

## Synthesis, Characterization, and Film Properties of Crosslinked Chitosan with Pentaerythritol Tris[3-(1-aziridinyl) propionate]

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**ABSTRACT:** The novel crosslinked chitosan derivative (CMCSCL) has been synthesized via carboxymethyl chitosan with pentaerythritol tris[3-(1-aziridinyl)propionate] (PEN) as crosslinking reagent. Its structure has been proved by Fourier transform infrared spectroscopy, X-ray diffraction, differential scanning calorimetry, thermogravimetric analysis and atomic force micrograph. The mechanical properties and water uptake of CMCSCL films have been measured. The results show that CMCSCL film with 4% PEN crosslinker has a high tensile strength (TS) 17.1 MPa, 13.1% elongation at break, and 99% water uptake. The film with 6% PEN has a TS 15.2 MPa, 10.2% elongation at break, and 111% water uptake. The dosage of the crosslinking agent is the principal factor affecting the mechanical properties and water uptake of these films. © 2012 Wiley Periodicals, Inc. *J. Appl. Polym. Sci.* 000: 000–000, 2012

**KEYWORDS:** chitosan; crosslinking; films; properties; characterization

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### INTRODUCTION

Chitin is the second most important natural polymer in the world after cellulose. The main exploited sources are two marine crustaceans, shrimp and crabs. Chitosan is deacetylated derivative of chitin by treatment with hot alkali.<sup>1–4</sup> The commercial or practical use of chitin and chitosan (including monomer and oligomers) has been confined to the unmodified forms. As a breakthrough in utilization, chemical modification to chitosan will be a key step.<sup>5–8</sup>

Crosslinking is one of kinds of chemical modifications for chitosan that can lead to the formation of a permanent covalent network and reinforce the properties of chitosan, such as chemical resistance, water resistance, mechanical and physical properties, and can form functional gel.<sup>9–12</sup> Chitosan can be crosslinked by some crosslinking agents such as glutaraldehyde, epichlorohydrin, and divalent calcium salts, to enhance properties.<sup>13,14</sup> The usual method of preparing hydrogels is also by chemically crosslinking amide groups with glutaraldehyde or other suitable crosslinker. However, some crosslinkers are highly toxic, or inactive, or hydrophobic.

Aziridine can easily react with a nucleophilic hydroxyl or carboxyl group in aqueous-based system. The reaction can occur immediately at ambient temperature, which provides a convenient alternative for preparing a highly crosslinked network in aqueous system.<sup>15–17</sup> To our knowledge, there is a lack of data

regarding chitosan film crosslinked with pentaerythritol tris[3-(1-aziridinyl)]-propionate (PEN).

In this paper carboxymethyl chitosan (CMCS) with certain degree of substitution was synthesized, then crosslinked with PEN to form the product abbreviated as CMCSCL. The CMCSCL structure has been characterized by Fourier transform infrared spectroscopy (FTIR), X-ray diffraction, differential scanning calorimetry (DSC), thermogravimetric analysis (TGA), atomic force micrograph, and chemical analysis. The water uptake and mechanical properties have also been measured.

### EXPERIMENTAL

#### Materials

Chitosan with medium molecular weight, ethanoic acid, and PEN were supplied by Aldrich. D<sub>2</sub>O<sub>s</sub> were supplied by Aldrich and Cambridge. The deacetylation degree of chitosan measured by 400 MHz <sup>1</sup>H NMR is 0.85.<sup>18</sup> The degree of substitution (DS) of CMCS measured by potential titration<sup>19</sup> is 0.574. All solutions were prepared in deionized water.

#### Instrumentation

FTIR was obtained on a Mattson Genesis Series FTIR instrument (Madison, WI).

The wide angle X-ray diffraction patterns were recorded at room temperature by an X-ray diffractometer (Rigaku 12 kW Rotaflex) using Cu K $\alpha$  radiation. The X-ray generator was

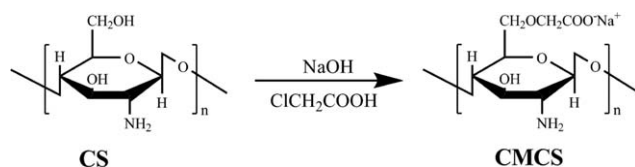


Figure 1. Schematic illustration for the synthesis of CMCS.

operated at 40 kV and 150 mA. The dried membranes of uniform thickness were mounted on aluminum frames and scanned from 2 to  $40^{\circ}2\theta$  at a speed of  $5^{\circ}2\theta/\text{min}$  and a chart speed of 2 cm/min.

DSC measurements were carried out on a Q1000 thermal analysis system (TA Instruments) at a heating rate of  $10^{\circ}\text{C}/\text{min}$  under the nitrogen atmosphere. The heat flow and temperature of DSC were calibrated with a standard material (indium) before measurements. The samples were dried under reduced pressure in a vacuum oven at room temperature for 2 days. Accurately weighed dry material (6–8 mg) was placed in an aluminum cup and hermetically sealed. Empty cup was used as reference.

Thermal stability was investigated in a TA 9900 thermal analyzer system from TA Instruments. Specimens accurately weighing 6–8 mg were subjected to thermal scan at a scan rate of  $10^{\circ}\text{C}/\text{min}$  in nitrogen gas.

Tensile properties were measured according to ASTM D 2370-92 using Instron 5567. Tensile measurements were made on rectangular specimens 8–9 mm wide, 0.06–0.08 mm thick, and with gauge length of 20 mm. A crosshead speed of 2 mm/min was applied to determine tensile strength (TS), and elongation at break. For each film, five samples were tested, and the average is reported.

AFM was performed with a DI multiMode scanning SPM in a tapping mode with a silicon tip. The samples used for film casting in aqueous solution were spinner coated on glass plates.

## EXPERIMENTAL

### Preparation of Carboxymethylchitosan (CMCS)<sup>20</sup>

Carboxymethylchitosan (CMCS) was synthesized under the following steps. Chitosan powder (5 g) was suspended in 40 mL propanol, in which 10 mL of 16.25 M sodium hydroxide solution was added like concentrated porridge. After stirring for 30 min at room temperature, the suspension was frozen at  $-20^{\circ}\text{C}$  for 24 h. Then 7.5 g of chloroacetic acid in 10 mL isopropanol was added in parts. The mixture was kept stirring at  $20^{\circ}\text{C}$  for 1 h then for another 4 h at  $50^{\circ}\text{C}$ . After dialyzing and vacuum dried, CMCS was obtained. DS of the product CMCS was estimated from potential titrations as 0.574.<sup>19</sup> As shown in Figure 1, it is for the synthesis of CMCS.

### Preparation of Crosslinked Chitosan (CMCSCL)

CMCS of 2.5 g was added into 6 mL deionized water. Then 2% PEN is dropped into the solution stepwise and stirred for 24 h, after vacuum dried for 24 h, extracted 24 h and vacuum dried, using methanol as solvent and dialyzing 24 h, the white solid was obtained for IR measurement.

### Preparation of CMCSCL Films

CMCS of 2.5 g was added into 6 mL deionized water. Then 2, 4, 6, and 8% (w/w) PEN to CMCS were added into the solu-

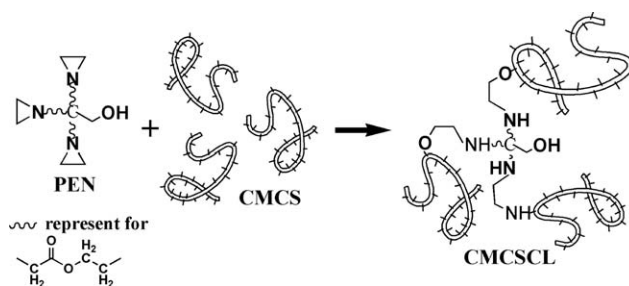


Figure 2. Schematic illustration for CMCSCL formation.

tion. Pour the solution onto a glass plate. After naturally drying 48 h, the films of CMCSCL can be peeled for measurement. The temperature in lab is  $24^{\circ}\text{C}$  and the degree of humidity is 85%. Figure 2 shows the schematic illustration for CMCSCL formation.

### Water Uptake of CMCSCL Film

The water uptake behavior of CMCSCL film was measured by soaping the films in deionized water at room temperature. Each sample (surface area approximately  $1\text{ cm}^2$ ) was weighed and then submerged into deionized water in a 250 mL beaker covered with parafilm to avoid evaporation. After 24 h, the film was withdrawn and its wet weights was determined after blotting with a filter paper followed by blowing with a stream of air to remove the surface water and immediately weighing the film. Average of three trials was taken. The water uptake was calculated using eq. (1).

$$\text{Wateruptake (\%)} = [(W_s - W_d)/W_d] \times 100 \quad (1)$$

where, water uptake is the percentage water absorption of sample,  $W_d$  and  $W_s$  are sample mass in the dry and swollen states, respectively.

## RESULTS AND DISCUSSION

### FTIR Spectra of CMCSCL

Figure 3 displays the formula of PEN and the mechanism of ring opening reaction of CMCS with PEN. The remaining hydroxyl and amino groups of CMCS have an open ring reaction with the aziridinyl groups of PEN.

Figure 4 presents FTIR spectra of CS, CMCS (DS 0.574), and CMCSCL. Compared with chitosan, CMCS shows disappearance of the  $\text{NH}_2$ - associate band at  $1650\text{ cm}^{-1}$ , which can be ascribed to primary amine N—H vibration deformation after carboxymethylation.<sup>21–23</sup> In the spectrum of CMCS, the C—O stretching band at  $999\text{ cm}^{-1}$  corresponding to the primary hydroxyl group decreases greatly, verifying a high reaction of OH-6 after chitosan carboxymethylation. Two peaks at  $1584$  and  $1400\text{ cm}^{-1}$  in CMCS and  $1587$  and  $1403\text{ cm}^{-1}$  in CMCSCL are observed due to the asymmetrical and symmetrical stretching of  $\text{COO}^-$  group.<sup>24,25</sup> The characteristic peak of second hydroxyl group at  $1023\text{ cm}^{-1}$  of chitosan has shifted to  $1045\text{ cm}^{-1}$  for CMCS. New peak appears at  $1740\text{ cm}^{-1}$  which indicates formation of a new ester carbonyl stretching in CMCSCL. No aziridinyl peaks at  $981$  and  $1310\text{ cm}^{-1}$  in CMCSCL also prove the ring open reaction between CMCS and PEN.<sup>17</sup>

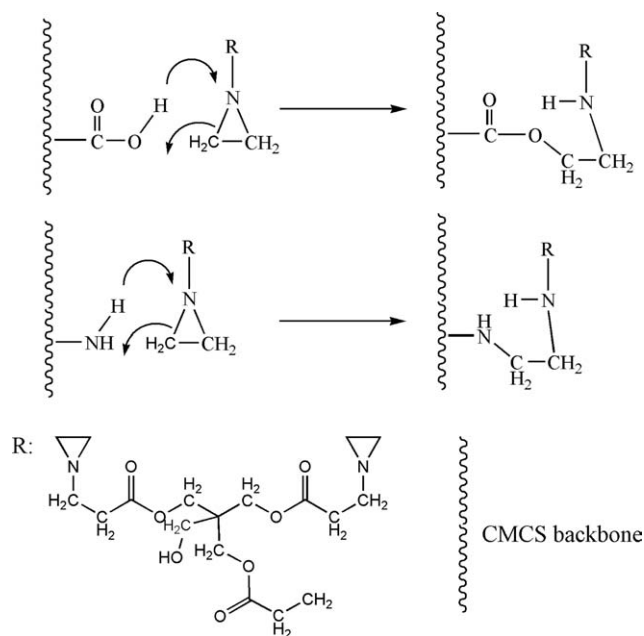


Figure 3. Mechanism of ring open reaction of CMCS by PEN.

### Wide Angle X-Ray Diffraction Analysis

The X-ray diffraction patterns of CMCSCL and CMCS are shown in Figure 5. It could be seen that there were some differences of peak height, width, and position between them.

CMCS consists of one major peaks at  $2\theta = 20^\circ$ . Compared with CMCS, CMCSCL has a relatively broader peak at  $2\theta = 20^\circ$ . It is well-known that the width of X-ray diffraction peak is related to the size of crystallite, the broadened peak usually results from small crystallites.<sup>26,27</sup> Hence, in this reaction, the crosslinking reaction first took place preferentially in the amorphous region and then proceeded very moderately from the edge to inside of the crystalline region and with further reaction with PEN, original crystalline structure of CMCS was destroyed and the crystallinity decreased.

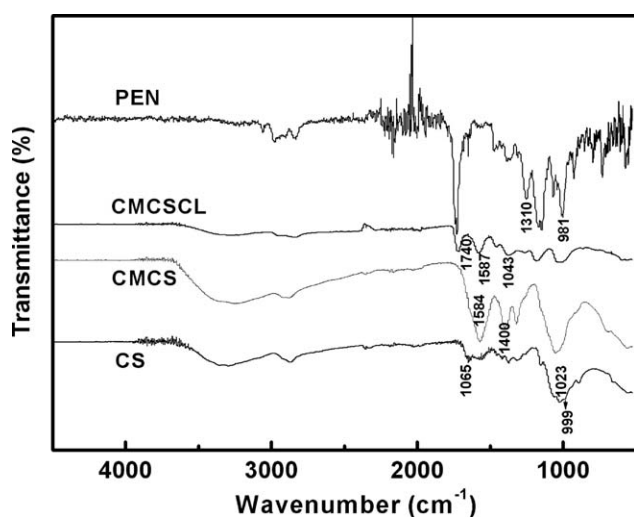


Figure 4. FTIR spectra of CS, CMCS, and CMCSCL.

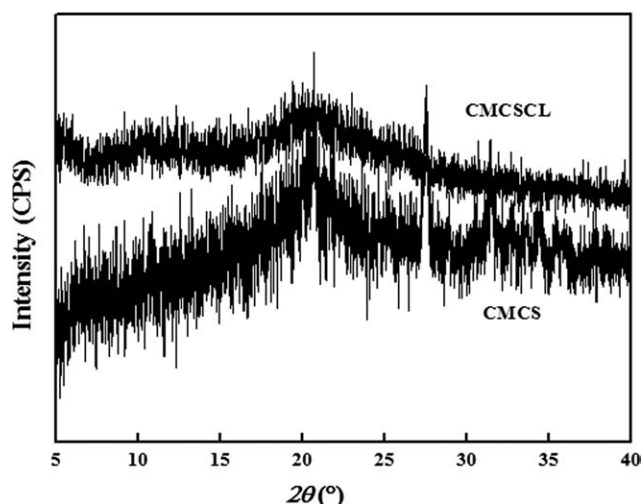


Figure 5. X-ray diffraction patterns of CS, CMCS, and CMCSCL (2%) film.

### DSC

DSC thermograms of CMCS and CMCSCL are shown in Figure 6. The thermal properties of chitosan and its derivatives are related to their structure.<sup>28</sup> As is known, polysaccharides have strong affinity for water. The hydration properties of these polysaccharides depend on the primary and supra-macromolecular structures, and the overall polymorphic nature of the macromolecule also has a great influence on the presence of bound water.<sup>29</sup> It is necessary that the samples should be dried completely to eliminate the effect of moisture.

The DSC peak area and position of sample CMCSCL are very different from CMCS in the different stages. Glass transition temperatures are not observed both for CMCS and CMCSCL. The thermogram of CMCS shows an endothermic peak at around  $165^\circ\text{C}$  owing to the dissociation of hydrogen bonds with the degradation of CMCS at about  $270^\circ\text{C}$ . The peak of dissociation of hydrogen bonds of CMCS in CMCSCL matrix is shifted to about  $175^\circ\text{C}$  because of the influence of stronger bonding in CMCSCL matrix. In contrast, CMCSCL also exhibits an endothermic peak at around  $209^\circ\text{C}$  due to the harder dissociation of hydrogen bond with the degradation of CMCSCL at

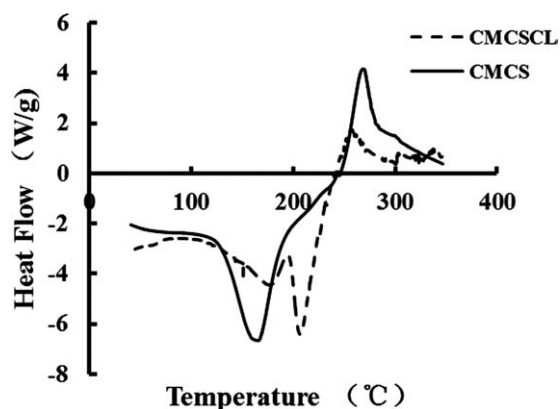
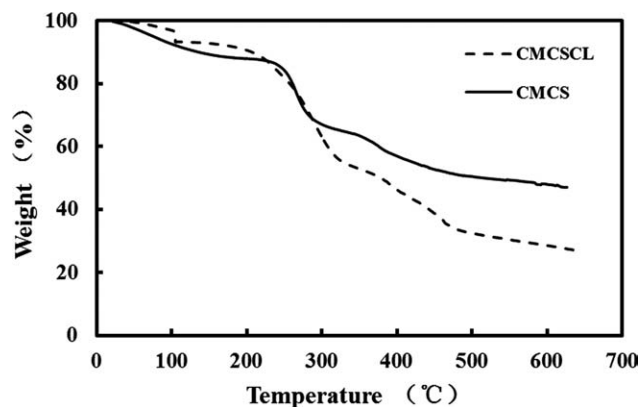


Figure 6. DSC thermograph for CMCS and CMCSCL.



**Figure 7.** TGA thermograph for CMCS (DS = 0.574) and CMCSCL (2%) film.

about 255°C. This can be supported by X-Ray results and TGA thermogram as well.

### Thermogravimetric Analysis

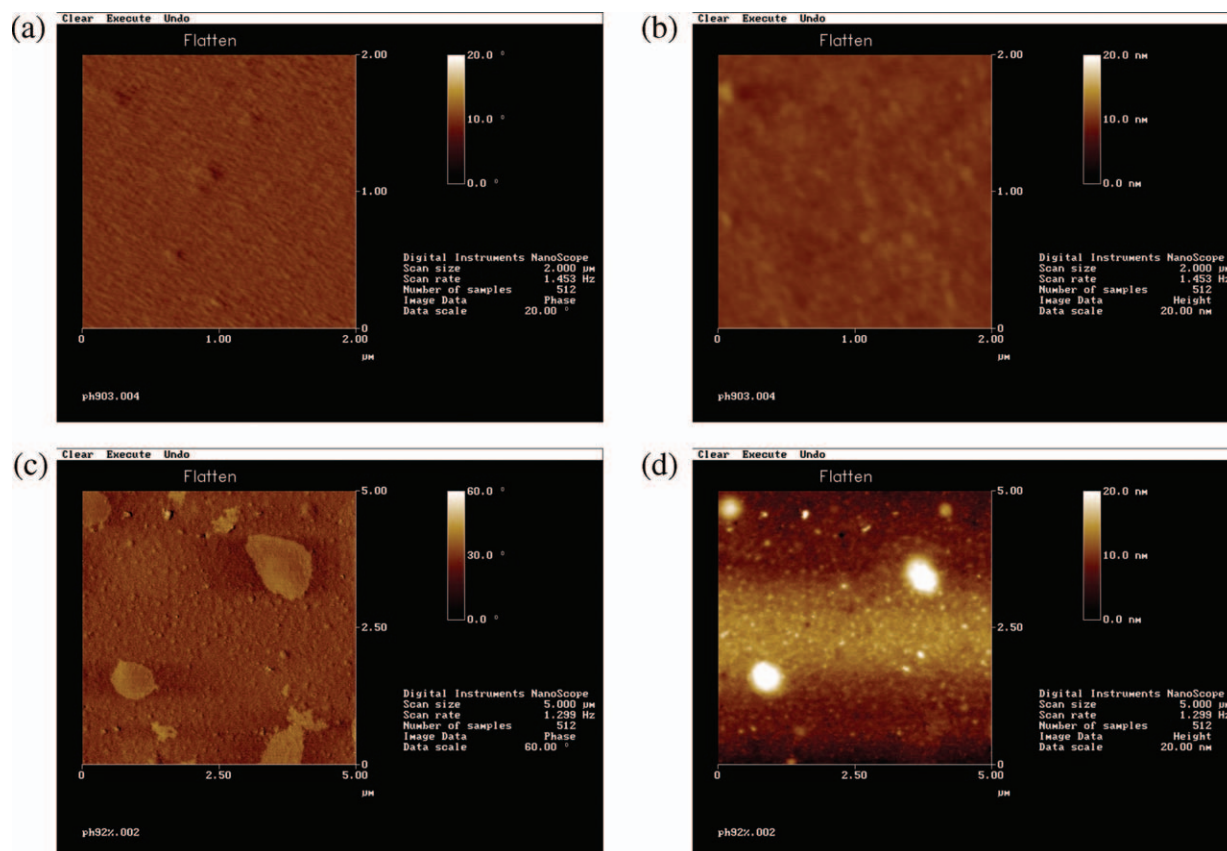
TGA of CMCS and CMCSCL (2%) is shown in Figure 7. The TGA of CMCS shows a weight loss in two stages. The first stage ranges between 30 and 105°C and shows 8% loss in weight. This may be corresponding to the loss of adsorbed and bound water. The second stage of weight loss starts at about 265°C and that continues up to 315°C during which there was 22% of

weight loss due to degradation of CMCS, whereas TGA of CMCSCL is different from its. It is observed that the latter has two stages of distinct weight loss between 20 and 330°C. The first stage ranges between 20 and 105°C with 7% of the absorbed and bound water weight loss. The second stage of weight loss starts at around 245°C that continues up to 330°C during which there was 34% of weight loss due to degradation of residual CMCS and CMCSCL.

### Atomic Force Microscopy

Tapping-mode AFM images yielded information about the surface features of CMCSCL film. Figure 8 shows the surface morphology of crosslinked CMCSCL and CMCS. Figure 8(a, b) show phase image and height image of CMCS film. The main surface topography is homogeneous, continuous, and relatively flat, without any pore or crack with good structural integrity and sparsely distributed small particles having peak heights of between 17 and 18 nm without any phase separation.

From AFM topographic images (c) and (d) of the film, particles of CMCSCL are distributed into the matrix of CMCS. Both phase contrast and slight phase separation were observed in the phase image. The second phase is the assembling structure of CMCSCL in agreement with X-ray, DSC, and TGA, which is like an island distributing into the sea and compatible. This textile structure benefits to the properties' improvement of film.



**Figure 8.** AFM topographic images of (a) CMCS film's phase; (b) CMCS film's height; (c) CMCSCL (2% PEN) film's phase; (d) CMCSCL (2% PEN) film's height. [Color figure can be viewed in the online issue, which is available at [wileyonlinelibrary.com](http://wileyonlinelibrary.com).]



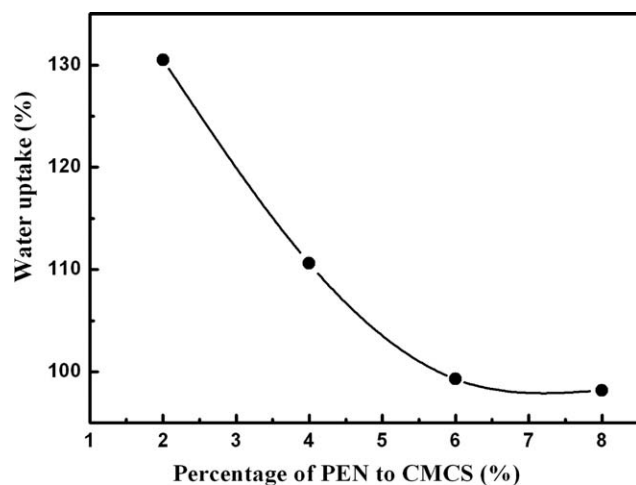


Figure 9. Water uptake dependence of PEN percentage in CMCSCL films.

The film height is between 8 and 18 nm. The particle height is about 18–20 nm.

#### Water Uptake of CMCSCL

From Figure 9, water uptake of CMCSCL reaches a minimum value at 8% of PEN, and it decreases with increasing PEN usage. For example, for percentages of 4 and 6%, water uptake of the resulting CMCSCL decreases from 111 to 99%, respectively. However, increasing the amount of PEN to an equivalent percentage does not decrease water uptake of CMCSCL, because a highly crosslinked network structure results in less free chain mobility, which limits further crosslinking reaction at ambient temperature.<sup>30</sup>

#### Mechanical Properties

Table I shows mechanical properties of CMCSCL films against PEN percentage. The TS values of the films with the different PEN percentage are also shown in Table I.

TS is at 9.94 MPa with 8.83% elongation at break of original CMCS and its value of cured CMCS increases first with the increasing curing dosage of PEN, due to the formation of crosslinking network. Typically, TS changes from original 9.94 MPa at 8.33% elongation at break to 17.1 MPa at 13.1% elongation for 4% curing system. However, that of CMCS with 6% PEN becomes 15.2 MPa at 10.2% elongation, which may be due to overcrosslinking between the two main components.

Table I. Mechanical Properties of CMCSCL Film as a Function of PEN Percentage

PEN content (%)	TS (MPa)	Elongation at break (%)
0	9.94	8.33
2.0	14.0	9.24
4.0	17.1	13.1
6.0	15.2	10.2

#### CONCLUSIONS

A new crosslinked chitosan derivative (CMCSCL) has been synthesized by crosslinking reaction between CMCS and pentaerythritol tris[3-aziridinyl] propionate (PEN) at ambitious temperature. Its structure has been proved by FTIR, X-ray, thermal analysis, and AFM. CMCSCL films have also been prepared. Their mechanical properties and water uptake properties have been measured. The results display that TS value increased first with increasing curing dosage of PEN, due to the formation of crosslinking network. However, to certain dosage of PEN, TS values decreased due to the post crosslinking between the two main components. The water uptake decreased with increasing PEN usage. However, increasing the amount of PEN to an equivalent percentage did not decrease water uptake of CMCSCL. The new derivatives could be expected to have application in coating and film area.

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