Drug Release Study of Nalidixic acid from the Chitosan-Carrageenan Ionic Matrix.

Zeki N. AL-Sokanee[1]*, Kadhmia W. AL- Chezzy[2],Tahseen A.Saki[1], Luma J.AL-Amber[2]

[1]Chemistry Department, College of Science, Basrah University
[2] Marine Science Center Basrah University.

Abstract

A new ionic matrix hydrogel based on chitosan and k-carrageenan was prepared by the ioncally crosslinked. Carrageenan is a sulphated polysaccharide (negatively charged), when mixed with chitosan (positively charged) the ionicaly matrix is formed. The matrix is characterized by the FT.IR spectroscopy and solubility properties. The aim of nalidixic acid loaded is to retarded the fast excretion of drug from the urinary tract by imbedded into the matrix and decreasing of the dose quantity of drug and removal the side effect of the crosslinking agents. A good release obtain by examination the matrix loaded by using different pH, the release of drug at pH=7.4 consumed 60 min on best quantity of drug.

دراسة الطرح المستمر لعقار حامض الندلكسك من الشبيكة الأيونية المحضرة من الكيتوسان و الكارجنان.

زكي ناصر السكيني* كاظمية الغزي** تحسين علي صاكي * لمى جاسم العنبر**

*قسم الكيمياء- كلية العلوم –جامعة البصرة.
** مركز علوم البحار- جامعة البصرة.

الخلاصة:

حضرت شبيكة أيونية هلامية جديدة مستندة إلى السكر المتعدد الكيتوسان والكاراجنان من خلال التشابك ألايوني الناتج بين مجاميع السلفيت السالبة الشحنة المتدلية في الكاراجنان ومجاميع الامونيوم الموجبة الشحنة في الكيتوسان. شخصت الشبيكة الجديدة المحضرة بواسطة تقنية الأشعة تحت الحمراء والصفات الذائبية.وحملت الشبيكة الأيونية بالمضاد الحيوي المعروف حامض الندلكسك إذ أن الهدف من تحميل الدواء داخل الشبيكة البوليمرية هو

لتقليل كمية الدواء المطروح إلى الخارج وزيادة زمن احتجازه داخل الشبيكة وأيضا تقليل الآثار الجانبية للعقار والعوامل المشبكة الكيماوية المستخدمة سابقا .تم الحصول على نتائج جيدة إذ وجد إن الدواء يطروح خلال 60 دقيقة وبكميات مدروسة عند الدالة الحامضية الفسيولوجية. كذلك تم دراسة الصفات الهلامية للشبيكة وكانت النتائج جيدة إذ وجد انه بإمكان الشبيكة إن تسحب كميات جيدة من الماء تتناسب والغرض المحضرة من اجله.

Introduction

One reason why chitosan has become of interest is undoubtedly because it can be obtained from natural sources that are abundant and renewable. Chitosan is prepared from chitin, the polymer second most abundant in nature after cellulose(Muzzarelli,1977). Chitin is the primary structural component of the outer skeletons of crustaceans, and of many other species such as molluscs, insects and fungi (Shepherd et al.1997).

The role played by chitin is similar to the roles played by cellulose in plants and collagen in higher animals. Chitosan is prepared from chitin to obtain a more reactive polymer. Chitosan is a linear polysaccharide

consisting of ß (1-4)-linked 2-amino-2- deoxy-D-glucose (D-glucosamine) and 2-acetamido-2-deoxy-D-glucose (N-acetyl-Dglucosamine) units (Fig. 1) (Ciechanska, 2004) . The structure of chitosan is very similar to that of cellulose (made up of ß (1-4)-linked D-glucose units), in which there are hydroxyl groups at C2 positions of the glucose rings. The term chitosan is used to describe a series of polymers of different degrees of deacetylation (*DD*), defined in terms of the percentage of primary amino groups in the polymer backbone, and average molecular weights (*Mw*). The *DD* of chitosan is usually between 70 and 95%, and the *Mw* between 10 and 1000 kDa. Changing the reaction conditions during the manufacture of chitosan from chitin can alter the *DD* and *Mw* of chitosan (Dodane and Vilivalam,1998).

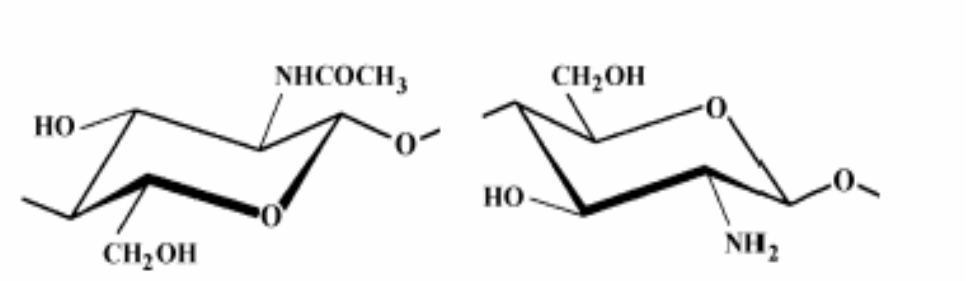

Figure 1. Structural units of chitosan and its parent substance chitin. Chitin consists mostly of N-acetyl-D-glucosamine –units (left). During the preparation of chitosan most units are deacetylated to D-glucosamine –units (right).

Several properties of chitosan make it potentially valuable as a pharmaceutical excipient. Good biocompatibility and low toxicity of chitosan, and the fact that sources of chitosan are abundant, are properties that any new excipient material should have (Jaworska et al.2003) . One property that makes chitosan particularly interesting for study as an excipient is its ability to become hydrated and form gels in acidic aqueous environments. Because of its gel-forming ability, a major area of interest since studies began has been use of chitosan to prepare slow release drug delivery systems (Aieden et al., 1997). Chitosan has been evaluated in vitro as a drug carrier in hydrocolloids and gels, and as a hydrophilic matrix retarding drug release in tablets, granules and microparticles. The hydrophilic nature of chitosan has also aroused interest in its use in immediate-release formulations, e.g. as a disintegrant in small amounts in tablets, where it has been found to have effects similar to or better than those of microcrystalline cellulose, and as an excipient to increase the rate of dissolution of poorly soluble drug substances. It has also been suggested that chitosan might be valuable for delivery of drugs to specific regions of the gastrointestinal tract, e.g. the stomach, small intestine, and buccal mucosa

(Orient et al.,1996). Delivery to other mucosa surfaces, e.g. delivery of peptide drugs on to the nasal epithelia has also been studied. Polysaccharides whose monomers are esterized to sulfuric acid residues and are moreover partially methylated have been detected in nearly all marine algae. They occur partially in the cell wall itself and partially in the intercellular substance. Sulfonated galactanes are typical for many red algae, depending on their origin are they called ,Carrageenan (Maim et al.,2004).Carrageenan contain exclusively D-compounds. Just like in alginates is the formation of gelatine one of the most important physical properties of this family of molecules. Carrageenans are hydrophilic, high molecular weight, anionic linear heteropolysaccharides extracted from marine algae *Rhodophyceae*. There are different types of carrageenans but kappa (κ), iota (ι), and lambda (λ) are used for pharmaceutical applications(Solima et al., 1980) .Carrageenans are sulfate esters of galactose and 3,6-anhydrogalactose copolymers, linked by alternating α-1,3 and β- 1,4 glycosidic linkages (Fig. 2). Due to its gelling property, it has been studied as a release retardant for ionic and nonionic drugs (Guven et al.,1990).

K-carrageenan is extensively used as a food additve in a wide range of products including cheese, cream. chocolate and ice creams (Solima et al., 1980). A number of properties have been attributed to carrageenan viz., antigenic; stimulation of the growth of connective tissue , antiviral against certain influenza viruses , anticoagulant and antithrombic , anti-peptic or anti-ulcer (Parekh et al.,1992). Therefore, the aim of the present study is to study the effect of ionic matrix model formed between the anionic polymers (κ-carrageenan)and cationic polymer (chitosan) on the drug release of nalidxic acid.

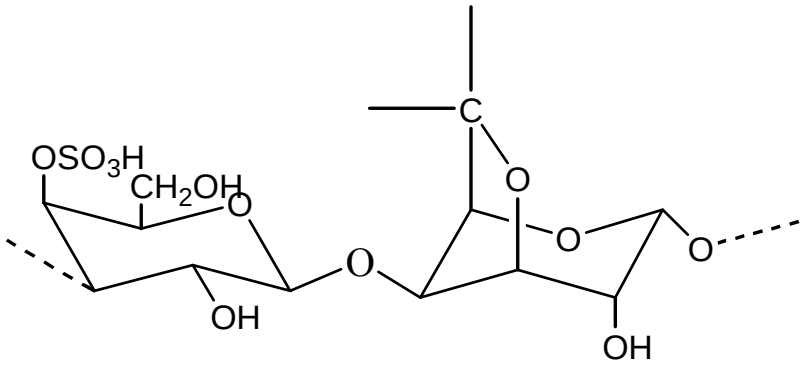

Figure 2. Structural units of carrageenan.

Materials and Methods

Chitosan from Shrimp shells isolated else where (Al-Sokanee et al. 2006), NaOH (BDH), HCl (Fluka), and Nalidixic acid (ASIA), Carrageenan (CECA S. A. France). F.T-I.R. - 8400S Fourier Transform Infrared spectrophotometer, Shimadzu, was used for recording I.R. spectra as a KBr disc. UV- visible, spectrophotometer (Shimadizu).

Preparation of the matrix

Chitosan was dissolved in 0.1M acetic acid, carrageenan dissolved in hot water was added with stirring to the chitosan solution,The feed mixture was allowed to gel for 24hrs at room temperature, followed by extensive washing with distilled water. The hydrogel was allowed to dry to a constant weight in room temperature. The same procedure was repeated with addition of proper amount of drug (Al-sokanee,2000).

Drug content

The matrix loaded the drug was treated with acid solution of HCl 12 N for 24 hrs and the drug content was estimated in UV measurement each 0.5 g of sample of matrix contain 0.1951g of drug.

Swelling Measurements

An ionic matrix thin film sample (0.10g) was immersed in 50 ml of pH media and allowed to soak to the interval times at room temperature. The swollen gel was allowed to drain by removal the water by filter papers until no more drops drained.

The swelling ratio (absorbency) was calculated using the following equation:

$$\text{Swelling Ratio}(Q)=(W_S-W_d)/W_d$$

Where w_s and w_d are the weight of the swollen gel and the dry sample, respectively. So, swelling ratio was calculated as grams of water per gram of matrix (g_{water}/g_{Gel}).

Experimental of Drug Release

In this study, several samples (with given weights) were pressed to discs (10-12 nm diameters), the samples were suspended in 25 ml of phosphate buffer solution (PBS) [KH_2PO_4 (2.14 g), NaH_2PO_4 (11.46 g) in 1000 ml of distilled water] containing (0.03 %, w/w) sodium azide NaN_3 to prevent bacterial growth. The pH of the PBS is equal to 7.4 at room temperature.

The drug release was determined at different intervals of times and was followed by measuring the UV (257nm) of samples each once . All the discs samples were placed in sterilized closed conical flasks. The percent of the drug release was evaluated by using the following definitions[13]:

$$\textbf{Drug Release (\%)} = \frac{\textbf{Amount of drug release (mg)}}{\textbf{Total weight of drug sample (mg)}} \textbf{ X 100}$$

Results and Discussion

Most of the crosslinkers used to perform covalent crosslinking may induce toxicity if found in free traces before administration. A method to overcome this problem and to avoid a purification and verification step before administration is to prepare hydrogels by reversible ionic crosslinking.Fig (3,4,and5) shows the FT.IR spectra of the substrates and the ionic matrix that formed. An obvious difference assigned to formation of new matrix. The broadness of all the peaks and shifting to the high wave length that attributed to ionic interaction between the positive charge and negative of chitosan and k-carrageenan, respectively . The other evidence of the formation of the new matrix, it is insoluble in water and acetic acid.

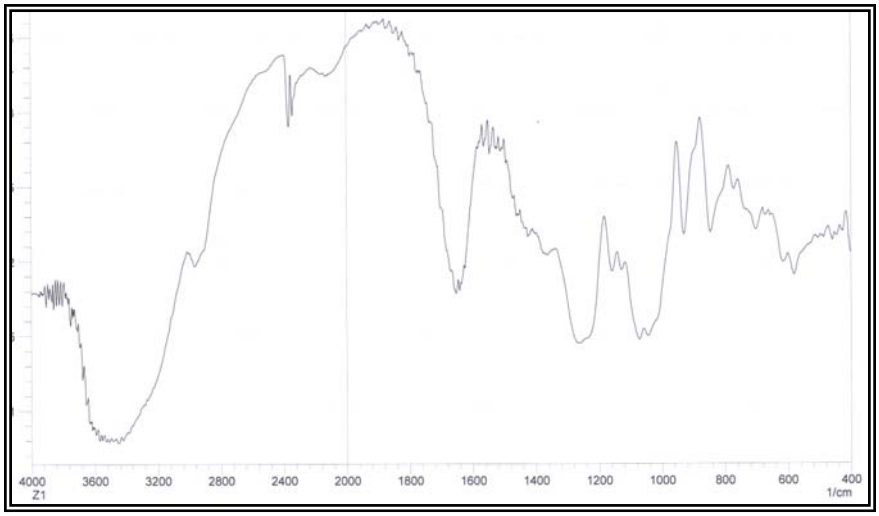

Fig. (3): FT. IR spectrum of Carrageenan.

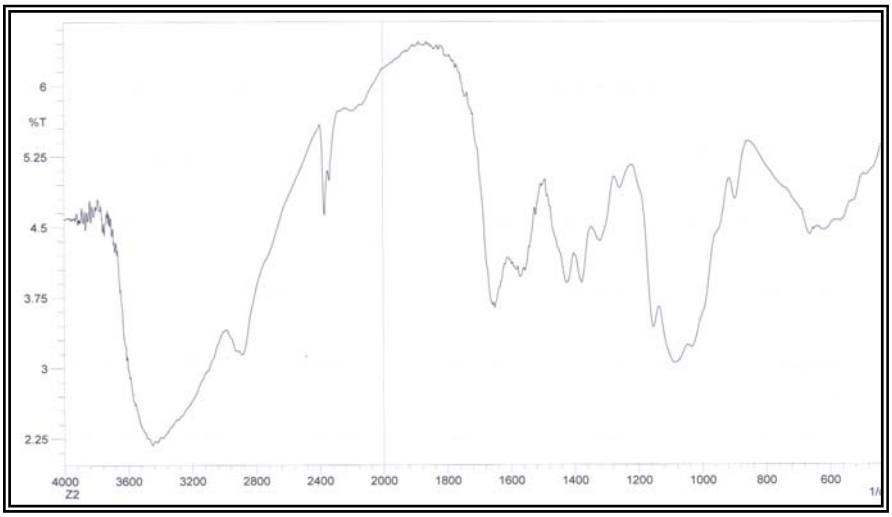

Fig. (4): FT. IR spectrum of Chitosan

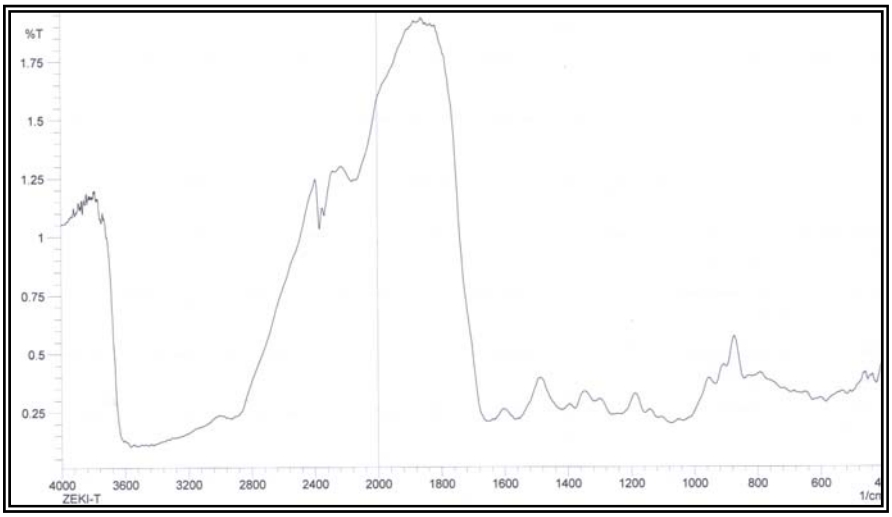

Fig. (5): FT. IR spectrum of matrix

Chitosan is a polycationic polymer, well known for its chelating properties. Therefore, reactions with negatively charged components, either ions or molecules, can lead to the formation of a network through ionic bridges between polymeric chains. Ionically crosslinked chitosan networks can be divided into two groups depending on the type of crosslinker used, either anions or anionic molecules.

Ionic interactions between the negative charges of the crosslinker and positively charged groups of chitosan are the main interactions inside the network (Majeti and Kumar,2000). Their nature depends on the type of crosslinker ($-SO_3^-$ in Carrageenan uints) these lead to electrostatic interactions formed between the positively charged ammonium groups ($-NH_4^+$) of chitosan and the anionic groups ($-SO_3^-$) used as crosslinker. Moreover, additional interactions can occur inside the network, such as hydrophobic interactions favoured by a decrease of the degree of deacetylation of chitosan or interchain hydrogen bonds due to the reduced electrostatic repulsion after neutralisation of chitosan by the crosslinker.

Ionic crosslinking is a simple and mild procedure. In contrast to covalent crosslinking, no auxiliary molecules such as catalysts are required, which is of great interest for medical or pharmaceutical applications. Indeed, ionic crosslinking can be ensured by the classical method of preparing a crosslinked network, namely by the addition of the crosslinker (Carrageenan), by solubilised to the chitosan solution. These methods allow the formation of a homogeneous hydrogel by a random crosslinking reaction. Fig.(6) shows the pH effects on the swelling ratio of the matrix.

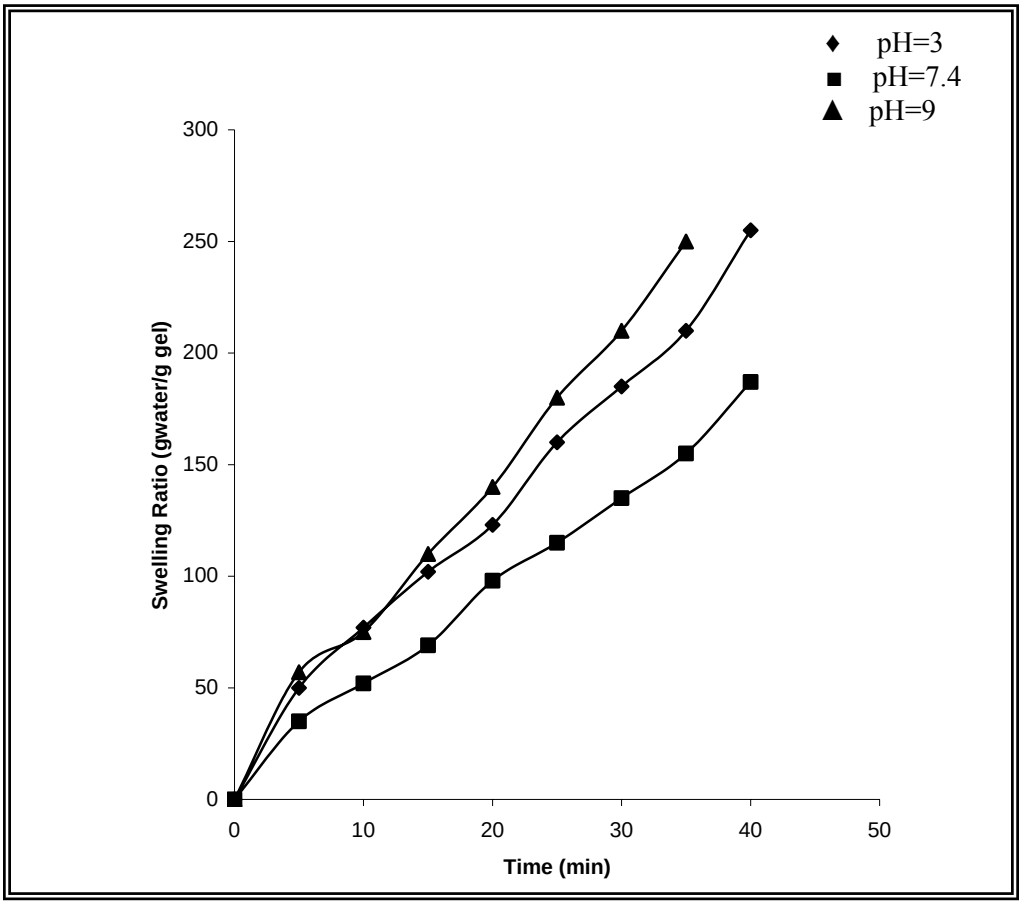

Fig.(6): The pH effect on swelling ratio of the matrix.

Networks formed by ionic crosslinking of chitosan are mainly used for drug delivery. The properties of pH dependent drug delivery systems can be controlled by the experimental conditions during preparation. They generally exhibit pH-sensitive swelling and drug release by diffusion through their porous structure .

Drug release is mainly influenced by ionic interactions between chitosan chains, which depend on the crosslinking density set during the formation of

the network. An increase in crosslinking density induces a decrease in swelling and pH-sensitivity, by improving the stability of the network, and results in decreased drug release. However, in ionically crosslinked hydrogels the crosslinking density is further modified by external conditions after administration, mainly by the pH of the application medium. It influences the global charge densities of chitosan and crosslinker, which directly determine the crosslinking density, interactions and swelling. In contrast, in covalently crosslinked hydrogels, the crosslinking density is not modified after administration since these hydrogels are linked by irreversible bonds. From the Fig. (7,8, and 9) we shows that in the ionically crosslinked hydrogels cannot only swell in acidic but also in basic conditions, which extends their potential applications. If the pH decreases, the charge density of the crosslinker and therefore the crosslinking density decrease, which leads to swelling and then the drug release increase, and this clear when seen the Fig. (8) that shows the drug release at first ten minutes reach to 45.6% in pH = 3, increasing to 76.7% at 30 mintues and 100% of drug release after 50 minutes.

Moreover, swelling is favoured by the protonation and repulsion of chitosan free ammonium groups. If the pH decrease, dissociation of ionic linkages and dissolution of the network can occur, leading to a fast drug release. If the pH increases (Fig. (9)), the protonation of chitosan decreases and induces a decrease of the crosslinking density, allowing swelling and then drug release when the pH increase, amino groups of chitosan are neutralised and ionic crosslinking is inhibited . If the crosslinking density becomes too small, interactions are no longer strong enough to avoid dissolution and the ionic crosslinker is then released .The drug release in

pH =9 was 40.2% at first 10 minutes then increasing to 69.2 % at 30 minutes and the completely drug release obtained at 60 minutes. On the other hand, a good release was obtained in pH = 7.4 (Fig. (7)) because of the nuterlized the matrix in physiological pH, the drug release at 10 minutes reach to 35.6 % and 62.4 % at 30 minutes and 100 % 60 mintues.

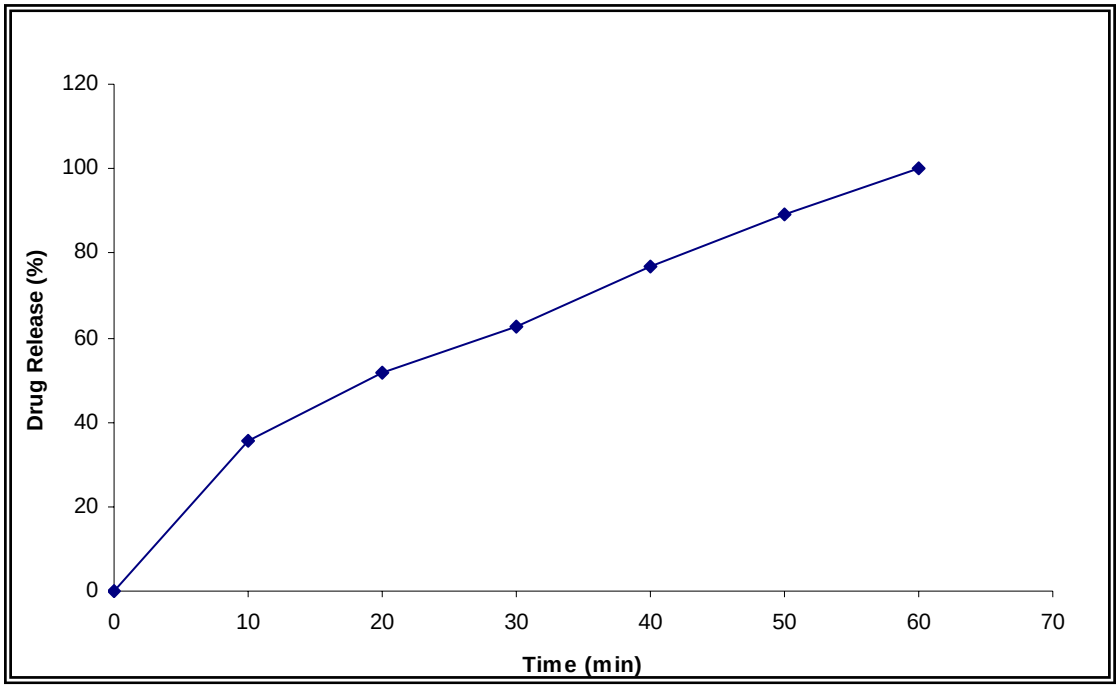

Fig (7):Nalidixic acid release at pH= 7.4.

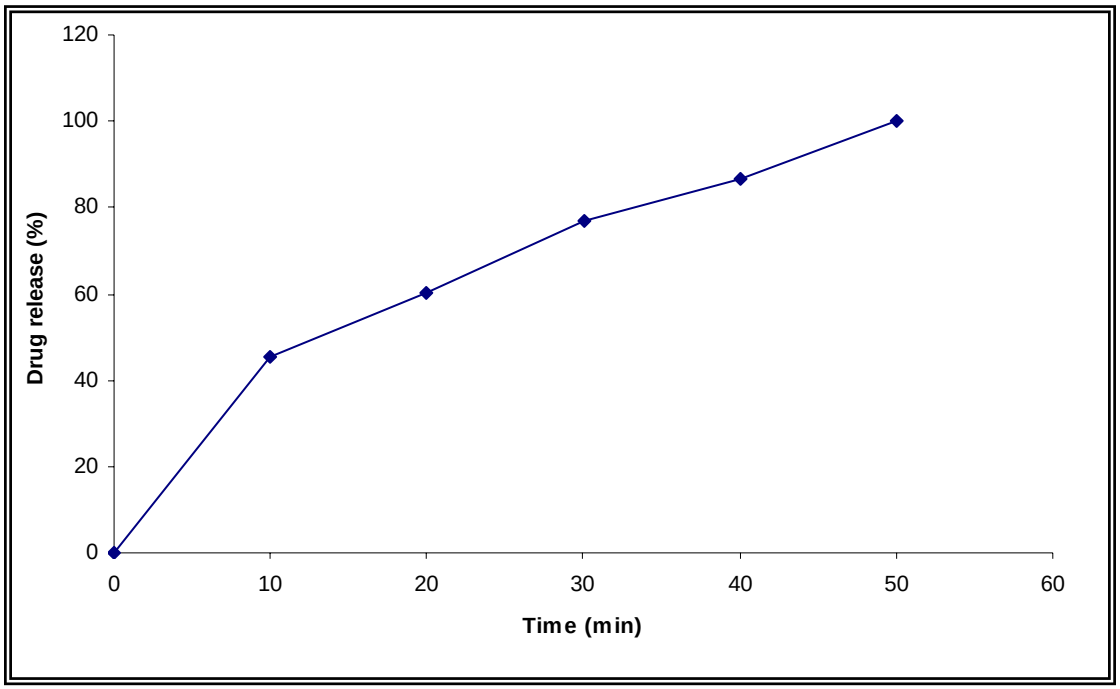

Fig (8):Nalidixic acid release at pH= 3.

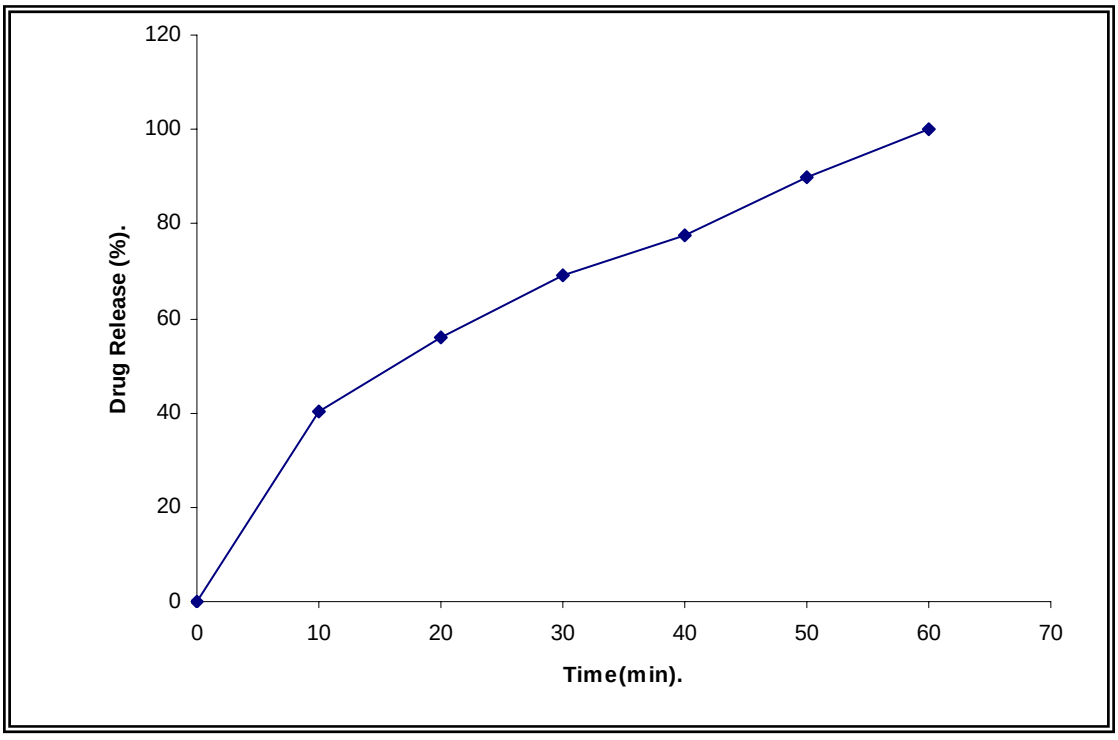

Fig (9): Nalidixic acid release at pH= 9.

Conclusion

A new ionic matrix for drug release was synthesized by ionic coupling between the cationic ammonium groups of chitosan and anionic sulfate groups of k-carrageenan under optimized conditions. The ionic crosslinked matrix was loaded with the antibacterial drug (Nalidixic Acid). The matrix was identified by the FT. IR. Spectroscopy and solubility, the drug release was investigated by varying pH of application media. When pH decrease the crosslinking density become lesser, that lead to increase the drug release in the high pH. In the high pH the protenation of amine groups decrease and the drug release become higher.

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