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Preparation and Characterization of Dual Sensitive Carboxylated Methyl Cellulose/Poly(vinyl alcohol) Physical Composite Hydrogel

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ABSTRACT: Neutral methyl cellulose (MC) was transformed into carboxylated methyl cellulose (CLMC) via the esterification reaction between MC and maleic anhydride. FTIR, X-ray diffraction, and thermogravimetry analysis results indicated the carboxyl groups were successfully incorporated onto the chains of MC. The carboxyl group percentages of CLMC ranged from 6.5 to 13.6%. CLMC and poly(vinyl alcohol) were mixed in aqueous solution, and then formed physical composite hydrogel. The equilibrium swelling ratios of the hydrogels in buffer solutions of pH 10 and 1.5 were 10.7 and 7.5, 7.7 and 6.0, 6.5 and 5.2 when the ratio of CLMC/PVA was 5 : 5, 4 : 6, and 3 : 7, respectively. The hydrogel exhibited both temperature- and pH-sensitive swelling properties, which implied not only the inherent temperature sensitive property of MC was retained but also one more function was acquired. The maximum cumulative release percentages of Rhodamine B from the CLMC/PVA (3 : 7) hydrogel in the mediums of pH 7.4 and 1.2 were 43.36 and 37.44%, respectively. Such a pH-responsive release behavior also confirmed that CLMC was anionic. © 2012 Wiley Periodicals, Inc. J. Appl. Polym. Sci. 000: 000–000, 2012

KEYWORDS: methyl cellulose; carboxylation; dual sensitive; physical; hydrogel

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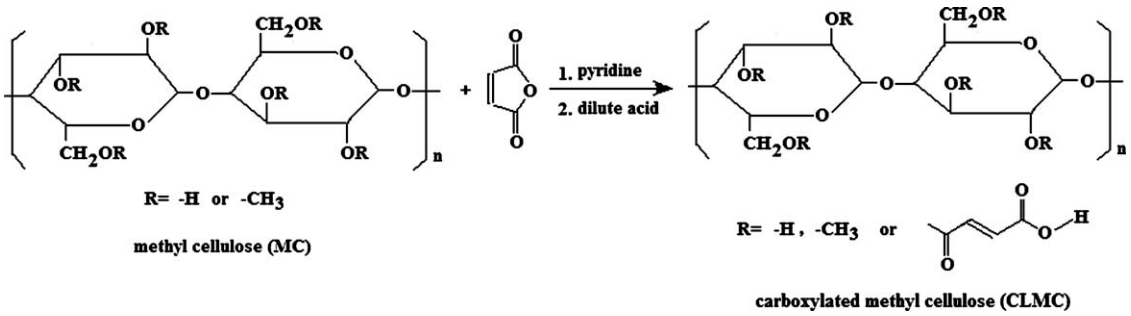
INTRODUCTION

Cellulose is the most abundant natural polysaccharide on earth. It is renewable, inexpensive, and biodegradable.[1] Cellulose is a linear biopolymer composed of D-glucose units. The high crystalline ability, strong inter- and intra-molecular interactions make cellulose rigid and poor soluble in common solvents.[2] Therefore, cellulose is usually modified before application. Cellulose acetate, methyl cellulose (MC), sodium carboxymethyl cellulose (NaCMC), and hydroxypropyl cellulose (HPC) are several major derivatives of cellulose.[3,4] Owing to the excellent properties such as water solubility, good processability, and biocompatibility,[5] they have been used widely in many areas such as food industry and biomedical fields.[6,7]

Among the derivatives of cellulose, MC is a kind of methyl ether. Portion of the hydroxyl groups on the glucose units are substituted with methoxyl groups.[8] Owing to the intermolecular hydrophobic interaction, the aqueous solution of MC exhibits temperature-sensitive sol-gel transition.[9] In addition, MC is neutral and has not any characteristics of polyelectrolyte.

As known, polyelectrolyte possesses many advantages such as forming polyelectrolyte complex with opposite ions and responding to the environmental pH or ionic strength change. Such physical procedures are mild and attractive for pharmaceutical and biomedical applications.[10,11] On the other hand, hydrogel contains a lot of water. It resembles the human tissues. Smart hydrogels, which are sensitive to more than one environmental stimulus, have been paid attention for decades. Physically formed intelligent hydrogels are regarded as good candidates as biomedical and biotechnological materials.[12] Polyelectrolytes are often chosen to prepare responsive hydrogels.

Poly(vinyl alcohol) (PVA) is a water-soluble and biocompatible synthetic polymer. By taking advantage of its capability of forming physical hydrogel through mild repeated freezing/thawing cycles, MC/PVA hydrogels with tunable thermosensitivity have been prepared in our lab.[13] In viewing of the hydroxyl groups on MC chains are active sites[6,14] for further chemical modification, we intend to incorporate new functional groups onto MC, which endows MC novel properties for meeting potential requirements. A facile way to functionalize MC, i.e., esterification reaction between MC and maleic anhydride, is conducted in this article. To the best of our knowledge, no similar work to incorporate carboxyl groups onto the chains of MC has been reported. Then the carboxylated MC is mixed with PVA in aqueous solution and subsequently formed composite hydrogels by freezing/thawing technique. The swelling and *in vitro* release behaviors of the hydrogels have been investigated in different mediums. The experimental results indicate that the functionalized MC is sensitive to temperature and pH as well, which proves our goal is feasible.



Scheme 1. The carboxylation of methyl cellulose via esterification between methyl cellulose and maleic anhydride.

EXPERIMENTAL

Materials

MC was purchased from Sinopharm Chemical Regent, China. Its average degree of substitution (DS), viscosity average molecular weight, and viscosity for a 2% in (w/v) aqueous solution at 20°C were 2.01, 2.2×10^5 and 350–550 cps, respectively. PVA was kindly donated by Fujian Chemical Fiber and Chemical Factory, China. Its viscosity average molecular weight and degree of hydrolysis were about 6.0×10^4 and 99%, respectively. Both PVA and MC were used without further purification. Maleic anhydride (MA, m.p. 54–55°C) was purified by recrystallization from benzene. Pyridine, absolute ethanol, hydrochloric acid (HCl), sodium hydroxide (NaOH) and *N*, *N*′-dimethylformamide (DMF) were all purchased from Shanghai Chemical Agents, China. Rhodamine B was purchased from Aladdin reagent. All of these reagents were analytical grade reagents and used as received.

Synthesis of Carboxylated Methyl Cellulose

CLMC was prepared according to the Ref. 15 by the esterification between MC and MA. Briefly, 6 g MC was dissolved in 120 mL DMF and transferred into a 250 mL three-necked flask. A predetermined amount of MA and pyridine (the molar ratio of MA and pyridine was 1 : 1) were dissolved in 15 mL DMF, respectively, mixed carefully and added into the flask in droplet under stirring. The mixture was allowed to react for 24 h with agitation at 30°C, and then acidified with 1 *M* HCl till the pH value of the solution was around 2. The crude product was precipitated from 200 mL anhydrous ethanol and purified by extracting with ethanol in a Soxhlet apparatus for 48 h. The dried yellow powder was pure CLMC.

Characterization of CLMC

The carboxyl group percentage (CGP) of CLMC was determined by titration[16] and calculated as CGP (%) = $(V_{NaOH} \times C_{NaOH} - V_{HCl} \times C_{HCl}) \times 45 \times 100/m$, where *C* and *V* were the concentration and volume of NaOH and HCl, and *m* was the weight of CLMC, respectively. The DS of CLMC was calculated as $M1 \times A/(1 - M2 \times A)$, where M1 was the molecular weight of the repeat unit of MC ($C_6H_7O_5(CH_3)_{DS}H_{DS+3}$, the average DS of MC is 2.01, thus M1 was approximately equal to 190), M2 was the molecular weight of COCH=CHCOOH moiety (M2 = 99), and A was the mole number of NaOH consumed by per gram CLMC sample [A = CGP/(45 × 100)].

The transparent films were obtained by casting 10 mL 0.010 g/mL CLMC aqueous solutions onto polypropylene plates and being dried at 50°C. The powdered MC was mixed with dry KBr and compressed into disk. Fourier transform infrared (FTIR) spectra of the samples were directly recorded with a Nexus 470 FTIR spectrophotometer.

X-ray diffraction (XRD) profiles of MC and CLMC powders were collected with a Bruker D8-Advanced diffractometer using Nickel-filtered Cu-Kα radiation ($\lambda = 0.15406$ nm) and scanned from 2° to 60° at a scan speed of 4°/min.

Thermogravimetric analysis (TGA) of CLMC was performed with a Shimadzu DTG-60H thermoanalyzer. Analyses were conducted over the temperature range from 25 to 900°C with a programmed temperature increment of 10°C/min using dry nitrogen purge at a flow rate of 50 mL/min.

CLMC samples containing different amount of carboxyl groups were dissolved in water to prepare solutions of 2 g/L. Sol-gel transition temperatures of CLMC solutions were determined over the temperature range from 50 to 80°C with a Shimadzu UV2450 UV-visible spectrophotometer that equipped with a temperature control accessory.

Preparation of CLMC/PVA Composite Hydrogels

CLMC samples of 6.5% carboxyl groups were mixed with PVA in the ratio of 3 : 7, 4 : 6, and 5 : 5 (w/w). One gram of each mixture was dissolved in 15 mL deionized water, respectively. The aqueous solutions were subjected to three freezing/thawing cycles, 16 h at −16°C and 6 h at ambient temperature, respectively. The formed complex hydrogels were maintained at 40°C until the weight of the samples was constant.

Dual Responsive Behaviors of CLMC/PVA Composite Hydrogel

To examine the pH-sensitive swelling property, the dried CLMC/PVA hydrogels were weighed and placed in vials that contained HCl–KCl (pH 1.5) and Na_2CO_3-$NaHCO_3$ (pH 10) buffer solutions, respectively. The ionic strength of both solutions was 0.2 *M*. The samples were kept at 25°C and removed at time intervals, blotted up the surface liquid of the samples with soft paper and weighed till the weights were constant. The swelling ratios (SR) of the samples were calculated from the weight of sample at different time (W_t) and the weight of dried sample (W_d): $SR = W_t/W_d$. CLMC/PVA hydrogels that contained 30 wt% CLMC were respectively maintained in buffer solutions of pH 1.5 and 10 for 24 h. Then the Au-coated cross-sections of freezing-dried CLMC/PVA hydrogels were examined with a Hitachi S-3500N scanning electron microscope (SEM).

Table I. Molecular Characteristics of Carboxylated Methyl Cellulose

Sample	Feeding ratio of reagents (repeat unit of MC/MA, mol/mol)[a]	Carboxyl group percentage (CGP, %)[b]	Average degree of substitution	Intrinsic viscosity (dL/g)[c]
MC	–	0	2.01	0.57
CLMC-1	1 : 1.5	6.5	0.27	0.76
CLMC-2	1 : 2	8.6	0.36	0.87
CLMC-3	1 : 3	13.6	0.58	1.07

[a]The carboxylation of MC was conducted at 30°C for 24 h by using pyridine as catalyst.
[b]It was determined by titration.
[c]It was measured with an Ubbelohde viscometer by using distilled water as solvent at 25°C.

To determine its reversible thermo-responsive property, the dried hydrogels were immersed in the phosphate-buffer saline (PBS, 0.1 *M*, pH 7.4), and the temperature was alternated between 37 and 70°C for three cycles. The swelling ratios of the samples were obtained as aforementioned.

The responsive properties of the hydrogels were further verified with examining the *in vitro* release behavior by using Rhodamine B as model drug. Fifty milligram Rhodamine B was mixed with CLMC and PVA (the ratio of CLMC/PVA in weight was 3 : 7, the total weight of CLMC and PVA was 1 g). The mixture was dissolved in 15 mL deionized water, and formed hrdrogels via three freezing/thawing cycles. The Rhodamine B encapsulated samples were slowly parched at 37°C, weighed, placed in conical bottles that contained 25 mL buffer solutions (pH 7.4 or 1.2) and maintained at 37°C. At defined time intervals, 5 mL buffer solutions were removed to determine the amount of released Rhodamine B with UV2450 UV-visible spectrophotometer by measuring the absorption at 512 nm. Five milliliters fresh buffer solutions were added respectively in the meantime.

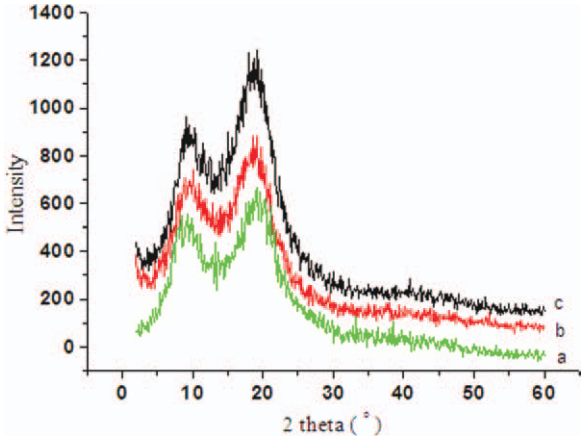


Figure 2. X-ray diffraction profiles of methyl cellulose (MC, a), the carboxylated methyl cellulose (CLMC) containing 8.6% (b), and 13.6% (c) carboxyl groups. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

RESULTS AND DISCUSSION

Synthesis of CLMC

Esterification is proved to be an effective way to transform the neutral polysaccharide into a polysaccharide-based anionic polyelectrolyte.[2,16] Accordingly, it is adopted to prepare MC-based anionic polyelectrolyte (Scheme 1). Such a carboxylated product has not been found in the literatures. As known, the esterification reaction between —OH and —COOH is reversible. In addition, the amount of free hydroxyl groups of MC is not abundant since DS of the starting MC is as high as 2.01. Consequently, it is able to anticipate that the content of —OCOCH=CHCOOH moieties incorporated onto MC chains is not high. As expected, it is found that the CGP and DS of CLMC are 6.5% and 0.27, 8.6% and 0.36, 13.6% and 0.58 as the feeding ratios (repeat unit of MC/MA, mol/mol) are 1 : 1.5, 1 : 2, and 1 : 3 (Table I), respectively. In other words, both the CGP and DS of CLMC are low but able to be varied with the

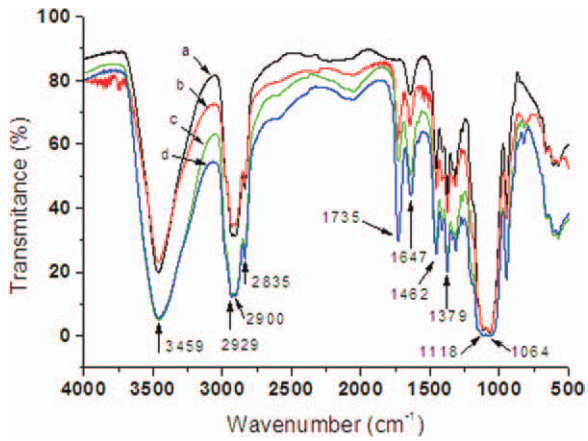


Figure 1. FTIR spectra of methyl cellulose (a) and the carboxylated methyl cellulose containing 6.5% (b, CLMC-1), 8.6% (c, CLMC-2), and 13.6% (d, CLMC-3) carboxyl groups. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

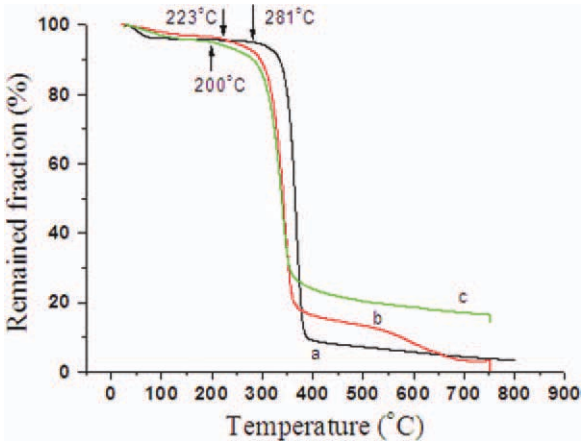


Figure 3. Thermogravimetric analysis profiles of methyl cellulose (MC, a), the carboxylated methyl cellulose (CLMC) containing 8.6% (b) and 13.6% (c) carboxyl groups. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

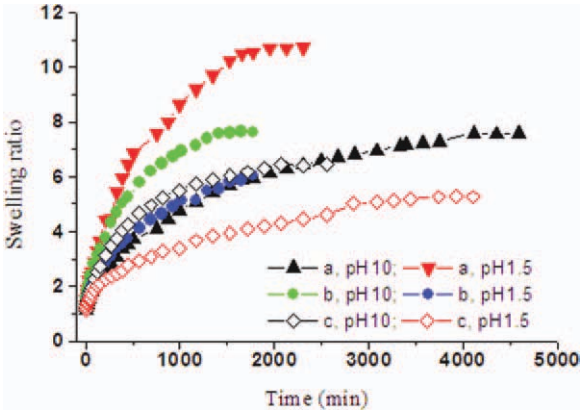


Figure 4. pH-sensitive swelling of CLMC/PVA complex hydrogels [the weight ratio of CLMC containing 6.5% carboxyl group and PVA: (a) 5 : 5; (b) 4 : 6; (c) 3 : 7)]. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

feeding ratio of MC/MA. Hereafter, we represent CLMC containing 6.5, 8.6, and 13.6% carboxyl groups as CLMC-1, CLMC-2, and CLMC-3, respectively.

Characterization of CLMC

The FTIR spectra of MC, CLMC-1, CLMC-2, and CLMC-3 are shown in Figure 1. As compared to the spectra of MC, one more characteristic absorption band appears at 1735 cm^{-1} on that of CLMC, which is attributed to the stretching vibration of carbonyl group. Moreover, the intensity of the peak appeared at 1735 cm^{-1} is increased with increasing the CGP of CLMC. These mean that the structure of CLMC is confirmed with FTIR. Two sharp peaks are exhibited around 9.7° and 18.8° on the XRD patterns of MC, CLMC-2, and CLMC-3 (Figure 2). Since the content of carboxyl groups is not high, the morphology of CLMC is similar to MC. In other words, CLMC is also semicrystalline. But the carboxylation has somewhat destroyed the steric regularity of MC chains, the crystallinity of CLMC is slightly lower than that of MC. The crystallinity of MC, CLMC-2, and CLMC-3, which is calculated from the crystalline peak area divided by whole peak area, are 47.6, 42.8, and 42.5%, respectively. This result is consistent with the thermal stability of MC and its derivative. Thermogravimetry analysis shows that the initial decomposition temperatures of MC, CLMC-2, and CLMC-3 are 281, 223, and 200°C (Figure 3), respectively. Evidently, higher carboxyl content decreases the crystallinity of CLMC and reduces its thermal stability more in turn. XRD and TGA measurements indicate that the carboxylation of MC is carried out once more.

To examine weather the intrinsic temperature-sensitivity of MC solution is retained after modification, the aqueous solutions of CLMC-1, CLMC-2, and CLMC-3 are subjected to heating. Meanwhile, the absorbency of the solutions is monitored by UV spectrophotometry. It is found that the transparent MC and CLMC solutions are respectively changed into turbid at different temperatures. The sol-gel transition temperatures of MC, CLMC-1, CLMC-2, and CLMC-3 are 71, 69, 66, and 63°C, respectively. As seen, higher CGP leads to conducting the sol-gel transition of CLMC solution at lower temperature. This phenomenon is probably attributed to the formation of inter- and intra-molecular hydrogen bonding between hydroxyl and carboxyl groups along the backbone of MC.[17] Anyhow, carboxylated MC does keep the temperature-sensitive characteristic of MC. In this regard, CLMC is superior to NaCMC. NaCMC is also an anionic derivative of cellulose, but it does not exhibit a low critical solution temperature in water.

Dual Sensitive Swelling Behaviors of CLMC/PVA Complex Hydrogel

To investigate the function and potential application of CLMC, we have prepared the physical CLMC/PVA hydrogels and then examined their swelling and release behaviors that respond to temperature and pH stimuli.

As shown in Figure 4, CLMC/PVA composite hydrogels that obtained from three ratios all show pH-sensitive swelling. The differences between the equilibrium swelling ratios of the hydrogels at pH 10 and 1.5 are 3.1, 1.8, and 1.2, respectively. Obviously, it is the protonation and deprotonation of carboxyl

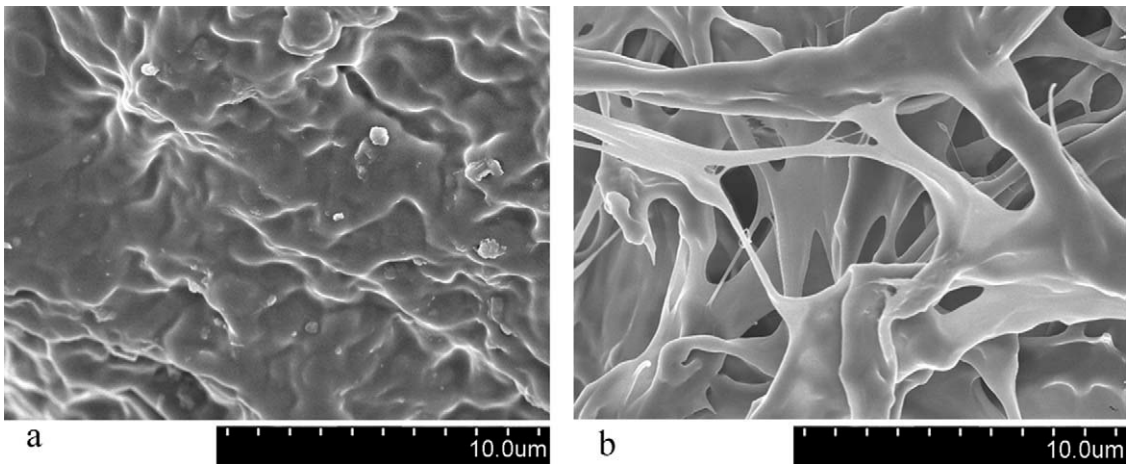


Figure 5. SEM images of the cross-sections of CLMC/PVA complex hydrogels (the weight ratio of CLMC containing 6.5% carboxyl group and PVA is 3 : 7) after maintaining in buffer solutions of pH 1.5 (a) and pH 10 (b) at 37°C, respectively.

groups that provide CLMC pH-sensitive swelling. At pH 1.5 and 10, most carboxyl groups exist in the form of —COOH and —COO^-, respectively. In other words, the electrostatic interactions are different in two situations. As a result, the hydrogels swell in different degree. This is verified with SEM analysis results. There are pores on the cross-section of CLMC/PVA hydrogel after swelling at pH 10, whereas no pore is found on that at pH 1.5 (Figure 5). In addition, more CLMC is mixed with PVA means that the composite hydrogel contains more carboxyl groups. Consequently, its pH-sensitive swelling behaves more evidently. On the other hand, —COOH is hydrophilic. Thus, more carboxyl groups enable CLMC/PVA hydrogel to reach higher equilibrium swelling ratio.

It is observed that the swelling of the CLMC/PVA composite hydrogel is reversible thermo-responsive. Being placed in buffer solution (pH 7.4) at 70°C, the hydrogels reach the equilibrium swelling. When the temperature is changed in the way of being cooled down to 37°C and subsequently heated to 70°C for three cycles, the swelling ratios of hydrogels will alternate accordingly (Figure 6). It is also found that the thermal-responsive phase transition relies on the amount of CLMC in the hydrogel. The maximum equilibrium swelling ratios for the hydrogels that contain 50 wt % CLMC and 40 wt % CLMC are 7.5 and 5.9, respectively. In addition, the time to finish three temperature-sensitive cycles for the samples that contain 40 and 50 wt % CLMC are 5530 and 7760 min, respectively. The reversible swelling–deswelling phenomenon reveals the composite hydrogels are stable enough, which can be attributed to the strong interaction, i.e., intermolecular hydrogen bonds, between carboxyl groups of CLMC and hydroxyl groups of PVA.[18]

Release Behavior of CLMC/PVA Complex Hydrogel

For the sake of exploring the application of CLMC as an anion polyelectrolyte, Rhodamine B is utilized as a model drug to examine the release behavior of the composite hydrogel. The *in vitro* release profiles of the model drug encapsulated hydrogels suggest that CLMC/PVA hydrogel is a good candidate as a carrier for drug controlled release. As shown in Figure 7, the maximum cumulative release percentages of Rhodamine B from

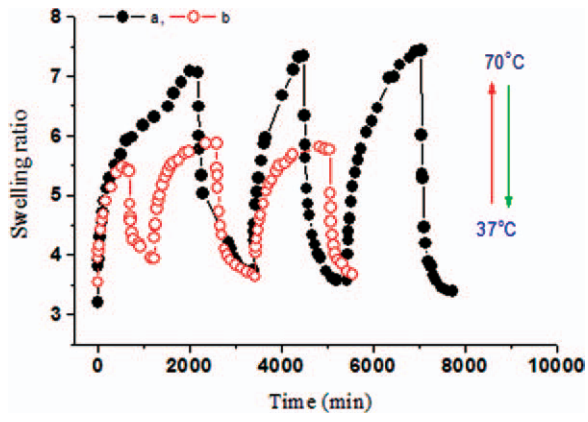


Figure 6. Temperature-sensitive reversible swelling of CLMC/PVA complex hydrogels (the weight ratios of CLMC containing 6.5% carboxyl group and PVA are: (a) 5 : 5 and (b) 4 : 6). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

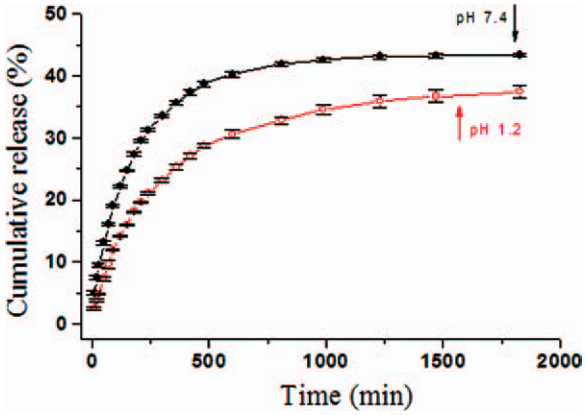


Figure 7. *In vitro* release of the CLMC/PVA hydrogel (the weight ratio of CLMC containing 6.5% carboxyl group and PVA is 3 : 7) that encapsulate Rhodamine B in buffer solutions of pH 1.2 and 7.4 at 37°C. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

the CLMC/PVA hydrogel are 43.36 and 37.44% in simulated body fluid (pH 7.4) and simulated gastric fluid (pH 1.2), respectively. Evidently, the release behavior is pH-sensitive as well. As Rhodamine B is a cationic electrolyte, the electrostatic interaction between opposite charged drug and CLMC may lead to immobilizing some Rhodamine B molecules tightly within the hydrogel. Thus, only around 40% Rhodamine B is released from the matrix after immersing in buffer solution for 30.5 h. In spite of this, the release analysis results suggest the CLMC/PVA composite hydrogel is probably suitable for sustained release of drug.[19]

CONCLUSIONS

The carboxylation of MC is successfully performed via esterification between MC and MA by using pyridine as a catalyst. The incorporation of carboxyl groups changes the structure and morphology of MC slightly. Therefore, the carboxylated MC still retain the thermal sensitivity. Moreover, it behaves as an anionic polyelectrolyte. The physically formed CLMC/PVA composite hydrogel exhibits pH-sensitive swelling and drug release behaviors. The CLMC/PVA complex hydrogels of different compositions show reversible thermal responsive swelling. The dual sensitive properties may provide CLMC as a material for more applications.

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REFERENCES

1. Stalling, S. S.; Akintoye, S. O.; Nicoll, S. B. *Acta Biomater.* **2009,** *5*, 1911.
2. Li, W. Y.; Jin, A. X.; Liu, C. F.; Sun, R. C.; Zhang, A. P.; Kennedy, J. F. *Carbohydr. Polym.* **2009,** *78*, 389.
3. Heinze, T.; Liebert, T. *Prog. Polym. Sci.* **2001,** *26*, 1689.

4. Song, Y. B.; Zhou, J. P.; Zhang, L. N.; Wu, X. J. *Carbohydr. Polym.* **2008,** *73*, 18.
5. Fettaka, M.; Issaadi, R.; Moulai-Mostefa, N.; Dez, I.; Le Cerf, D.; Picton, L. J. *Colloid Interface Sci.* **2011,** *357*, 372.
6. Barbucci, R.; Leone, G.; Monici, M.; Pantalone, D.; Fini, M.; Giardino, R. J. *Mater. Chem.* **2005,** *15*, 2234.
7. Kim, J.; Yun, S.; Ounaies, Z. *Macromolecules.* **2006,** *39*, 4202.
8. Patel, T. R.; Morris, G. A.; de la Torre, J. G.; Ortega, A.; Mischnick, P. Harding, S. E. *Macromol. Biosci.* **2008,** *8*, 1108.
9. Desbrières, J.; Hirrien, M.; Rinaudo, M. *Carbohydr. Polym.* **1998,** *37*, 145.
10. Dautzenberg, H. *Macromolecules.* **1997,** *30*, 7810.
11. Hartig, S. M.; Carlesso, G.; Davidson, J. M.; Prokop, A. *Biomacromolecules.* **2007,** *8*, 265.
12. Hendrickson, G. R.; Smith, M. H.; South, A. B.; Lyon, L. A. *Adv. Funct. Mater.* **2010,** *20*, 1697.
13. Xiao, C. M.; Geng, N. N. *Eur. Polym. J.* **2009,** *45*, 1086.
14. de Melo, J. C. P.; Filho, E. C. D. S.; Santana, S. A. A.; Airoldi, C. *Colloids Surf. A* **2009,** *346*, 138.
15. Lu, D. R.; Xiao, C. M.; Xu, S. J.; Ye, Y. F. *eXPRESS Polym. Lett.* **2011,** *5*, 535.
16. Xiao, C. M.; Tan, J.; Xue, G. N. *eXPRESS Polym. Lett.* **2010,** *4*, 9.
17. Nurkeeva, Z. S.; Mun, G. A.; Khutoryanskiy, V. V.; Mangazbaeva, R. A. *Polym. Int.* **2000,** *49*, 867.
18. Mc Gann, M. J.; Higginbotham, C. L.; Geever, L. M.; Nugent, M. J. D. *Int. J. Pharm.* **2009,** *372*, 154.
19. Maderuelo, C.; Zarzuelo, A.; Lanao, J. M. *J. Control. Release* **2011,** *154*, 2.