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Studies of cross-linking reaction on chitosan fiber with glyoxal

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Abstract

Taking glyoxal as a reagent for cross-linking to chitosan fiber to improve latter's mechanical properties, this paper studied the effects from the preparation of glyoxal solution, e.g. the concentration and pH, and the reaction condition, e.g. time and temperature. Results showed that the glyoxal solution prepared with a concentration of about 4% with pH at 4 to be better, while the reaction condition to be better at about 40 °C and 60–70 min. Based on comparison of IR spectra for chitosan fiber before and after cross-linking, the enhancement of the mechanical property for chitosan fiber has been found due mainly to the occurrence of acetalization and Schiff base reaction among the glyoxal and the hydroxyls and amino groups of chitosan. Of them, the Schiff base reaction seems to be of a dominator comparing to the former with respect to their IR behaviors observed.

Keywords: Chitosan fiber; Cross-linking; Glyoxal; Acetalization reaction; Schiff base reaction

1. Introduction

During the past decades, chitosan has got great attention and been broadly applied in medical and other areas with respect to its excellent biological characteristics of unique antiphlogistic effect, bio-compatibility, absorptivity, non-hypersensitivity, biodegradation property and wound healing (Chen, Liau, & Tsai, 1998; Kweon, Kang, & Chung, 1998; Okamoto et al., 1995; Tsai & Su, 1999). As known, chitosan has been currently used as scaffold for hepatocyte attachment (Yagi et al., 1997), bacterial antigens (Lillo & Matsuhiro, 1997), antimicrobial finishing reagents (Chung, Lee, & Kim, 1998; Chung, Schmidt, Hamlyn, Sagar, & Andrews, 1998) and capsules (Tozaki et al., 1999). Because chitosan has been also found available to accelerate the healing of human wounds (Chung, Lee, & Kim, 1998; Chung, Schmidt et al., 1998; Denuziere, Ferrier, Damour, & Domard, 1998; Koide, 1998; Kratz, Back, Arnander, & Larm, 1998; Le, Anand, & Horrocks, 1997; Pikiel & Kopczewski, 1998), it was thus observed that this natural polymeric material has been thus converted into fiber as reported elsewhere (East & Qin, 1993). Obviously, this is an interest because fiber can play a role different to other forms. However, it is noted that chitosan fiber seems to be low in its strength, especially in wet statue (East & Qin, 1993). To overcome this shortage, some efforts have been therefore tried. For example, East and Qin (1993) have tried to acetylate chitosan fibers to enhance its mechanical properties, and Knaul, Hooper, Chanyi, and Creber (1998) and Knaul, Hudson, and Creber (1999) have applied two methods, i.e. one is the drying and another is the cross-linking, to improve the mechanical properties for chitosan fiber. According to these authors, an aqueous solution containing glutaraldehyde and glyoxal may be available to enhance the mechanical properties for chitosan fiber.

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Noted, Jegal and Lee (1999), Moon, Pal, and Huang (1999) and Shin and Ueda (1999) have tried to use aldehyde as reagent to cross-link chitin, chitosan fibers and membranes.

Since a high strength chitosan fiber is expected by medical area for applying to different cases such as bone settling, human body tissue, medical paper, wound dressing, anti-fungus fabric textile and others, the aim of this work is proposed to enhance the mechanical properties for chitosan fiber using the cross-linking technique. In addition to Knaul et al. (1999), in this case we are able to employ glyoxal as a cross-linking reagent to enhance the mechanical properties for chitosan fiber, and meanwhile to understand the mechanism related. This is because glyoxal has been assumed may have steric effect on the network structure of cross-linked fiber in comparing to glutardialdehyde to yield tight supermolecular structure with higher crystallinity.

2. Experiments

2.1. Materials

Powder chitosan was supplied by Shanghai Tian-Qing Biomaterial Company as received. According to company analysis, its viscosity is of about 625 mPa S and its degree of deacetylation is 91.2%.

A glyoxal solution (30%) was employed in this case as cross-linking reagent, it was purchased from Shanghai Chemical Reagent Company as received without further purification.

2.2. Preparation of chitosan fiber

The chitosan fiber was prepared initially by dissolution of the chitosan powder 3.5 wt% in an acetic acid solution. Then the spinning solution was filtered and flowed into a spinneret to form a filament by controlling the flow rate using a metering pump under a nitrogen pressure condition. After that, the fiber was coagulated in coagulating bath comprising NaOH and Na_2SO_4 and drawn in a drawing bath. Finally, the drawn fiber was washed and dried. This wet-spinning process was also recently described in detail recently (Wang, Shen, & Gu, in press).

2.3. Cross-linking chitosan fiber

To avoid cross-linking process resulting in shrinking for fiber, the prepared chitosan fiber was initially reeled in a 'U' type glass rod then immersed in glyoxal solution to start the cross-linking reaction. Since cross-linking reaction is considerable to be influenced by many factors, a set of experiments were designed by taking the tenacity as a unique evaluation target and four vary parameters, e.g. time, temperature, pH and concentration of reagent.

After cross-linking reaction, sample fiber was washed by distilled water for several times then dried in an oven condition, e.g. 50 °C for 16 h.

2.4. Fiber analysis and characterization

The tenacity of fibers was measured using a XQ-1 type fiber strength-elongation testing machine as the same as Wang et al. (in press) described. In order to know the cross-linking reaction how to influence chitosan fiber, the same measurement was also conducted for an uncross-linked sample. For instance, the tenacity for original chitosan fiber was found to be of about 1.10 cN/dtex.

FTIR spectra of fibers were recorded using a Nicolet 520 FTIR spectrophotometer from 400 to 4000 cm^{-1}.

3. Results and discussion

3.1. FTIR spectroscopy investigation of possible mechanism for glyoxal cross-linking to chitosan fiber

Though dialdehyde compounds, e.g. glutaraldehyde and glyoxal, have been extensively used as cross-linking reagent for chitosan (Hsien & Rorrer, 1995; Knaul et al., 1999), cellulose (Yagi et al., 1998) and starch (Ivanov et al., 1992), it is truly that the mechanism on these reagents bond to chitosan is still not fully understood. Considering the halobiosic chitosan has a structure with two hydroxyl groups and an amino group in one glucosamine ring (Fig. 1(a)), and the glyoxal molecule has two carbonyl groups (Fig. 1(b)), especially the carbon of carbonyl groups due to located in sp^2 easily hybridized to cause attachment from other three atoms lying in the same plane to yield the bond angles among these three atoms to form a trigonal coplanar structure, e.g. approximately 120° (Fig. 1(c)). Moreover, the carbonyl carbon and carbonyl oxygen are available to present several positive and negative charges influenced by the electronegative oxygen and the resonance contribution of the second structure (Fig. 1(d)). To observe these evidences is thus of interest and probably in relation to understanding of the mechanism for using glyoxal to cross-link chitosan.

FTIR spectra for chitosan fiber before and after cross-linking were presented in Fig. 2. Observed there has a new peak appeared at 1110 cm^{-1} for chitosan fiber after cross-linking reaction. This indicates that glyoxals have reacted with the hydroxyls of the glucosamine rings due to acetalization and in good agreement with Solomons (1980). According to Solomons (1980), dissolution of aldehyde in alcohol can yield hemiacetal by nucleophilic addition of the alcohol to the carbonyl group, and this process has a basic feature on hemiacetal, i.e. its –OH and –OR groups to be attached by the same carbon atom. Moreover, as known from Solomons (1980) that hemiacetal is of unstable and it is easily further isolated in

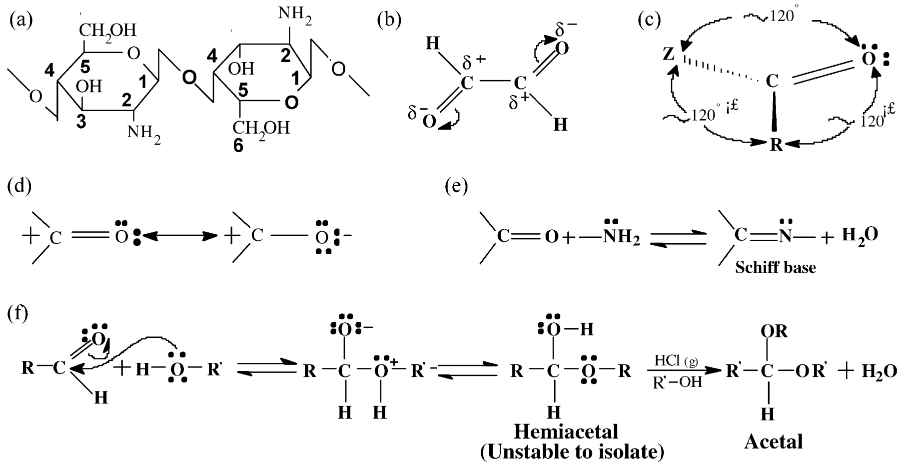

Fig. 1. Possible reaction routs for glyoxal cross-linking to chitosan fiber. (a) Chitosan, (b) Glyoxal, (c) Carbonyl, (d) Resonance structure for the carbonyl group, (e) Schiff base reaction and (f) Acetalization reaction.

the presence of catalysts, e.g. gaseous HCl, to cause it reacts with the alcohol again to result in an acetal with two –OR groups attached by the same CH group as Fig. 1(f) described.

Because carbonyl group can also reacted with Schiff base in theory due to the presence of amino group adding to aldehyde in resulting of imines (Fig. 1(e)) (Pine, Hendrickson, Cram, & Hammono, 1980). Assuming this reaction occurrence in this case, to observe its effect on chitosan fiber is thus another interest and should be appeared in IR spectrum. As expected, since Fig. 2 presented a quite strong peak observed to locate at 1666 cm^{-1} assigning to C=N stretching, this is availably taken as an evidence for supporting expected evidence as described in Fig. 3(b). This is in good agreement with Knaul et al. (1999). Since these two organic reactions have been showed with visible larger or smaller peaks, e.g. the larger for Schiff base and the smaller for acetalization, it is also considered that the Schiff base reaction might be a domination in this case as comparing to another. On this conclusion, though it seems to be not in agreement with general knowledge on Schiff base unstable (Solomons, 1980), it is, however, perhaps possible because Solomons (1980) indicated that Schiff base unstable in solution while our sample showed in Fig. 2 was in solid and that might be in relation to enamines with covalent structure or reform (Roberts, 1992). In fact, since the IR spectrum (Fig. 2) was obtained for sample after cross-linking from a solution with pH 4 where the Schiff base has been found strongly taken place (Pine et al., 1980), the obtained solid sample presented obviously Schiff base structure is probably.

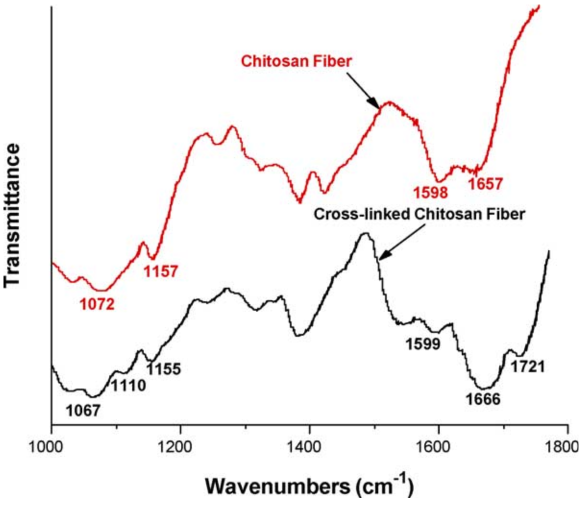

Fig. 2. FTIR spectra of chitosan fiber before and after cross-linking.

Based on above discussion, the possible mechanism for glyoxal cross-linked to chitosan fiber was furthermore outlined in Fig. 3.

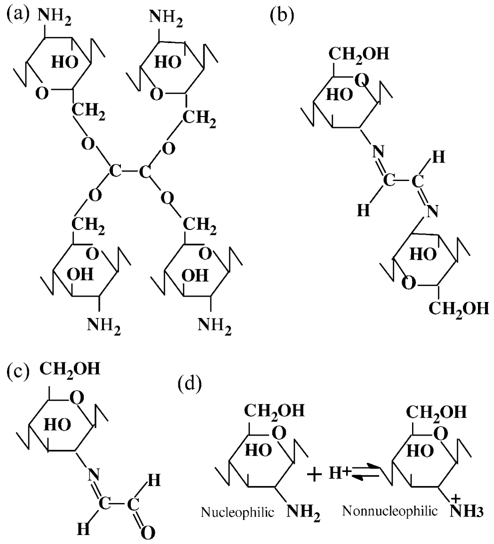

Fig. 3. A description of the mechanism for glyoxal cross-linking to chitosan fiber. (a) Glyoxal reacts with hydroxyl groups in chitosan, (b) Glyoxal reacts with amino groups in chitosan, (c) one carbonyl in a glyoxal reacts, (d) Effect of acid on chitosan nucleophilicity.

Table 1
An analysis of chitosan fiber cross-linked with glyoxal based on FTIR spectra presented in Fig. 2

Groups	Vibration mode	Uncrosslinked chitosan fiber		Crosslinked chitosan fiber	
		Peak (cm^{-1})	Intensity	Peak (cm^{-1})	Intensity
N=C–C=O	Stretching	–	–	1721	Weak
C=N	Stretching	–	–	1666	Very strong
–NH_2	Scissor	1598	General	1599	General
C–O–C–O–C	Asymmetrical stretching	–	–	1110	Weak

Based on Dolphin and Wick (1977), Fig. 2 presented peaks were also assigned and summarized in Table 1.

3.2. *Effects on glyoxal cross-linking with chitosan fiber*

By varying the concentration of reagent from 1 to 8% and taking the tenacity as a function, Fig. 4 presented several curves. Of them, temperature was also varied in the range of 25–60 °C. Since Fig. 4 showed that each curve presented a unique peak and all these peaks corresponding to a critical concentration of about 4%, it is in reasonable to consider that this critical concentration may be better for using glyoxal as reagent for cross-linking to chitosan fiber. From Fig. 4, it is also evidenced that the better temperature for mentioned case to be of about 40 °C. Based on Fig. 4 in relation to Fig. 3 outlined mechanism for mentioned case, it is thus further considered that the role of glyoxal molecules played is to interlink chitosan at its backbone of the carbonyls to cause two or more chitosan macromolecules joining together to form a network molecule caused the increase of the molecular weight (Knaul et al., 1999).

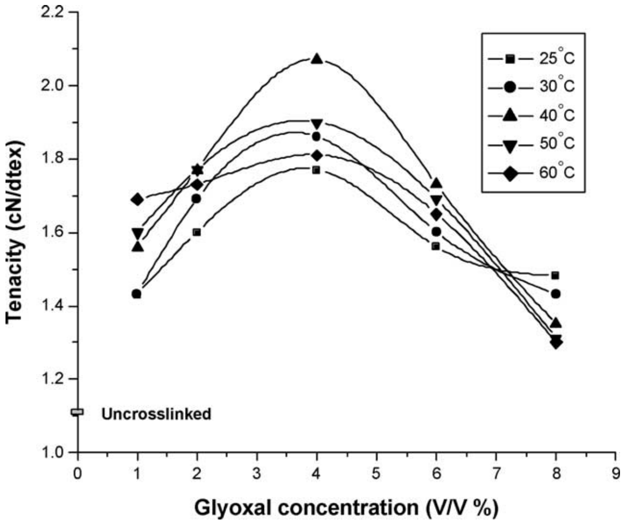

Fig. 4. Effect of the variety of glyoxal concentration, the pH was kept at about 3.73, on the tenacity of cross-linked chitosan fiber under different reaction temperatures and after reaction of about 30 min.

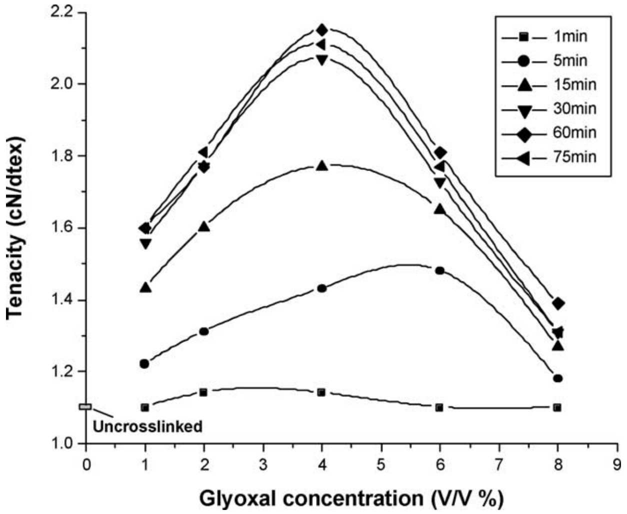

Fig. 5. Effect of glyoxal concentration, the pH was kept at about 3.73, on the tenacity of cross-linked chitosan fiber under different reaction times and fixed temperature, e.g. about 40 °C.

Of interest the result obtained in Fig. 4 was furthermore observed and certified in Fig. 5. For example, to take tenacity and reaction time as functions, the same plot showed in Fig. 5 again indicated that there has a critical concentration for glyoxal solution. However, this figure suggested that reaction time required for this case to be better at about 60–75 min. On the basis of Figs. 4 and 5, the reason for understanding the presented critical concentration, e.g. about 4%, for glyoxal could be explained as that the lower or the higher of the concentration of glyoxal both impossible for acceptation is due to the former unable to causing function groups to cross-link to chitosan to form net leading to increase the tenacity while the latter, however, may cause redundant glyoxal molecules and to lost

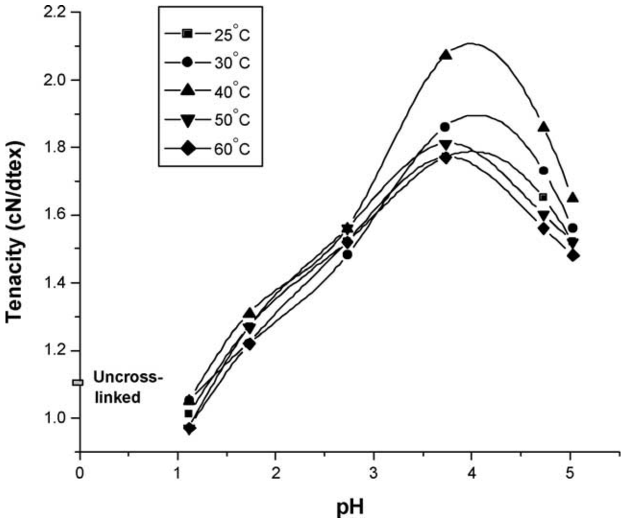

Fig. 6. Effect of the variety of pH for glyoxal solution on the tenacity of cross-linked chitosan fiber under a reaction condition, e.g. the temperature of about 40 °C, glyoxal concentration about 4% and reaction time about 30 min.

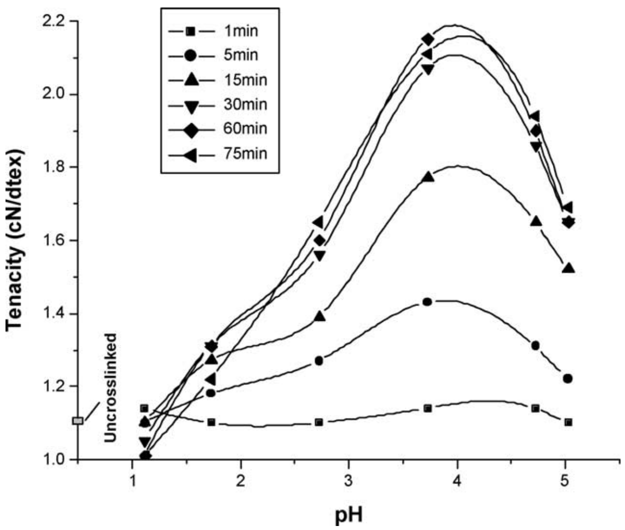

Fig. 7. Effect of the variety of pH for glyoxal solution on the tenacity of cross-linked chitosan fiber under different reaction times with fixed glyoxal concentration, e.g. of about 4%, and reaction temperature, e.g. of about 40 °C.

opportunity to bond to the chitosan glucosamine rings as expected to increase the mechanical property.

In general, pH is an important factor in this case because the pKa value of chitosan has been known to be of 6.5 (Strand, Tømmeraas, Vårum, & Østgaard, 2001) while the pH of solution in the range of 3–5 has been also known available to enhance Schiff's base reaction (Pine et al., 1980). The effect from solution's pH is thus subsequently showed in both Figs. 6 and 7. Observed, the tenacity of cross-linked chitosan fiber is gradually increased with the increase of pH until to 4, then suddenly reduced. This suggests that the peak related pH to be better for applying to cross-linking process. Because the whole pH used are in an acidic range, and the strongest tenacity observed corresponding to pH 4, it is, therefore, considered that an acidic environment may suit for cross-linking to chitosan reaction occurrence. The reason for glyoxal cross-linking to chitosan better in this pH is assumed to be as IR spectra showed (Fig. 2) due to the Schiff base reaction taking place in the presence of acid catalysis (Pine et al., 1980). This can also be understood from Figs. 1 and 3 because the primary amine located at the backbone of chitosan is nucleophilic owing to its unshared pair of electrons on nitrogen. Therefore, once the acidity of the reaction solution increased, the amine could be soon protonated to present non-nucleophilic property (Fig. 3(d)) to cause the carbonyl group lost chance to take place of addition reaction to yield the low tenacity for chitosan fiber as Figs. 6 and 7 showed. From these Figs. 6 and 7, the same phenomena as that of Figs. 4 and 5 were observed again, e.g. the better temperature and time for cross-linking reaction to be at 40 °C and 60–70 min. In general, these values are in reasonable because a high temperature usually causes degradation taking place for polymer material especially chitosan. On the understanding of the reaction time from Fig. 7, it is also probably because the increase of the mechanical properties for chitosan fiber using cross-linking method must be obtained until reaction stabilized whereas it sis time dependence.

4. Conclusion

This study indicated that the mechanical properties of chitosan fiber could be enhanced using glyoxal as reagent and the cross-linking method, and of this process the effects are basically from the preparation of the cross-linking solution, e.g. the concentration and pH, and reaction condition, e.g. temperature and time.

Based on IR spectra presented tracks, the possible mechanism for using glyoxal to cross-linking to chitosan fiber to enhance the mechanical property is found to be due to the occurrence of acetalization and Schiff base reaction, i.e. probably taking place among the glyoxal and the hydroxyls and amino groups of chitosan.

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