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Novel pH-sensitive citrate cross-linked chitosan film for drug controlled release

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Abstract

Turbidimetric titration revealed that there were electrostatic attractive interactions between citrate and chitosan in the pH region of 4.3–7.6, depending on their degree of ionization. Citrate cross-linked chitosan film was prepared simply by dipping chitosan film into sodium citrate solution. The swelling ratio of citrate/chitosan film was sensitive to pH, ionic strength etc. Under acidic conditions, citrate/chitosan film swelled and even dissociated in the pH less than 3.5, and the model drugs (brilliant blue and riboflavin) incorporated in the film were released quickly (usually within 2 h released completely in simulated gastric fluid at 37°C) while under neutral conditions the swelling ratio of citrate/chitosan film was less significant and the release rate of brilliant blue and riboflavin was low (less than 40% released in simulated intestinal fluid in 24 h). Sodium chloride weakened the electrostatic interaction between citrate and chitosan, and therefore facilitated the film swelling and accelerated drug release. The parameters of film preparation such as citrate concentration, solution pH etc. influencing the film swelling and drug release profiles were examined. The lower concentration and the higher pH of citrate solution resulted in a larger swelling ratio and quicker riboflavin release. To improve the drug controlled release properties of citrate/chitosan film, heparin, pectin and alginate were further coated on the film surface. Among them only the coating of alginate prolonged riboflavin release noticeably (for 80% of drug released the time was extended from 1.5 to 3.5 h with 0.5% w/v alginate used). The results indicated that the citrate/chitosan film was useful in drug delivery such as for the site-specific drug controlled release in stomach.

Keywords: Chitosan film; Sodium citrate; pH-sensitive; Drug controlled release

1. Introduction

Chitosan with excellent biodegradable and biocompatible characteristics is a naturally occurring polysaccharide. Due to its unique polymeric cationic character, its gel and film forming properties, chitosan has been extensively examined in the pharmaceutical industry for its potential in the development of drug delivery systems (Yao et al., 1995; Illum, 1998).

Chitosan films were usually prepared by chemical cross-linking with glutaraldehyde etc. (Nakatsuka and Andrady, 1992; Thacharodi and Rao,

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1993; Illum, 1998). These films swelled under acidic conditions due to the ionization of amino groups but remained in a shrunken state under neutral condition. Moreover, chitosan was reported to have intragastric-floating characteristics and prolonged retention of the dosage form in the stomach. By utilizing these advantages, chitosan films or other dosage forms have been exploited widely for oral sustained drug delivery in the stomach (Inouye et al., 1988; Chandy and Sharma, 1993; Patel and Amiji, 1996; Gupta and Ravi Kumar, 2000). To improve the pH-sensitive performance, blended chitosan films have usually been prepared. For example, polyether oxide/chitosan film was reported to have excellent pH-sensitivity (Yao et al., 1993; Angelova et al., 1995; Patel and Amiji, 1996).

However, the chemical cross-linking agents possibly induce toxicity and other undesirable effects. To overcome these disadvantages, recently reversible physical cross-linking by electrostatic interaction was applied in the preparation of chitosan film (Illum, 1998). Polyanions were usually used as a component to prepare these films. For example, Yao et al. (1996) reported the preparation of pectin/chitosan films by dissolving this polyelectrolyte complex in formic acid and then evaporating the solvent. Chu et al. (1995) also prepared xanthan/chitosan complex film by the solvent evaporation method in the existence of concentrated sodium chloride (ca. 0.5 M) and then treatment at a high temperature.

On the other hand, the use of low molecular weight ions to prepare an ionic cross-linking polymeric matrix was found to be very simple and mild, and the cross-linking process was accomplished just by dipping the polymer films into cross-linking ion solution (Al-Musa et al., 1999). For instance Remuñăn-López and Bodmeier (1997) prepared tripolyphosphate cross-linked chitosan film by dipping chitosan film into tripolyphosphate aqueous solution.

However, up to now, no other anion cross-linked chitosan film is reported in the literature. In our previous experiments, we found that there was electrostatic interaction between sodium citrate and chitosan, and citrate cross-linked chitosan beads or microspheres were prepared using our recently developed method (Shu and Zhu, 2000a,b). In this paper, we aim to prepare citrate cross-linked chitosan film and investigate the pH-sensitive performances of citrate/chitosan film. The preliminary results of citrate/chitosan film as pH-dependent drug controlled release matrix are also reported.

2. Materials and methods

2.1. Materials

Chitosan was obtained from Tianbao Chitosan Co. Ltd (China), and refined twice by dissolving in dilute HAc solution and precipitating from dilute ammonia, the degree of deacetylation was 86%, Mv was 460 000. Pectin (USP XXII) and sodium alginate (low viscosity) were obtained from Sigma (USA). Heparin (Mw 11 000, from porcine intestinal mucosa) was a gift from Jiuyuan Gene Co. Ltd (China). Riboflavin (Mw 376.37), theophylline (Mw 180.17) and 5-fluorouracil (5-FU, Mw 130.08) were all purchased from Aldrich (USA). Coomassie brilliant blue R250 (Mw 825) was purchased from Fluka A.G. (Switzerland) and used after sieving (less than 50 μm). Sodium citrate (analytical grade) and other reagents were all commercially available and used as received.

2.2. Turbidimetric titration

The interactions of sodium citrate and chitosan were investigated by turbidimetric titration according to the reported method (Park et al., 1992; Mattison et al., 1995). A solution of 0.2 g/l sodium citrate and 0.2 g/l chitosan was prepared at pH 1.0. Titrant (0.01–0.2 M NaOH) was delivered with a microburette into the solution with gentle stirring at 20 ± 0.2°C, and the pH was monitored by a digital pH meter with a precision of ± 0.01. Changes in turbidity were monitored at 420 nm with an UV–vis spectrophotometer and reported as $100 - \%T$ which is linearly proportional to the true turbidity for $T > 0.9$. The time interval between turbidity measurements was ca. 4 min.

coating of heparin and pectin only retarded drug release slightly. The significant difference of citrate/chitosan film swelling and model drug release profiles in SIF and SGF indicates that these films may be useful for site-specific drug delivery in the stomach.

Acknowledgements

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References

Al-Musa, S., Fara, D.A., Badwan, A.A., 1999. Evaluation of parameters involved in preparation and release of drug loaded in cross-linked matrices of alginate. J. Control. Release 57, 223–232.

Angelova, N., Manolova, N., Rashkov, I., Maximova, V., Bogdanova, S., Domard, A., 1995. Preparation and properties of modified chitosan films for drug release. J. Bioact. Compat. Polym. 10, 285–298.

Arguelles-Monal, W., Garciga, M., Peniche-Covas, C., 1990. Study of the stoichiometric polyelectrolyte complex between chitosan and carboxylmethyl cellulose. Polym. Bull. 23, 307–313.

Chandy, T., Sharma, C.P., 1993. Chitosan matrix for oral sustained delivery of ampicillin. Biomaterials 14, 939–944.

Chu, C.-H., Sakiyama, T., Yano, T., 1995. pH-sensitive swelling of a polyelectrolyte complex gel prepared from xanthan and chitosan. Biosci. Biotech. Biochem. 59, 717–719.

Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A., Smith, F., 1956. Colorimetric method for determination of sugars and related substances. Anal. Chem. 28, 350–356.

Gupta, K.C., Ravi Kumar, M.N.V., 2000. Drug release behaviors of beads and microgranules of chitosan. Biomaterials 21, 1115–1119.

Illum, L., 1998. Chitosan and its use as a pharmaceutical excipient. Pharm. Res. 15, 1326–1331.

Ikeda, S., Kumagai, H., Sakiyama, T., Chu, C.-H., Nakamura, K., 1995. Method for analyzing pH-sensitive swelling of amphoteric hydrogels — application to a polyelectrolyte complex gel prepared from xanthan and chitosan. Biosci. Biotech. Biochem. 59, 1422–1427.

Inouye, K., Machida, Y., Sannan, T., Nagai, T., 1988. Buoyant sustained release tablets based on chitosan. Drug Des. Del. 2, 165–175.

Kihuchi, Y., Noda, A., 1976. Polyelectrolyte complexes of heparin with chitosan. J. Appl. Polym. Sci. 20, 2561–2563.

Macleod, G.S., Collett, J.H., Fell, J.T., 1999. The potential use of mixed films of pectin, chitosan and HPMC for bimodal drug release. J. Control. Release 58, 303–310.

Mattison, K.W., Brittain, I.J., Dubin, P.L., 1995. Protein-polyelectrolyte phase boundaries. Biotechnol. Prog. 11, 632–637.

Nakatsuka, S., Andrady, A.L., 1992. Permeability of vitamin B-12 in chitosan membranes. Effect of cross-linking and blending with poly(vinyl alcohol) on permeability. J. Appl. Polym. Sci. 44, 17–28.

Park, J.M., Muhoberac, B.B., Dubin, P.L., Xia, J., 1992. Effects of protein charge heterogeneity in protein-polyelectrolyte complexation. Macromolecules 25, 290–295.

Patel, V.R., Amiji, M.M., 1996. Preparation of characterization of freeze-dried chitosan–poly(ethylene oxide) hydrogels for site-specific antibiotic delivery in the stomach. Pharm. Res. 13, 588–593.

Remuñán-López, C., Bodmeier, R., 1997. Mechanical, water uptake and permeability properties of cross-linked chitosan glutamate and alginate films. J. Control. Release 44, 215–225.

Shu, X.Z., Zhu, K.J., 2000a. A novel approach to prepare tripolyphosphate/chitosan complex beads for controlled release drug delivery. Int. J. Pharm. 201, 51–58.

Shu, X.Z., Zhu, K.J., 2000b. Chitosan/gelatin microspheres prepared by modified emulsification and ionotropic gelation. J. Microencapsulation, in press.

Thacharodi, D., Rao, K.P., 1993. Propranolol hydrochloride release behavior of cross-linked chitosan membranes. J. Chem. Tech. Biotechnol. 58, 177–181.

Thu, B., Bruheim, P., Espevik, T., Smidsrød, O., Soon-Shiong, P., Skjåk-Bræk, G., 1997. Alginate polycation microcapsules II. Some functional properties. Biomaterials 17, 1069–1079.

Yalpani, M., Hall, L.D., 1984. Some chemical and analytical aspects of polysaccharide modification. 3. Formation of branched-chain, soluble chitosan derivatives. Macromolecules 17, 272–279.

Yao, K.D., Peng, T., Goosen, M.F.A., Min, J.M., He, Y.Y., 1993. pH-sensitivity of hydrogel based on complex-forming chitosan: polyether interpenetrating polymer network. J. Appl. Polym. Sci. 48, 343–354.

Yao, K.D., Peng, T., Yin, Y.J., Xu, M.X., 1995. Microcapsules/microspheres related to chitosan. J.M.S.-REV. Macromol. Chem. Phys. C35, 155–180.

Yao, K.D., Liu, J., Cheng, G.X., Lu, X.D., Tu, H.L., 1996. Swelling behavior of pectin/chitosan complex films. J. Appl. Polym. Sci. 60, 279–283.

Yoshihisa, T., Yoshioka, I., Segi, N., Ikeda, K., 1991. Acid-induced and calcium-induced gelation of alginic acid: bead formation and pH-dependent swelling. Chem. Pharm. Bull. 39, 1072–1074.

However, only the coating of alginate prolonged riboflavin release significantly, which indicated that there were possibly other reasons but not electrostatic interaction resulting in the above result. It was reported that the aqueous solubility of alginate under acidic conditions was very poor (Yoshihisa et al., 1991), though in SGF the salt-bonds between alginate and chitosan dissociated, the precipitated alginate layer was still kept on the surface of citrate/chitosan film, which may limit film swelling and prolong drug release.

3.3.5. Model drug nature

The model drug properties, especially solubility, affected their release behavior from citrate/chitosan film seriously. Fig. 12 shows the loading percent and loss percent of brilliant blue, riboflavin, theophylline and 5-FU during cross-linking process. No obvious loss of brilliant blue and riboflavin occurred because they slightly dissolved in water, and even taking 4 h for cross-linking the loading percents were both larger than 97. But for more water-soluble and also smaller molecular weight theophylline and 5-FU, the loading efficiency decreased greatly with cross-linking time, and the loading efficiencies were both less than 20% in 2.5 h.

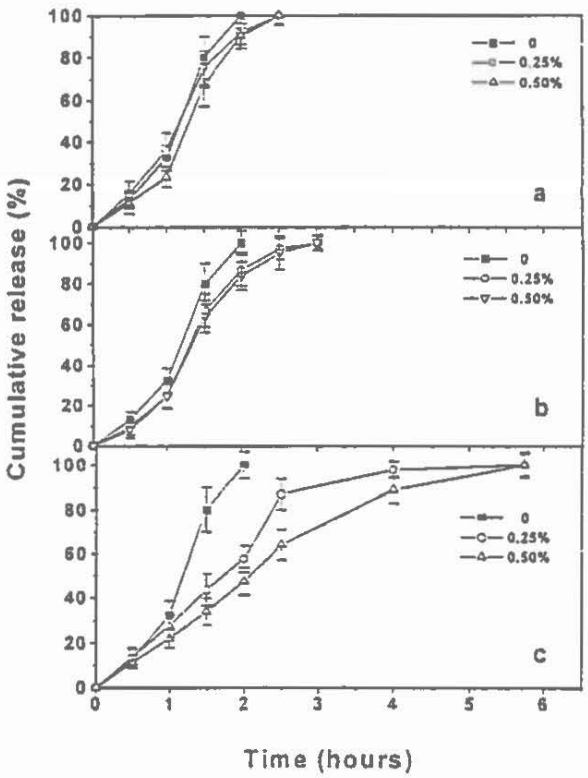

Fig. 11. The release of riboflavin from polyanion coating citrate/chitosan film. The films were prepared with 5.0% w/v sodium citrate (pH 7.0) and a cross-linking time of 1 h, and then dipped into polyanion solutions (pH 5.5) with different concentrations (w/v) for 15 min, (a) pectin, (b) heparin, and (c) sodium alginate.

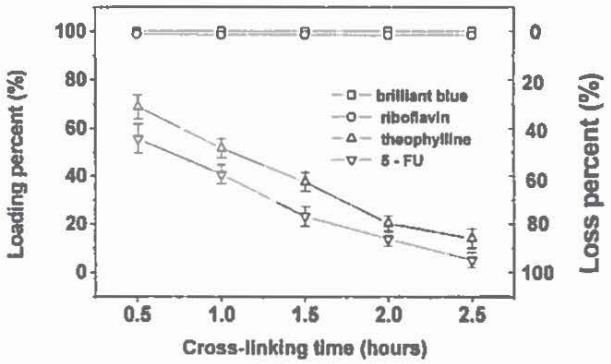

Fig. 12. The loading percent and loss percent of model drugs during cross-linking process, 5.0% w/v sodium citrate, pH 7.0.

The release of theophylline and 5-FU from citrate/chitosan film in SIF was very quick, in most cases more than 90% drug released in 2 h, while under the same condition the release percents of brilliant blue and riboflavin were both less than 5%.

4. Conclusions

Novel citrate cross-linked chitosan film was prepared by dipping chitosan film into citrate solution. Citrate/chitosan film possessed pH-sensitive swelling and drug controlled release properties. Sodium chloride weakened ionic cross-linking and facilitated film swelling and model drug release. Sodium citrate solution concentration and pH during cross-linking process affected film swelling and drug controlled release profiles, and using higher concentration and lower pH of sodium citrate resulted in less swelling and slower drug release. The further coating of alginate on the surface of citrate/chitosan prolonged riboflavin release in SGF significantly, while the

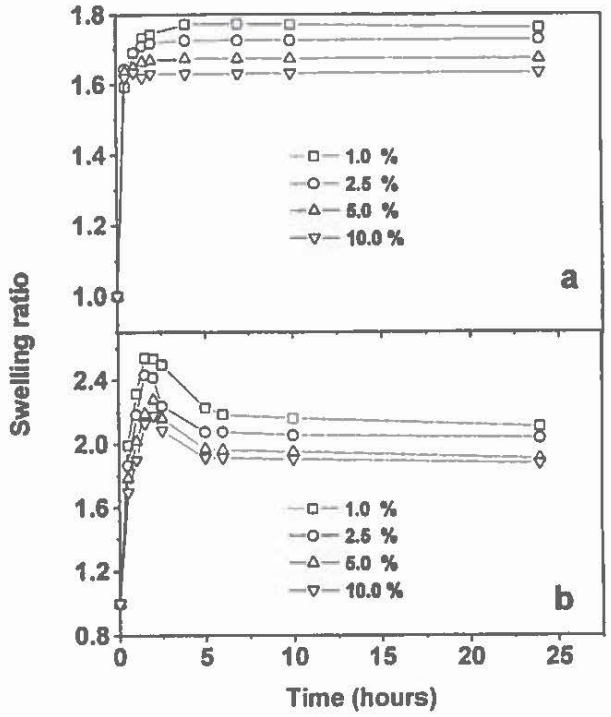

Fig. 8. The influence of sodium citrate concentration on the swelling of blank citrate/chitosan film in distilled water (a) and SIF (b) (cross-linking time 1.0 h, pH 7.0).

3.3.4. Polyanion coating

In low pH (1.0–3.5), citrate/chitosan film usually dissociated and the model drug released quickly (Figs. 5 and 6). For example, in SGF the release of riboflavin was usually completed within 2 h. To prolong the drug release from citrate/chitosan film in SGF, polyanions were further coated on the surface of citrate/chitosan film. However, the coating of pectin and heparin only slightly retarded riboflavin release in SGF (Fig. 11a and b). On the other hand, the coating of alginate greatly prolonged riboflavin release; the time period for 80% riboflavin released was extended from 1.5 to 2.4 and 3.5 h after being coated with 0.25 and 0.50% alginate (w/v), respectively.

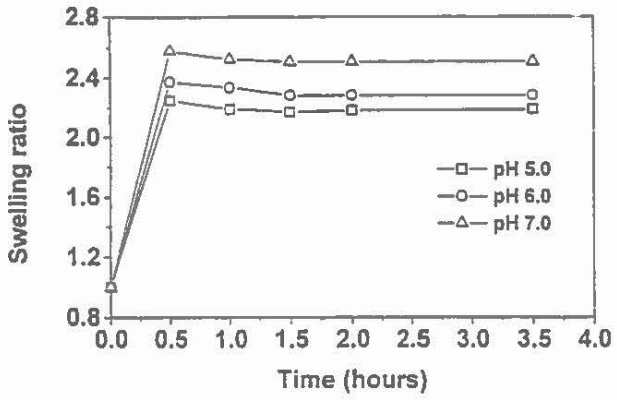

Fig. 9. The swelling curves of blank chitosan film in sodium citrate solution (5.0% w/v) with different solution pHs during cross-linking process.

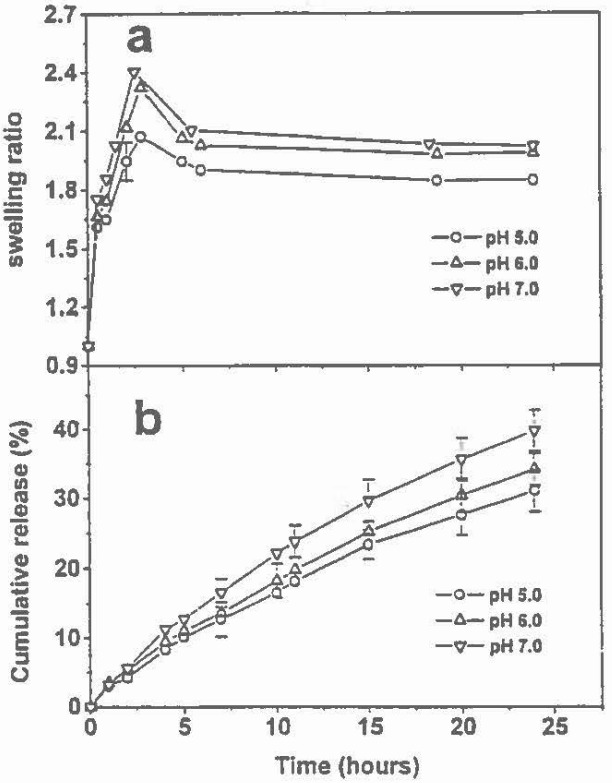

Fig. 10. The swelling of blank citrate/chitosan film (a) and the release of riboflavin from citrate/chitosan film (b) in SIF. The film was prepared with sodium citrate 5.0% w/v (pH 5.0, 6.0 or 7.0) and a cross-linking time of 1.0 h.

From the point of polyelectrolyte interaction, the coating of heparin should retard drug release in SGF most effectively, because the interaction between heparin and chitosan was the strongest due to the highest charge density of heparin (carboxylic and sulfonic groups) (Kihuchi and Noda, 1976). As for pectin and alginate, the weakly acidic carboxyl groups protonated in SGF (pH 1.0–1.1), and the electrostatic attractive force between pectin (or alginate) and chitosan disappeared (Macleod et al., 1999; Yao et al., 1996).

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