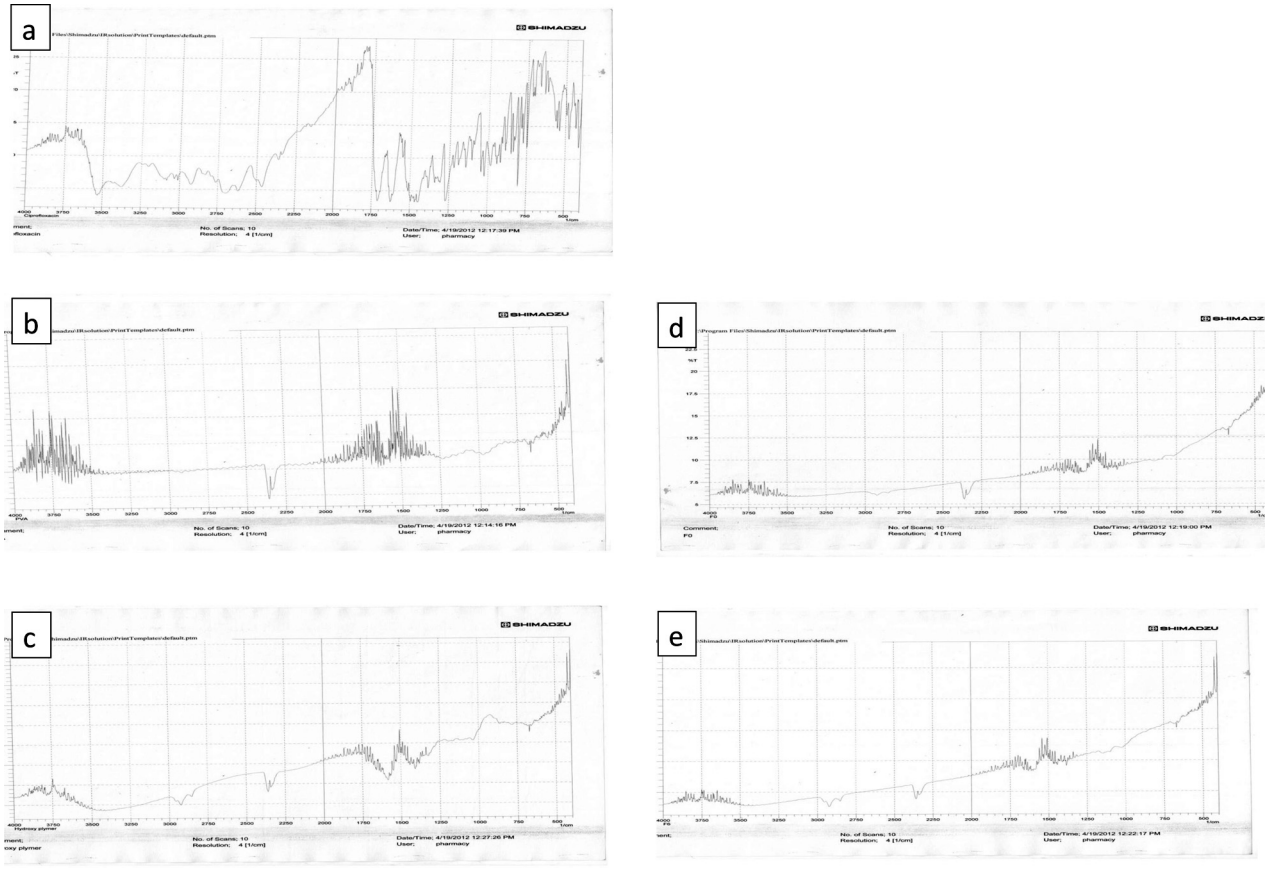

Fig. 2. FTIR spectra of (a) pristine drug, (b) PVA, (C) hydrolyzed SAP, (d) blank microspheres and (e) drug-loaded microspheres.

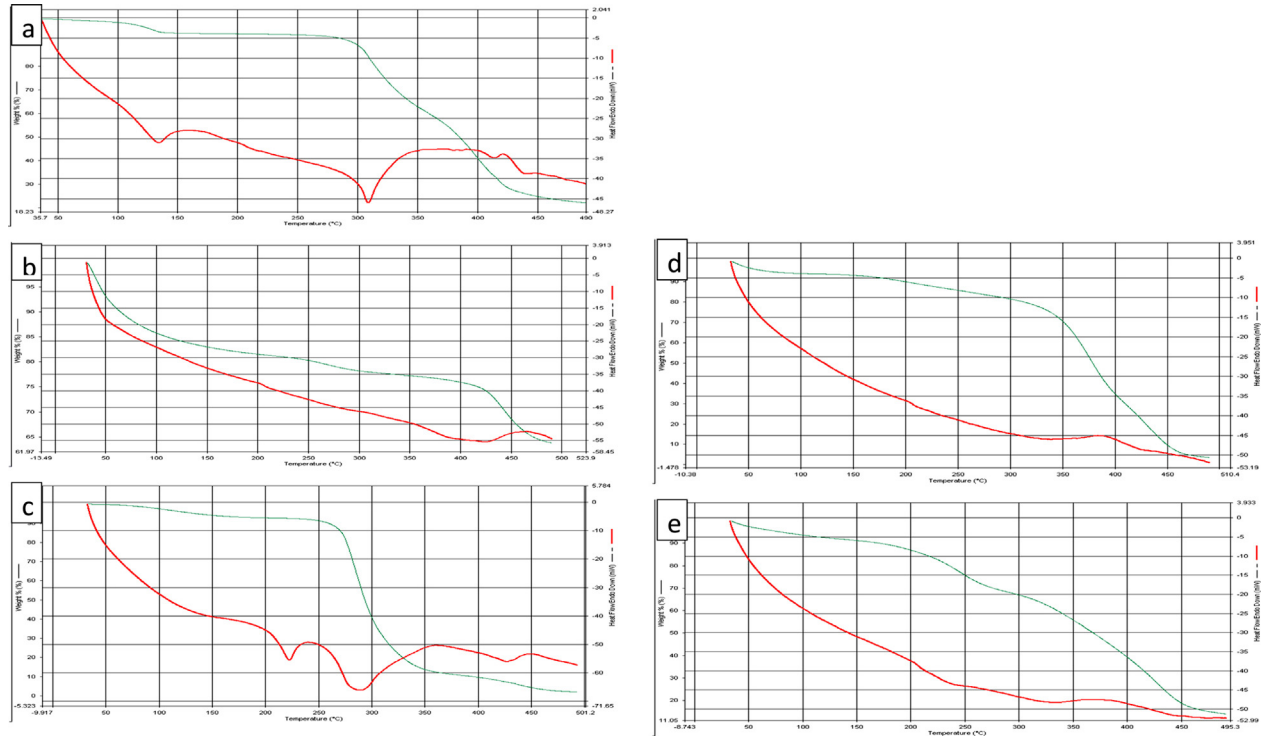

Fig. 3. DSC thermograms of (a) pristine drug, (b) hydrolyzed SAP, (c) PVA, (d) blank microsphere and (e) drug-loaded microspheres.

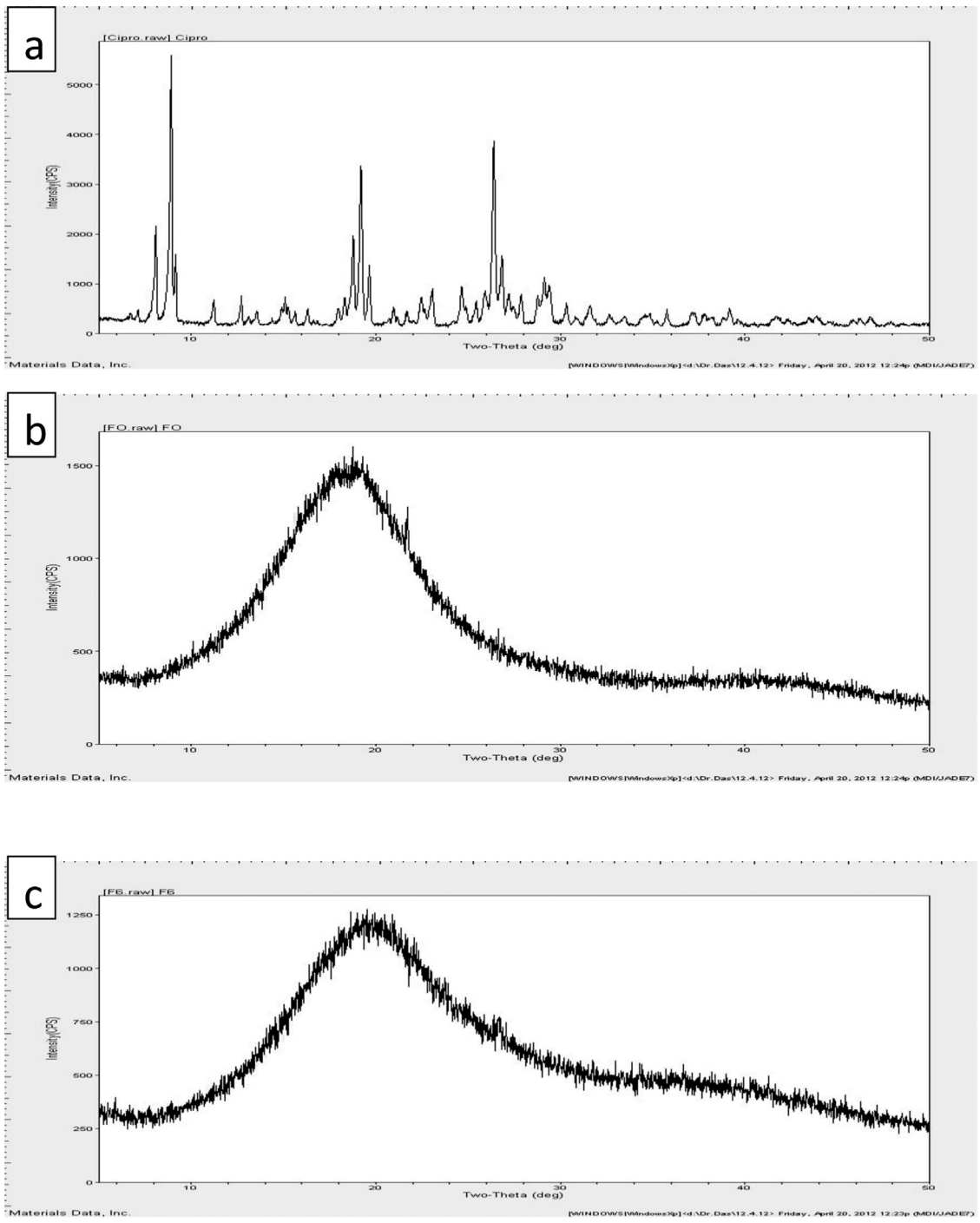

Fig. 4. X-ray diffraction pattern of (a) pristine drug (b) blank microsphere and (c) drug-loaded microspheres.

peak at 315 °C corresponds to its melting point. The decomposition process followed the melting of the drug. Hydrolyzed polymer showed no characteristic melting peak which indicates the amorphous nature of the polymer (Fig. 3(b)). PVA exhibits two characteristic endothermic peaks at around 224 °C and 285 °C (Fig. 3(c)) due to loss of water on evaporation. In case of blank microsphere no sharp peak were observed due to endothermic transmission of the polymeric matrix and formation of rigid IPN matrix structure as because of chain entanglements and formation of a homogeneous matrix structure (Fig. 3(d)).However drug loaded IPN microsphere exhibits two broad endothermic peaks at around 240 °C (Fig. 3(e)), probably due PVA inducing water evaporation phase and a weak endothermic peak at 325 °C corresponding to the melting point of pristine CIPRO.

3.5. X-ray diffraction (X-RD) studies

XRD patterns recorded for pristine drug, blank microsphere and drug loaded microspheres are presented in Fig. 4(a)–(c). XRD pattern of CIPRO showed the important crystallographic reflection at different scattering angle ranges from 6° to 27° to the interplanner distances at 2θ were due to the crystalline nature of CIPRO (Fig. 4(a)). However, these signal intensities were very weak for

blank microsphere and drug-loaded microsphere (Fig. 4(b) and (c)). Thus, the X-ray crystallographic data indicates that the relative degree of crystallinity (RDC) value was found to be 16.77%, thereby indicating the change of drug from its crystalline to amorphous state, where most of the drug dispersed at molecular level in the polymer matrices since no indication about the crystalline nature of the drugs was observed in case of drug-loaded IPN microspheres.

3.6. Estimation of percent yield values

The yield value of IPN hydrogel microspheres was improved from 51.49% to 75.27% (Table 2). The impact of crosslinker content during the production of drug-loaded microspheres was studied. The change in GA content from 10 ml to 15 ml was associated with increase in percentage yield of microspheres due to higher extent of crosslinking, which numerously breaks the polymeric chains producing higher quantity of microspheres. Again as the hydrolyzed SAP content of the microspheres increases, the yield of microspheres was improved from 64.07% to 75.27% (Table 2). This could be due to when the concentration of the polymer was increased the viscosity in the polymeric solution was also increased, thereby producing bigger droplets during emulsification which ultimately increases the quantitative weight of the formulations.

3.7. Estimation of drug entrapment efficiency

The percent encapsulation efficiency was found to be in the range between 37.5% and 65.7% (Table 2). The percent encapsulation efficiency showed a dependence on the extent of crosslinker content and hydrolyzed SAP concentration. The effect of crosslinking on percent encapsulation efficiency showed a significant effect. As the concentration of crosslinker content was increased, an increase in percent encapsulation efficiency was observed. For instance, F_1 containing crosslinker content 10 ml, encapsulation efficiencies were decreased, in comparison with microsphere crosslinked with 15 ml of GA i.e. F_2. This could be due to higher extent of crosslinking, resulting in the formation of a more rigid network structure, which reduces the possibility of leaching out of the drug during the microsphere preparation. So, ultimately it causes retention of more drug particles into the core of IPN matrix structure during the microsphere preparation. Results of % encapsulation efficiency included in Table 2 show decreasing trends with increasing amount of hydrolyzed SAP. Encapsulation efficiency of 65.7% was observed for F_2 but for F_6, it was 40.6%. Such smaller values may be due to the incorporation of a higher amount of hydrolyzed SAP into microspheres leading to lower encapsulation efficiencies due to the formation of a loose network that allows the leaching out of more of drug particles during the production stage of the microspheres (Banerjee et al., 2012).

3.8. Percent equilibrium water uptake study

The percentage equilibrium water uptake data of the crosslinked microspheres (Table 2) indicate that as the amount of GA in matrices increases, the equilibrium water uptake in both acidic and alkaline media decreases significantly, which is evident from the enhanced swelling behavior of formulation F_1 (175.28%, 221.47%)in comparison to F_2 (155.22%, 207.81%). The reduction in water uptake capacity was due to the formation of a rigid network structure at the higher concentration of cross-linking which ultimately reduces the pore volume of IPN matrix. Whereas it was found that formulations containing higher amounts of hydrolyzed SAP showed higher percentages of equilibrium water uptake than formulations containing small amounts of hydrolyzed SAP. Formulation F_5 showed higher water uptake capacity than F_1 due to the hydrophilic nature of hydrolyzed SAP, thereby leading to higher water uptake capacity.

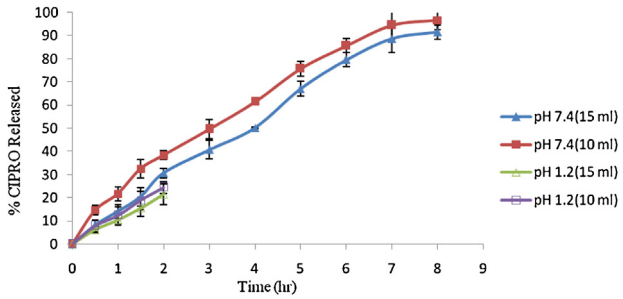

Fig. 5. Release profiles of drug in acidic solution (open symbols) and phosphate buffer solution (closed symbols) from IPN microspheres prepared by variation in the concentration of GA (10–15 ml) at a fixed ratio of PVA: hydrolyzed SAP (3:3) and CIPRO (50%). Key: GA = (□) 10 ml, (△) 15 ml.

3.9. In vitro drug release studies

In vitro drug release was performed in pH 1.2 and pH 7.4 and percentage cumulative release versus time data were presented in Fig. 5 to investigate the extent of crosslinking on the in vitro drug release profiles. The formulation containing lower amount of glutaraldehyde showed a higher release rate than higher amount of glutaraldehyde and release was slower for those formulations in which a higher amount of glutaraldehyde was used compared to those where lower glutaraldehyde was used. This confirms the formation of a denser network structure, which reduces the rate of swelling as well as the rate of drug release from the matrix. The cumulative percentage release vs time for the microspheres prepared with different ratios of PVA: hydrolyzed SAP loaded with CIPRO were presented in Fig. 6. The cumulative percentage released was higher in the case of F_6 than F_2. This indicates that with the increase in the hydrolyzed SAP in the matrix, swelling of the matrix increases due to the hydrophilic nature of polymer, which leads to the higher release of CIPRO from the matrix. This sort of release behavior was previously studied by us.

3.10. Kinetics analysis of drug release

The release data were further analyzed by Korsmeyer–Peppas equation by using the least-squares procedure at the 95% confidence limit. The calculated n and k values for all formulations, and these data along with the correlation coefficients (r^2) were included in Table 2. The values of n and k were directly proportional to the extent of crosslinking and inversely proportional with the increase in hydrolyzed SAP content in the IPN matrix. The lower n value indicates the formation of a loosely crosslinked polymer network,

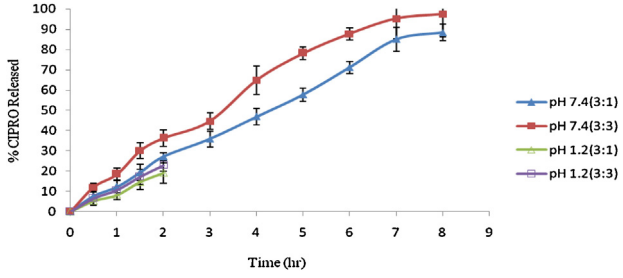

Fig. 6. Release profiles of drug in acidic solution (open symbols) and phosphate buffer solution (closed symbols) from IPN microspheres prepared by variation in PVA: hydrolyzed SAP ratio at a fixed CIPRO loading (50%) and in 15 ml GA concentration. Key: PVA: hydrolyzed SAP = (△) 3:1, (□) 3:3.

which leads to increased swelling. It was evident from (Table 2) that the correlation coefficient values approached unity, suggesting best fit to the non-Fickian model.

4. Conclusion

Two hydrophillic polymers, viz., xanthan gum based superabsorbent polymer and poly vinyl alcohol have been chosen to develop interpenetrating polymeric network hydrogel microspheres for sustaining the release of CIPRO. FTIR and XRD analysis were used to confirm the formation of IPN matrix structure. Microspheres with a narrow size distribution of sizes ranging from 325 to 607 μm. Swelling kinetics was investigated in terms of the extent of crosslinking density and the amount of xanthan gum based superabsorbent polymer used. Extent of crosslinking density and xanthan gum based superabsorbent polymer content into IPN matrix influenced the release of CIPRO. Release mechanism followed a non-Fickian type behavior. It is demonstrated that microspheres of this study were useful as controlled release devices to control the release rates of CIPRO through the polymeric matrices developed.

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