Sorption and Drug Release Studies from Semi-interpenetrating Polymer Networks of Chitosan and Xanthan Gum

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Hydrogel films of Chitosan (CS) and Xanthan gum (XA) of compositions 100/0, 90/10, 80/20, 70/30, 60/40 and 50/50 (w/w) % were prepared and swollen in simulated gastric fluid (SGF) of pH 1.2 and simulated intestinal fluid (SIF) of pH 7.4. To impart stability in acidic environment, semi-interpenetrating polymer network (semi-IPNs) films were formed using glutaraldehyde (GA) as the crosslinking agent. With increase in XA concentration, equilibrium degree of swelling reduced in SGF as well as SIF indicating maximum intermolecular interactions for 50/50 CS/XA semi-IPN. The swelling data was observed to follow second order kinetics. Spectroscopic and thermal analyses of these semi-IPN films also suggest maximum intermolecular interactions for 50/50 CS/XA semi-IPN. The potential of using 50/50 semi-IPN in drug delivery was studied using amoxicillin. *In-vitro* drug release studies indicated higher drug release in SGF than in SIF suggesting dependence of amoxicillin release kinetics and diffusion coefficient on pH of the environment and drug loading. The results suggest that CS-based semi-IPNs with different crosslinker and XA concentration could be promising candidates for formulation in oral gastrointestinal delivery systems.

Key words:
Amoxicillin, chitosan, glutaraldehyde, hydrogels

Introduction

Hydrogels are crosslinked three-dimensional hydrophilic polymer networks capable of retaining a significant amount of water within their structures, and swell without dissolving in water.[1] A hydrogel is formed when an organic polymer (natural or synthetic) is crosslinked via covalent, ionic, or hydrogen bonds to create a three-dimensional structure.[2] Recently, much work has been carried out on the synthesis and characterization of pH- and temperature-sensitive hydrogels by copolymerization and crosslinking. Interpenetrating polymer networks (IPNs) have also been synthesized and used in a wide range of applications including artificial implants, dialysis membranes, wound dressings, etc., suggesting their enormous potential as drug delivery systems. IPNs are defined as a combination of two or more polymers, each in a network form, at least one of which is synthesized and/or crosslinked in the immediate presence of the other. Several reviews have been published describing both applications and fundamental theory of IPNs.[3]

In the recent past, there has been considerable interest in developing controlled drug delivery systems using natural polymers due to their non-toxicity, biodegradability and biocompatibility. Chitosan (CS) is a linear polysaccharide composed of randomly distributed β-(1–4)-linked D-glucosamine (deacetylated unit) and N-acetyl-D-glucosamine (acetylated unit) (Fig. 1).[4] It is produced commercially by deacetylation of chitin, the structural element in the exoskeleton of crustaceans (crabs, shrimp, etc.).[5] CS is widely used in biomedical and pharmaceutical applications because of its unique chemical and biologic properties. It is a cationic polysaccharide (pKa ~6.3) soluble at acidic pH, and bioadhesive, which increases retention at the site of application and readily binds to negatively charged surfaces such as mucosal membranes. Mucoadhesive formulations have been developed for ocular, nasal, buccal, gastrointestinal, and vaginal drug ad-

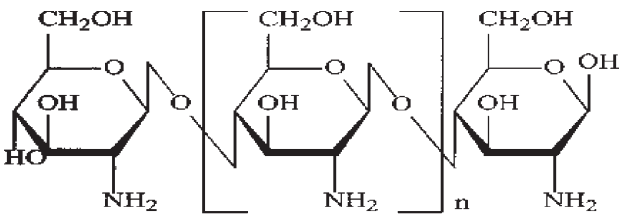

Fig. 1 – *Structure of Chitosan (adapted from J. H. Hamman, "Chitosan based polyelectrolyte complexes as potential carrier materials in drug delivery systems,"[4])*

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ministration. [6] Biodegradability and biocompatibility of CS has been exploited for use in controlled drug delivery systems.[7]

Xanthan gum (XA) is another extensively investigated water-soluble heteropolysaccharide natural polymer (Fig. 2). It is an anionic polymer with high molecular weight (1–2 million), produced by pure culture fermentation of a carbohydrate by naturally occurring bacterium *Xanthomonas compestris*,[8,9] and is commonly used as viscosity controller in food industries due to its high viscosity in aqueous media. XA has received considerable attention in the medical field as it has been found to retard drug release.[10,11]

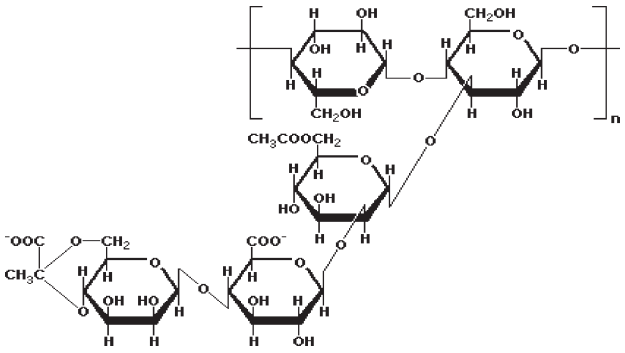

Fig. 2 – *Structure of Xanthan gum (adapted from A. Lachke, Xanthan – A Versatile Gum.pdf[8])*

Since CS and XA are oppositely charged, they are capable of forming polyelectrolyte complexes through electrostatic forces of attraction. Other secondary forces like hydrogen bonding, hydrophobic interactions, Van der Waals forces can also play a role in the formation of complexes. As polyelectrolyte complex hydrogel films are pH sensitive, it was proposed to prepare and study these for potential application as a drug delivery system. The aim of this work was to develop biopolymer hydrogel films based on CS and XA that could be used under varied pH conditions of gastrointestinal tract. Water absorption capacity and pH stability are important properties to be considered for their use as a drug delivery system in addition to other relevant properties. The hydrogel films were investigated for their swelling behavior in SGF and SIF as a function of XA and GA concentration. Additionally, release of model drug amoxicillin from hydrogel films was studied in simulated gastric and intestinal fluid.

Experimental

Material used

Chitosan (low viscous) was purchased from Fluka Biochemika. Xanthan gum (pure, food grade) was purchased from Loba Chemie. Amoxicillin was kindly gifted by Ind-Swift Drugs Ltd, Parwanoo, India. Glutaraldehyde (25 %, w/v) and glacial acetic acid were purchased from Loba Chemie and hydrochloric acid (35 %, w/v) was procured from Qualigens. Disodium dihydrogen phosphate, sodium hydroxide and potassium chloride were of analytical grade.

Characterization of chitosan

The degree of deacetylation (DDA) of CS was calculated by the method described by Alovarez[12] using (1), from the carbon (C) and nitrogen content (N) of CS, which were determined using a Perkin-Elmer Elemental Analyzer:

$$DDA = \frac{6.861 - (C/N)}{6.861 - 5.145} \cdot 100 \qquad (1)$$

Determination of molecular weight of chitosan

Intrinsic viscosity of chitosan in 0.2 mol L^{-1} NaCl/0.1 mol L^{-1} CH_3COOH was measured using an Ubbelohde capillary viscometer in a constant temperature water bath (model CT 1450, Schott Gerate, Germany) at 25 ± 0.1°C in triplicate. Solution concentrations were adjusted based on the viscosity of the samples, so that the flow time was kept in the range of 100–150 s. The intrinsic viscosity [η] was determined by the common intercept of Huggins equation (η_{sp}/C vs C) (2) and Kraemer's equation ($\ln \eta_r/C$ vs C) (3) on the ordinate at infinite dilution ($C \rightarrow 0$).[13]

$$\frac{\eta_{sp}}{C} = [\eta] + k'[\eta]^2 C \qquad (2)$$

$$\frac{\ln \eta_r}{C} = [\eta] - k''[\eta]^2 C \qquad (3)$$

The viscosity-average molecular weight of chitosan was calculated using the classical Mark-Houwink eq. (4):[13]

$$[\eta] = K_m M^a \qquad (4)$$

where $[\eta]$ is the intrinsic viscosity of chitosan, $K_m = 1.81 \cdot 10^{-3}$ and $a = 0.93$ are constants for a given solute–solvent system and temperature.[12,13]

Synthesis of physically crosslinked hydrogel films

Chitosan films were cast from 2 % (w/v) CS solution prepared in 0.3 mol L^{-1} acetic acid on a petri dish ($\Phi = 90$ mm) and allowed to dry at 37 °C. The film was further dried in vacuum oven (20 in. Hg) to remove traces of moisture for about 24 h at 40 °C and stored in a desiccator. For CS/XA blends, 2 % (w/v) solution was prepared with CS and XA in varying weight ratios of 100/0, 90/10, 80/20,

70/30, 60/40 and 50/50. XA solution was made in distilled water, while CS solution was prepared in 0.3 mol L^{-1} acetic acid. CS solution was added to XA solution in appropriate amounts resulting in the formation of a non-homogeneous solution containing suspended particles of the polymer. This solution was stirred continuously for 72 h on a magnetic stirrer to obtain a homogeneous solution and then cast on a petri dish.

Synthesis of chemically crosslinked semi-IPN hydrogel films

Chemically crosslinked CS[25] and CS/XA hydrogel films were prepared by adding 5 % (v/v) GA solution to homogeneous polymer solution prepared in the above section. To study the effect of crosslinker concentration on swelling behavior, varying amounts of crosslinker were used *i.e.* 0.1, 0.2, 0.3 and 0.4 mL of 5 % v/v GA.

Synthesis of drug loaded hydrogel films

The incorporation of the active agent into the semi-IPN hydrogel was done by adding the active agent during the synthesis process. The drug was added to the homogeneous CS/XA solution to which the required amount of GA was added for crosslinking. After crosslinking, a semi-IPN structure is formed and the drug gets entrapped in the polymer structure. When the hydrogel is placed in the medium of study, its swelling causes the mesh size to increase, which results in the diffusion of drug out of the system. In this study, the active agent amoxicillin was dissolved in the polymer solution at different drug loadings of 50 %, 75 % and 100 % (mg of drug/g of polymer). Casting of the films followed the same procedure.

Swelling studies

Swelling behavior of hydrogel films was investigated by placing 1cm × 1cm samples in SGF (pH 1.2) and SIF (pH 7.4). The dry samples were weighed using Sartorius weighing balance with an accuracy of ± 0.0001 g. Dry samples were placed in 20 mL of buffer solution at 37 °C and carefully taken out from the solution at various time intervals, placed on filter paper and blotted superficially to remove the free solvent on the surface of the film, and then weighed. After weighing, the samples were placed in the same media. The process continued until no further change was observed in the weight of the swollen film, which was taken as the equilibrium state. The degree of swelling (S) was determined using (5):[14]

$$\text{Degree of swelling, } S = \frac{W_w - W_{di}}{W_{di}} \qquad (5)$$

where W_w is the weight of swollen sample at time t, and W_{di} is the initial dry weight of the sample. When a hydrogel sample reached its equilibrium state under a fixed condition, its degree of swelling was referred to as equilibrium degree of swelling. The equilibrium state was achieved in 5 h for all the samples. All measurements were replicated three times for each sample.

Fourier transform infrared spectroscopy

Spectroscopic structural elucidation of semi-IPN films was done by FTIR using spectrophotometer (Bruker, model Tensor 27). The transmission spectra were collected at a resolution of 4 cm^{-1}, in the range of 4000–500 cm^{-1}.

X-Ray diffraction

X-ray diffraction patterns were measured using a wide-angle X-ray XPERT-PRO diffractometer system in the range $2\theta = 4°$ to 40°. X-ray diffraction patterns of samples were obtained at room temperature, with Cu as anode material, operated at a voltage of 45 kV. The process parameters were set at scan step size of 0.0170° (2θ) and scan step time of 25.1978 s.

Thermogravimetric analysis

The thermogravimetric analysis (TGA) of samples was conducted on Perkin Elmer STA 6000 under nitrogen atmosphere from 40 to 550 °C at a heating rate of 10 °C min^{-1}.

In vitro drug release studies

The drug release experiments were carried out in simulated gastric fluid (SGF) and simulated intestinal fluid (SIF) at 37 °C. The 1 cm × 1 cm samples were immersed in the buffer medium, and the amount of drug (Amoxicillin) released in the medium was sampled periodically and analyzed using UV-2450 (Shimadzu) spectrophotometer by monitoring the UV absorbance at λ_{max} = 272 nm. Sink conditions were used in all the experiments. The amount of drug released M_t at a time t was determined using a calibration curve with $R^2 = 0.999$.

Results and discussion

Characterization of chitosan

The degree of deacetylation of low viscous chitosan was determined as 84.55 % based on carbon and nitrogen content using eq. (1) and the molecular weight was evaluated as $3.5 \cdot 10^5$ Da from dilute solution viscosity studies using eq. (4).

Swelling properties of CS and CS/XA physically crosslinked films

Swelling behavior is one of the most important properties to be characterized of pH sensitive drug delivery hydrogel system. Fig. 3 represents the degree of swelling of physically crosslinked CS and CS/XA films at 37 °C in SIF. Samples of CS film when placed in SGF (pH 1.2) disintegrated into small parts within five minutes due to protonation of amino groups of chitosan (NH_2 becomes NH_3^+). This indicates that the crystal structure is broken down in acidic conditions due to the electrostatic repulsion between protonated amine groups in chitosan. When CS/XA blend films were investigated for swelling in SGF, they also disintegrated implying the hydrogen bonds between the two polymers were not strong at acidic pH. However, in SIF (pH 7.4), CS as well as CS/XA films were found to be stable, which may be due to the fact that the amino groups were deprotonated, and the amine and hydroxyl groups on glucosamine unit of CS could form strong intra- and inter-molecular hydrogen bonds. However, when hydrogel samples were placed in the medium of study (SIF), a decrease in weight with time was observed, indicating erosion of the polymer matrix. Therefore, when degree of swelling, *S*, is evaluated using eq. (5) and plotted against time, it draws a negative slope. The weight loss was observed to decrease from 29 % to 13 % as XA concentration increased from 10 to 50 wt % in CS/XA hydrogel.

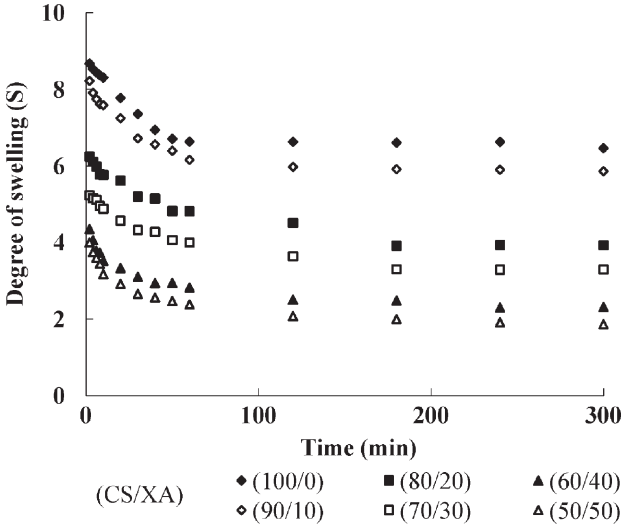

Fig. 3 – *Swelling curves of physically crosslinked CS and CS/XA hydrogels in pH 7.4 at 37 °C*

Swelling properties of chemically crosslinked CS and CS/XA semi-IPN films

Swelling curves of chemically crosslinked CS and CS/XA semi-IPN films at pH 1.2 and 7.4 are shown in Fig. 4 and Fig. 5, respectively. Initially, swelling occurred at a high rate which gradually decreased until the hydrogels reached an equilibrium state shown by the plateau region. The equilibrium degree of swelling of CS film was observed to be 3.13 and 2.91 in buffer of pH 1.2 and 7.4, respectively. The swelling effect is due to solvent penetration in hydrophilic polymers within the matrix. The chemically crosslinked films showed a lower equilibrium degree of swelling than physically crosslinked films (pH 7.4) because crosslinking hinders the mobility of polymer chains and hence lowers the swelling capability.

Swelling of films in acidic medium (pH 1.2) is due to protonation of amine groups of chitosan (NH_2 becomes NH_3^+) causing the repulsions between polymer chains and swelling to increase, while at pH 7.4, deprotonation of amino groups reduces the repulsions in the polymer chains causing swelling to decrease. In case of CS/XA semi-IPNs, with increase in XA concentration, equilibrium degree of swelling

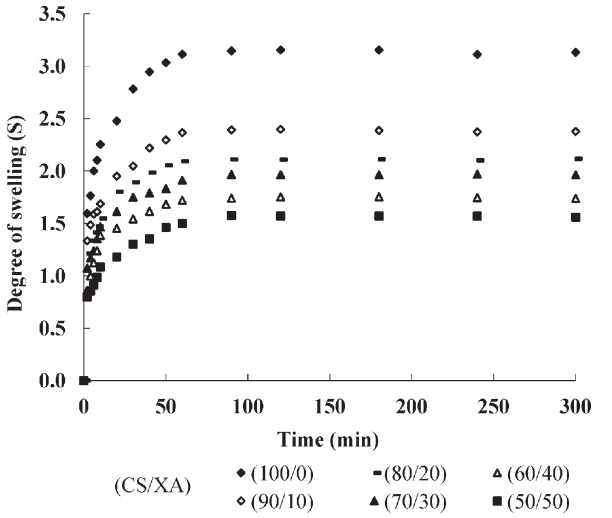

Fig. 4 – *Swelling curves of chemically crosslinked CS and CS/XA semi-IPN films with 0.1 mL 5 % (v/v) GA in pH 1.2 at 37 °C*

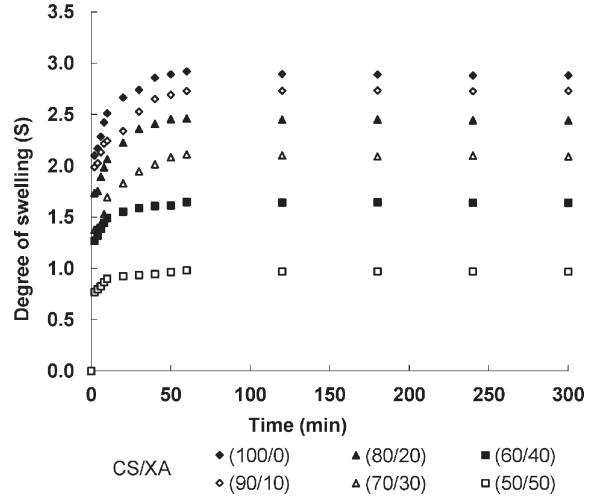

Fig. 5 – *Swelling curves of chemically crosslinked CS and CS/XA semi-IPN films with 0.1 mL 5 % (v/v) GA in pH 7.4 at 37 °C*

was found to decrease to 0.97 at pH 7.4, and to 1.58 at pH 1.2. This may be due to the strong interactions between positively charged amino groups in chitosan (NH_3^+) and negatively charged carboxylic groups (COO^-) in xanthan,[15] which leads to tightening of the network, and hence to the decreased swelling observed in the swelling media.

Efforts were also made to assess the degradation (weight loss) of chemically crosslinked CS and CS/XA semi-IPN films when placed in buffer mediums. After attaining equilibrium swollen state, the films were dried completely and the dry weights determined. The difference in initial dry weight and final dry weight gives an idea about weight loss occurring (in the film with time) during the swelling measurements. The weight loss for films was determined as given in Table 1. About 11.5 % and 10 % weight loss occurred in CS film in buffer solution of pH 1.2 and 7.4, respectively, which decreased to 5.08 % in pH 1.2, and 4.76 % in pH 7.4 with increase in XA content in CS/XA semi-IPNs. The weight loss was observed to be the least for 50/50 CS/XA blend suggesting maximum interactions between CS and XA at this composition.[16,17]

Table 1 – *Weight loss data of semi-IPN hydrogel films at pH 1.2 and 7.4, 37 °C*

S.No.	CS/XA (w/w)%	Weight loss (%)*	
		pH 1.2	pH 7.4
1	100/0	11.47	10.00
2	90/10	10.38	8.11
3	80/20	8.03	7.35
4	70/30	7.50	6.14
5	60/40	6.15	5.55
6	50/50	5.08	4.76

$$*Weight\ loss = \left(\frac{W_{di} - W_{df}}{W_{di}}\right) \cdot 100$$

where, W_{df} is the final dry weight of sample at time t and W_{di} is the initial dry weight of sample.

Effect of crosslinker concentration on degree of swelling of CS/XA semi-IPN

In order to study the influence of crosslinking density on swelling behavior of CS/XA semi-IPN, 60/40 CS/XA semi-IPNs were prepared by varying the amount of crosslinker. Different amounts of GA i.e. 0.1, 0.2, 0.3 and 0.4 mL of 5 % v/v (GA) were added to 20 mL of 60/40 CS/XA polymer solution. It was observed that equilibrium degree of swelling (***S***) decreased from 1.64 to 1.04 with increase in GA concentration at pH 7.4, and from 1.75 to 1.17 at pH 1.2 (Table 2) due to increase in crosslinking density and network stability. Highly crosslinked semi-IPNs have a tight structure as crosslinking hinders the mobility of polymer chain and hence lowers swelling ability of hydrogels.[18]

Table 2 – *Effect of crosslinker concentration on equilibrium degree of swelling of 60/40 CS/XA semi-IPN at 37 °C*

Volume of 5 % (v/v) GA / 20 mL polymer solution	Equilibrium degree of swelling, *S*	
mL	pH = 7.4	pH = 1.2
0.0	2.50	–
0.1	1.64	1.75
0.2	1.47	1.56
0.3	1.29	1.31
0.4	1.04	1.17

Swelling kinetics

The swelling kinetics of the semi-IPNs was analyzed by the following relation (eq. 6) which could be used for hydrogels exhibiting extensive swelling behavior.[19,26]

$$\frac{dS}{dt} = k\left(S_{max} - S\right)^2 \tag{6}$$

where, S is the degree of swelling at time t, and S_{max} represents equilibrium degree of swelling or maximum swelling, and k is the kinetic constant. The relation represents second order kinetics. Linearized form of eq. (6) is expressed as:

$$t/S = A + Bt \tag{7}$$

where, B is the inverse of maximum swelling = $1/S_{max}$ and $A = 1/(dS/dt)_o$ is the reciprocal of the initial swelling rate (r_o) of the hydrogel. Representative swelling kinetic curve for CS/XA (50/50) semi-IPN in SGF (pH 1.2) and SIF (pH 7.4) is shown in Fig. 6 having a regression coefficient of 0.999. Sim-

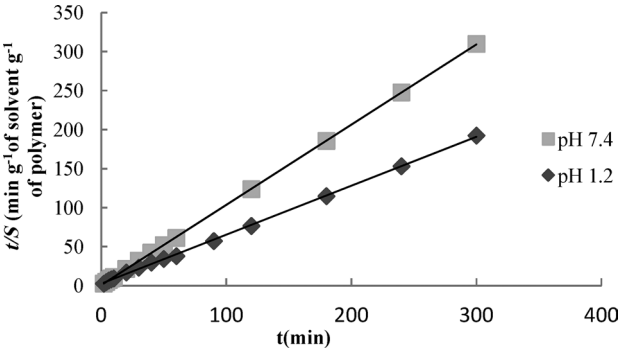

Fig. 6 – *Swelling kinetics curve of chemically crosslinked 50/50 CS/XA semi-IPN blend film with 0.1 mL 5 % (v/v) GA in pH 7.4 and 1.2*

Table 3 – *Swelling parameters in buffer solution of pH 1.2 and 7.4 at 37 °C*

S.No	CS/XA (w/w)%	pH = 1.2					pH = 7.4				
		S_{max} (Eq. 7)	r_o (min^{-1})	S_{exp} (Eq. 5)	MRQE (Eq. 8)	AAE (%) (Eq. 9)	S_{max} (Eq. 7)	r_o (min^{-1})	S_{exp} (Eq. 5)	MRQE (Eq. 8)	AAE (%) (Eq. 9)
1	100/0	3.17	1.07	3.13	0.07	2.79	2.89	2.93	2.91	0.04	3.17
2	90/10	2.41	0.82	2.38	0.08	0.293	2.74	2.27	2.74	0.06	2.30
3	80/20	2.14	0.76	2.12	0.08	0.328	2.46	2.07	2.46	0.05	2.52
4	70/30	1.99	0.63	1.96	0.08	0.235	2.10	1.92	2.09	0.09	6.14
5	60/40	1.77	0.58	1.75	0.03	1.07	1.64	1.50	1.64	0.01	0.122
6	50/50	1.59	0.39	1.56	0.10	0.155	0.97	1.32	0.98	0.03	0.3309

ilar behaviour was observed for other semi-IPNs. The initial swelling rate and the value of maximum swelling of the hydrogel were evaluated from the intercept and slope of the lines respectively, and the values are listed in Table 3. It can be observed from the data that as XA concentration increased in the hydrogel, the value of S_{max} decreased in both the mediums. The data shows close correspondence between theoretical and experimental values of equilibrium degree of swelling which was substantiated by finding mean relative quadratic error (MRQE)[14] and percentage absolute average error (% AAE)[15] using eq. (8) and (9), respectively, which further corroborated that the experimental data is well represented by eq. (7) with low values of MRQE and % AAE (Table 3).

$$MRQE = \sqrt{\frac{\sum\left(\frac{S_{cal} - S_{exp}}{S_{exp}}\right)^2}{N-1}} \quad (8)$$

$$\%AAE = \frac{100}{N}\sum_{1}^{N}\left|S_{exp} - S_{cal}\right| \quad (9)$$

where, S_{cal} is the calculated value of degree of swelling (eq. 7), S_{exp} is the experimental degree of swelling (eq. 5) and N is the number of observations.

Fourier Transform Infrared Analysis

FTIR spectrum of CS, XA and CS/XA semi-IPNs is shown in Figs. 7 and 8. CS exhibits distinctive bands at 1646 cm^{-1} and 1557 cm^{-1} corresponding to amide I (–CONH–), and amide II, (–NH–) respectively.[20] Bands at 1149 cm^{-1} (asymmetric stretching of C–O–C bridge), 1061 cm^{-1} and 1024 cm^{-1} (skeletal vibrations involving C–O stretching in alcohol group of chitosan) are characteristics of its saccharide structure. The band in the region of 3200–3700 cm^{-1} is assigned to N–H and O–H stretching bands of chitosan.

FTIR spectra of pure xanthan exhibits peaks at 3271 cm^{-1}, 2885 cm^{-1} and 1601 cm^{-1} which are as-

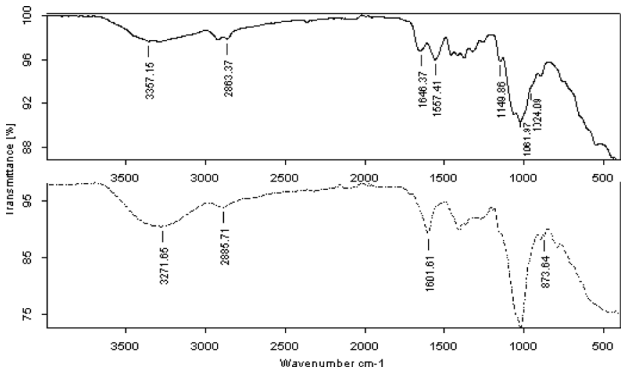

Fig. 7 – *FTIR spectra of Chitosan film and Xanthan*

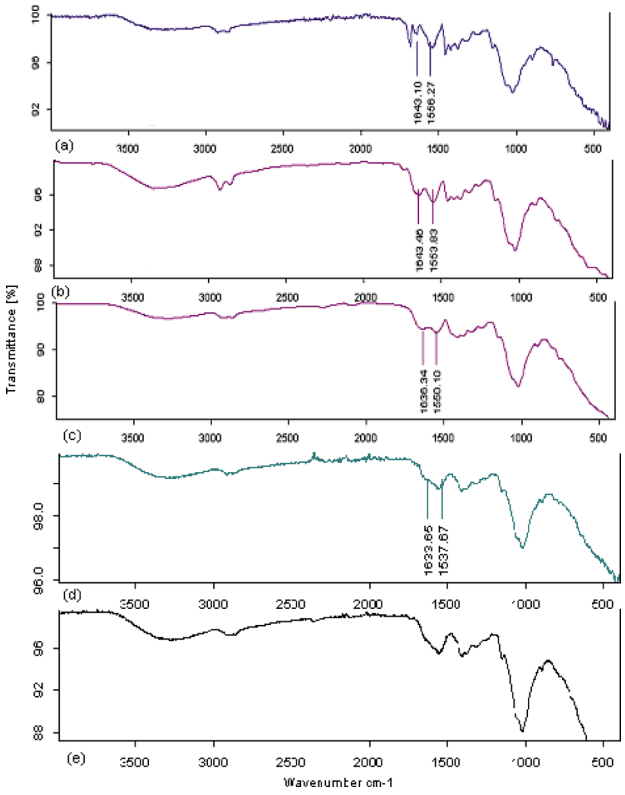

Fig. 8 – *FTIR spectra of CS/XA blend films (a) 90/10 (b) 80/20 (c) 70/30 (d) 60/40 (e) 50/50*

signed to O–H stretching, typical C–H stretching vibrations and –C=O of pyruvate group in xanthan. It could be observed that the broad peak occurring in the region 3200–3700 cm^{-1} assigned to N–H and O–H stretching bands of chitosan, shifts to lower wavenumber with increase in XA concentration in the hydrogel (from 3357 cm^{-1} for pure CS to 3339 cm^{-1} for 50/50 CS/XA semi-IPN, Fig. 8). The maximum shift occurs for 50/50 CS/XA semi-IPN and broadens, indicating strong interactions between $–NH_2$ group of CS and –OH group of XA. Also, the amide II peak appearing at 1557 cm^{-1} for CS shifts to lower wavenumber for 90/10, 80/20, 70/30 and 60/40 CS/XA semi-IPNs, whereas 50/50 CS/XA semi-IPN exhibits a broad peak suggesting strong interactions between CS and XA through hydrogen bonds.

FTIR of drug loaded films was also conducted to study interactions between the drug and the polymer. Amoxicillin exhibits peaks at 1772 cm^{-1} and 1685 cm^{-1} (Fig. 9) which are assigned to –C=O stretching in carboxylic group and ketonic group respectively. It is observed that –C=O stretching vibration, which appears at 1685 cm^{-1} for the active agent, is shifted to lower wavenumber at 1644 cm^{-1} in drug loaded semi-IPN. These peaks are absent in unloaded semi-IPN as shown in Fig. 9. Interaction is known to occur between the drug and the polymer if appearance of new peaks or disappearance of peaks is observed. Drug loaded 50/50 CS/XA semi-IPN shows peak at 1556 cm^{-1}, which is intermediate of 1553 cm^{-1} (appearing in 50/50 CS/XA semi-IPN) and 1614 cm^{-1} (appearing in amoxicillin), also indicating strong interactions between the drug and polymer.[21] Drug loaded 50/50 CS/XA semi-IPN shows that the peak in the region 3200–3500 cm^{-1} is

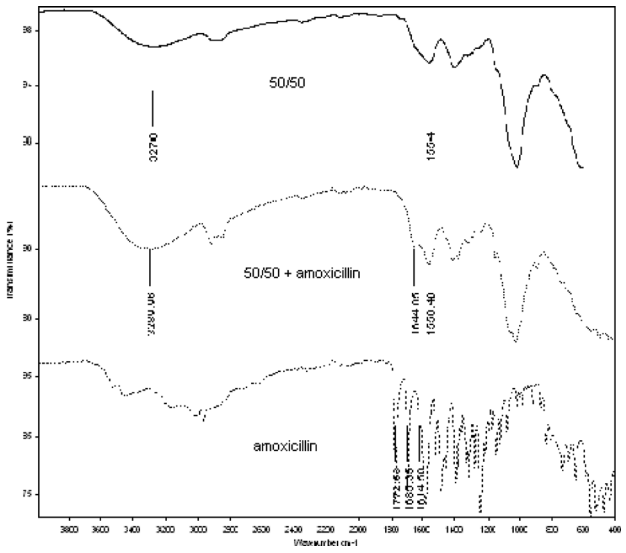

Fig. 9 – *FTIR spectra of 50/50 CS/XA blend film, 50/50 CS/XA blend loaded with Amoxicillin and pure Amoxicillin*

Table 4 – *FTIR of CS/XA blend films (Amide-II band)*

S.No.	(CS/XA) (w/w)%	Wave number (cm^{-1})
1	100/0	1557.41
2	90/10	1556.27
3	80/20	1553.83
4	70/30	1550.10
5	60/40	1537.67
6	50/50	–

broadened, which clearly indicates the presence of hydrogen bonding between the hydrogel and drug.

X-ray diffraction

Diffractogram of pure chitosan shows no sharp peak, confirming its amorphous character. XRD analysis of XA shows broad peaks at 8° and 20° indicating its partly crystalline character. It is observed that 50/50 CS/XA blend is also amorphous in character as no sharp peaks are observed as shown in Fig. 10. With the addition of xanthan to chitosan, the peaks of xanthan are diminished, clearly indicating the presence of strong interactions between chitosan and xanthan. Fig. 11. shows

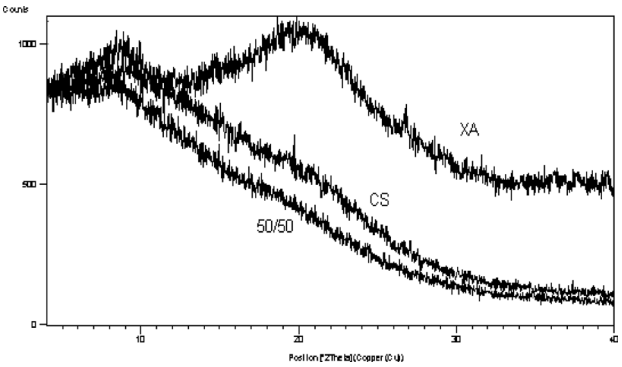

Fig. 10 – *XRD curves of pure Xanthan, pure Chitosan and 50/50 CS/XA semi-IPN blend film*

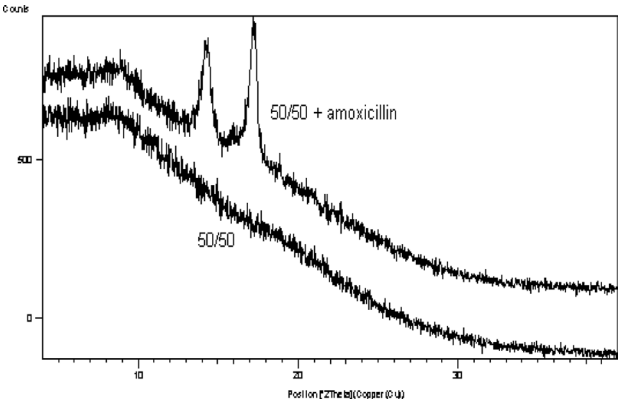

Fig. 11 – *XRD curve of Amoxicillin loaded and unloaded chemically crosslinked 50/50 CS/XA semi-IPN blend films*

diffractogram of amoxicillin loaded 50/50 CS/XA film with two sharp peaks indicating the crystalline nature of drug loaded 50/50 CS/XA film blend.

Thermogravimetric analysis

TGA curves of CS/XA semi-IPN are shown in Fig. 12. and Fig. 13. The first thermal event is observed in the temperature range 50–150 °C, where all samples present a mass loss ranging from 9 % to 17 %. This weight loss is attributed to the evaporation of water. The onset method was used to determine degradation temperature of physically and chemically crosslinked films, as listed in Table 5.

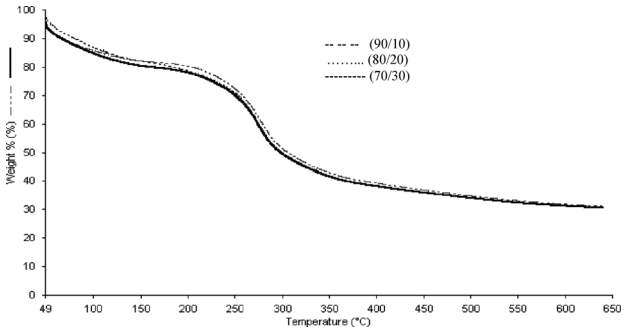

Fig. 12 – *TGA curves for chemically crosslinked CS/XA (90/10, 80/20, 70/30) films*

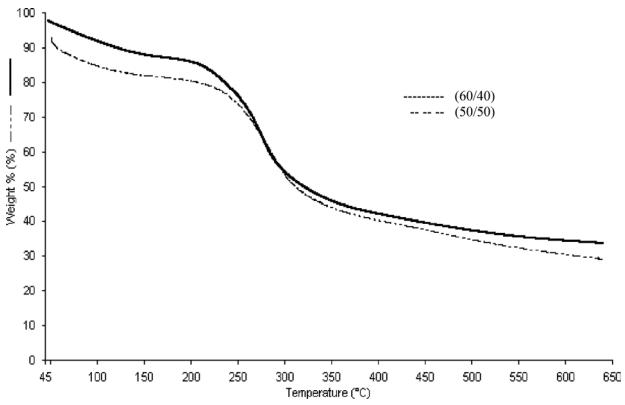

Fig. 13 – *TGA curves for chemically crosslinked CS/XA (60/40, 50/50) semi-IPN films*

The degradation temperature of CS film was found to be 235.99 °C, which increased to 253.09 °C when the XA concentration in the blend increased to 50 wt %. Chemically crosslinked pure CS film showed thermal degradation temperature of 240.62 °C. With increase in XA concentration in chemically crosslinked semi-IPN blends, the thermal degradation temperature increased to 262.43 °C as shown in Table 5. The degradation temperatures of all chemically crosslinked CS/XA blends were observed to be higher than their respective physically crosslinked blends suggesting increase in the intermolecular attractions between the two polymers.

Table 5 – *Thermal degradation temperatures of physically and chemically crosslinked pure CS and CS/XA semi-IPN blend films*

S. No.	Blend composition (CS/XA) (w/w)%	Thermal degradation temperature (°C)	
		Physically crosslinked	Chemically crosslinked*
1	100/0	235.99	240.62
2	90/10	238.29	243.36
3	80/20	241.65	244.06
4	70/30	246.26	250.87
5	60/40	250.02	253.92
6	50/50	253.09	262.43
7	0/100	229.00	–

*(0.1 mL of 5 % v/v GA/20 mL of CS/XA blend solution)

In vitro drug release

Drug release studies were carried out for crosslinked CS and CS/XA semi-IPN films at different drug loadings. Drug release profiles of crosslinked CS films with drug loadings (mg/g) of 50 % and 100 % at pH 1.2 and 7.4 are shown (Fig. 14). It can be seen that the drug released immediately with about 70 % and 45 % of amoxicillin being released within 30 minutes at pH 1.2 and 7.4, respectively for 50 % drug loading. However, a gradual release profile is observed for 50/50 CS/XA semi-IPN as compared to sudden release profile for CS film (Fig. 15(a) and 15(b)). Unlike pure CS film, about 61 % of amoxicillin was released in 1 hour in 1.2 and 37 % at 7.4 pH. The total amount of drug release from pure CS film was more than from 50/50 CS/XA semi-IPN. This may be attributed to the fact that CS/XA semi-IPN forms a compact matrix. The strong interactions between positively charged chitosan (NH_3^+) and negatively charged carboxylic functional groups (COO^-) of xanthan gum[18] led to a

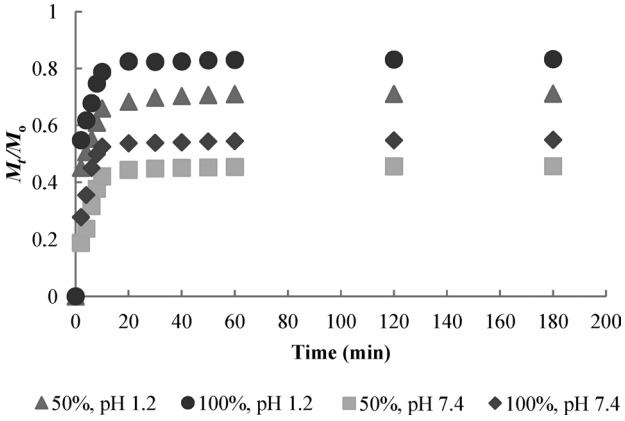

Fig. 14 – *Drug release profile from crosslinked CS film at pH 1.2 and 7.4, 37 °C at different drug loadings*

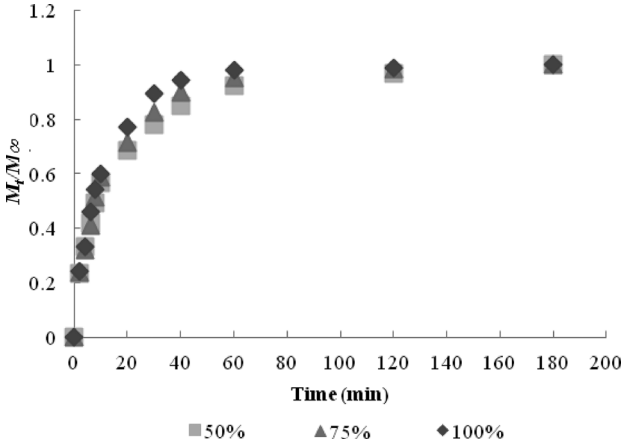

Fig. 15(a) – *Drug release profile from 50/50 CS/XA semi-IPN films at pH 1.2, 37 °C at various drug loadings (%)*

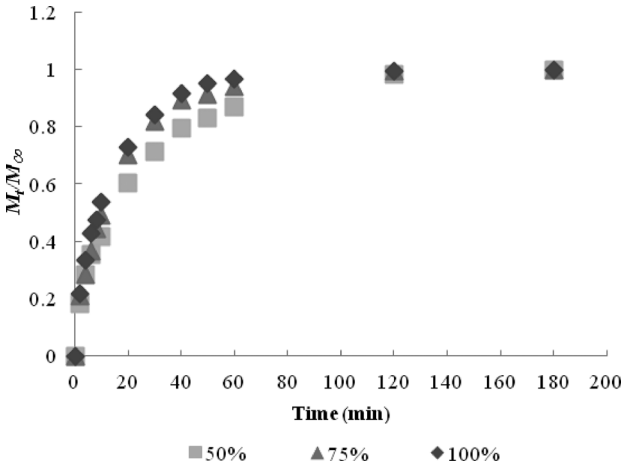

Fig. 15(b) – *Drug release profile from 50/50 CS/XA films at pH 7.4, 37 °C at various drug loadings (%)*

decreased equilibrium degree of swelling, and hence the path for drug diffusion became more obstructed leading to lower amount of drug release. It can also be observed that a higher amount of drug is released in SGF (pH 1.2) than in SIF (pH 7.4), which could be attributed to the higher degree of swelling obtained in SGF. At pH 7.4, the protonation of chitosan decreases, hence repulsions in the polymer chains recede allowing less swelling and thus lower amount of drug release. The transport mechanism of amoxicillin from semi-IPN was observed to be anomalous in nature, indicating drug release occurring due to the swelling mechanism rather than the erosion mechanism. This is also evident from swelling studies and weight loss studies of semi-IPN films. In case of 50/50 CS/XA semi-IPN film, only about 4–5 % weight loss occurred in 5 hours indicating insignificant erosion mechanisms in the polymer matrix.

The effect of drug loading (50 %, 75 % and 100 %) on release kinetics was also studied. The results are presented in Fig. 14 and Fig. 15(a,b) for drug release studies conducted at pH 1.2 and 7.4 for pure crosslinked CS film and 50/50 CS/XA semi-IPN film, respectively. Pure chitosan film with 50 % drug loading showed 70 % and 45 % release at pH 1.2 and 7.4, respectively, while drug loading of 100 % showed 83 % and 54 % release at pH 1.2 and 7.4. The amount of drug released was found to be dependent on the matrix drug loading. It was observed that at higher drug loading, the rate of drug release from hydrogel film also increased due to higher concentration gradient available. With higher drug loading, more drug molecules are available leading to higher drug release.

Analysis of the amoxicillin release kinetics from 50/50 CS/XA semi-IPN was performed by calculating the diffusion coefficient. The solution of Fick's second law of diffusion under the initial and boundary conditions equivalent to those of testing in this work is an infinite series given by (10):[22]

$$\frac{M_t}{M_\infty} = 1 - \sum_{n=0}^{\infty} \frac{8}{(2n+1)^2 \pi^2} \exp\left[\frac{-(2n+1)^2 \pi^2 D}{L^2} t\right] \quad (10)$$

where, D is the diffusion coefficient in $m^2\ s^{-1}$, t is release time in seconds, L is the thickness of polymer sample in m, M_t/M_∞ denotes the fraction of drug molecules released up to any time t. To enable reasonable modeling of the diffusion process, the 'early-time' approximation, (11), and 'late-time' approximation, (12), were used. The early-time approximation method[12] is valid for the first 60 % of drug release data from crosslinked polymers in a chosen solvent, and the late-time approximation is valid for the latter 40 % of drug release data:

$$\frac{M_t}{M_\infty} = 4\left[\frac{D_E t}{\pi L^2}\right]^{1/2} \quad (11)$$

$$\frac{M_t}{M_\infty} = 1 - \frac{8}{\pi^2} \exp\left[\frac{-\pi^2 D_L t}{L^2}\right] \quad (12)$$

where, D_E and D_L are the early-time and late-time diffusion coefficients.

For drug loaded 50/50 CS/XA semi-IPN hydrogel, the values of D_E and D_L in SGF and SIG calculated from the model fits of the release process using eq. (11) and (12) respectively, are listed in Table 6. In general, there was no significant difference in the values of early-time drug diffusion coefficients in SGF and SIF, which may be due to the negligible difference in the degree of swelling of the same composition of semi-IPNs in the two media. Also, there was no dramatic change in the diffusion coefficients between early- and late-time for a given drug loading in the media of study. It was observed that the value of D_E increases as the

Table 6 – *Early-time and late-time drug diffusion coefficients and release exponents for drug loaded 50/50 CS/XA semi-IPNs in buffer solution of pH 1.2 and 7.4 at 37 °C*

Drug loading % (mg g^{-1})	pH 1.2			pH 7.4		
	Early-time drug diffusion coefficient, $D \cdot 10^{-13}$, $m^2\ s^{-1}$	Late-time drug diffusion coefficient, $D \cdot 10^{-13}$, $m^2\ s^{-1}$	Release exponent, n	Early-time drug diffusion coefficient, $D \cdot 10^{-13}$, $m^2\ s^{-1}$	Late-time drug diffusion coefficient, $D \cdot 10^{-13}$, $m^2\ s^{-1}$	Release exponent, n
50	4.83	3.48	0.539	2.94	2.48	0.508
75	4.95	3.97	0.559	3.69	3.47	0.539
100	5.54	5.46	0.581	4.57	3.97	0.559

amount of drug increases in the matrix because of higher concentration gradient available. The early-time diffusion coefficients ranged from $4.83 \cdot 10^{-13}$ m^2 s^{-1} to $5.54 \cdot 10^{-13}$ m^2 s^{-1}, and late-time diffusion coefficients ranged from $3.48 \cdot 10^{-13}$ m^2 s^{-1} to $5.46 \cdot 10^{-13}$ m^2 s^{-1} at pH 1.2. The early-time diffusion coefficients at pH 7.4 ranged from $2.94 \cdot 10^{-13}$ m^2 s^{-1} to $4.57 \cdot 10^{-13}$ m^2 s^{-1}, and late-time diffusion coefficients ranged from $2.48 \cdot 10^{-13}$ m^2 s^{-1} to $3.97 \cdot 10^{-13}$ m^2 s^{-1}.

Another empirical equation developed by Ritger and Peppas for the early-time approximation assumes a time-dependent power law function.[23,24]

$$\frac{M_t}{M_\infty} = kt^n \tag{13}$$

Here, k is a structural/geometric constant for a particular system, and n is the release exponent representing the solute transport mechanism. When n is equal to 0.5, the transport mechanism is Fickian (as was assumed for the early-time approximation equation) and the drug release rate is time-dependent. When n is between 0.5 and 1.0, anomalous transport occurs where the polymer relaxation as well as Fickian diffusion control the drug release, and the drug release rate is time-dependent. For n equal to 1.0, polymer relaxation governs solute release and is called Case II type diffusion leading to zero-order release. For drug loaded 50/50 CS/XA semi-IPN hydrogels, M_t/M_∞ vs t plots were plotted at different drug loadings, and values of n were calculated from the curves. The values of the n obtained from eq. (13) are given in Table 6.

As can be seen, the values of n lie in the range $0.5 < n < 1.0$, indicating non-Fickian drug diffusion mechanism, where both diffusion and polymer relaxation control the overall rate of drug release.[14]

Conclusion

Physically crosslinked CS/XA films converted into thick gel in SGF of pH 1.2 almost instantaneously and in SIF of pH 7.4, films were too fragile to be useful as a drug delivery system. Semi-IPNs prepared using GA as the crosslinker were found to be stable in both the buffer mediums. It was observed that 50/50 CS/XA semi-IPN showed least equilibrium degree of swelling in both the buffer mediums, indicating the existence of maximum interactions between the two polymers in the hydrogel. The degree of swelling was found to decrease with increase in crosslinker concentration. Drug release studies conducted on 50/50 CS/XA semi-IPN film showed non-Fickian drug diffusion mechanism, with higher release in SGF solution than in SIF solution, indicating that these semi-IPN films could serve as potential candidates for antibiotic delivery in an acidic environment. The CS/XA semi-IPN films were analyzed using FTIR, XRD and TGA analysis which show that interactions between the two polymers increase with the XA concentration in the semi-IPN.

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