Control of Thermogelation Properties of Hydrophobically-Modified Methylcellulose

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ABSTRACT: Aqueous solutions that undergo reversible thermosensitive gelation around body temperature were developed based on hydrophobically-modified methyl cellulose (HMMC). The approach involved HMMC as the main component of aqueous compositions to provide a system with fast gelling properties, which has not been accomplished with aqueous solutions of unmodified methyl cellulose (MC). MC was modified with the stearyl group as a hydrophobic modifier by controlling the degree of modification. The gelation rate of aqueous solutions containing identical amounts of HMMC and NaCl increased as the temperature increased. The HMMC solutions gelled at a fixed temperature and concentration range, while the unmodified MC solutions did not show sol-to-gel transition. In addition, HMMC solutions exhibited much faster gelation than MC solutions at given polymer and NaCl concentrations. The HMMC/NaCl solutions exhibited the reversible gel-to-sol transition upon cooling below 25°C. The rate of sol-to-gel transition at body temperature, and the reversible gel-to-sol transition at room temperature, were modulated by adjusting the concentration of HMMC and NaCl, respectively. The HMMC/NaCl compositions provided a simple system for accurate control of the thermogelling temperature and the thermogelation rates.

KEY WORDS: thermogelation, hydrophobical modification, methycellulose, sol-gel phase transition, reversible gelation

INTRODUCTION

Thermally reversible gelation of aqueous solutions of macromolecules has generated a great deal of interest due to their

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applicability in pharmaceutical and biomedical fields [1]. Gel-forming properties are attractive in the area of drug delivery in that they can offer the possibility of gelation of an administered formulation containing bioactive agents at the body temperature [2,3]. Typical examples of polymers for thermoreversible gelation are triblock copolymers consisting of poly(ethylene oxide) and poly(propylene oxide) (PEO-PPO-PEO, Pluronics), their analogs and hydrophobically modified cellulose [4,5]. Hydrophobically-modified celluloses, such as methylcellulose (MC) and ethyl(hydroxyethyl)cellulose (EHEC), have been widely investigated [6–9].

MC is a water-soluble polymer that has been used as a binder or thickener in pharmaceutical, food and ceramic processing [5,10,11]. MC has a heterogeneous structure consisting of substituted regions with methoxy groups called hydrophobic zones and less substituted regions called hydrophilic zones [12]. Aqueous solutions of MC are known to undergo sol-to-gel transition when the temperature is increased. Since the first observation by Heymann on reversible sol-to-gel transitions [10], significant efforts have been made to clarify the nature of thermogelation of MC solutions [5,7–9,11,13–16]. The main driving force for gelation of MC is the temperature-triggered intermolecular association of hydrophobic groups containing methoxy substitution. At the lower temperatures, polymers are fully hydrated and only weak polymer–polymer interactions exist through simple entanglement. As the temperature increases, the hydration of polymers by water is gradually weakened and eventually a polymer–polymer association becomes more pronounced, thereby resulting in the formation of a gel structure. Gelation of MC solutions is completely reversible in that gels warming up to a certain temperature return to the original sol state upon cooling.

Inorganic salts are a third component in MC solutions that influence the gelation properties of MC since they cause conformational changes in polymer chains [17]. In particular, salting-out solutes such as NaCl, KF and $(NH_4)_2SO_4$, lower the gelation temperature because of their water-structure formation properties. Although the effect of salts on the thermogelation of MC solutions is known [6], the salt effect on HMMC has not been studied, especially with control of gelation rates at body temperature.

The goal in this study was to investigate the salt effect on the gelation properties of aqueous HMMC solutions at body temperature. The stearyl group, as a hydrophobic modifier, was attached to MC to produce HMMC. The thermosensitive gelation properties of aqueous compositions, consisting of HMMC as the main component and NaCl as a salting-out solute, were evaluated.

EXPERIMENTAL SECTION

Materials and Equipment

Methyl cellulose (MC) with number average molecular weights (M_n) of 14,000 (MC14000) and 40,000 (MC40000) were purchased from Aldrich Co. and used without further purification. The average degree of methoxy substitution in MCs was in the range of 1.6–1.9. Stearoyl chloride, stearic acid, 1,3-dicyclohexylcarbodiimide (DCC), 4-dimethylaminopyridine (DMAP) and triethylamine (TEA) were also purchased from Aldrich Co. and used as received. Dimethylsulfoxide (DMSO), *N,N*-dimethylacetamide (DMAc) and ethanol were reagent grade. ^{1}H and ^{13}C NMR spectra were obtained on a Bruker ARX300 spectrometer at 300 MHz and 75 MHz, respectively.

Synthesis of Hydrophobically-modified MC (HMMC)

MC was hydrophobically-modified with stearyl groups. Two different procedures were used in the synthesis of HMMC. One reaction used stearyl chloride with MC in the presence of TEA. The other used a DCC-mediated coupling reaction between MC and stearic acid. For the first method, a solution of MC (0.3 g) and TEA (0.016 g, 1.6×10^{-4} mol) in dry DMAc (10 mL) was stirred at 70°C for 3 h until the solutes were completely dissolved. Stearoyl chloride (0.024 g, 8.0×10^{-5} mol) was then added to a stirred solution of MC and TEA and the reaction mixture was stirred at room temperature under nitrogen for 24 h. MC modified with the stearyl group was precipitated in ethanol (100 mL). The final product was washed with ethanol twice, filtered and dried *in vacuo* at 50°C for 24 h. For the second method, a solution of MC (1 g) in dry DMSO (30 mL) was added to stearic acid (0.076 g, 2.7×10^{-4} mol). This solution was stirred at 70°C for ~1 h until the solutes were completely dissolved. DCC (0.111 g, 5.4×10^{-4} mol) and DMAP (0.013 g, 1.6×10^{-4} mol) were then added with stirring. The reaction mixture was maintained at room temperature under nitrogen for 24 h. Purification of HMMC14000-2 was carried out using the same method used for HMMC14000-1. HMMC40000 was also synthesized by the second synthetic procedure.

Thermogelling Properties

Reversible thermogelling behavior was examined as follows: an aqueous polymer solution (0.5 g) was prepared by dissolving the HMMC in a 3 mL vial with an inner diameter of 11 mm to obtain a homogeneous solution. The diameter and the thickness of the samples for the thermosensitive transition experiment were 11 and 5.5 mm, respectively. Aqueous solutions with different concentrations ranging from 2.5 wt% to 4.5 wt% of HMMC14000 were prepared. To these HMMC14000 solutions, NaCl was added in concentrations from 0.65 M to 0.9 M. A polymer concentration of 2 wt% and 1.0 M NaCl were employed for the HMMC40000 sample. After immersing the vials in a water bath at a desired temperature, the rates for the sol-to-gel and the reversible gel-to-sol transitions were measured. Temperatures of 33°C and 37°C were used for the evaluation of the sol-to-gel transition, and temperatures of 4°C, 20°C and 25°C were employed to determine the gel-to-sol transition. The gel-to-sol transition was examined by the tube-rotating method from vertical to horizontal position in a water bath. When the sample was immobile for 1 min, it was regarded as a gel. The transition was observed with an accuracy of ± 1°C.

RESULTS AND DISCUSSION

Synthesis of HMMC

HMMC was synthesized by two different methods as illustrated in Scheme 1. One method involved the reaction of stearoyl chloride with

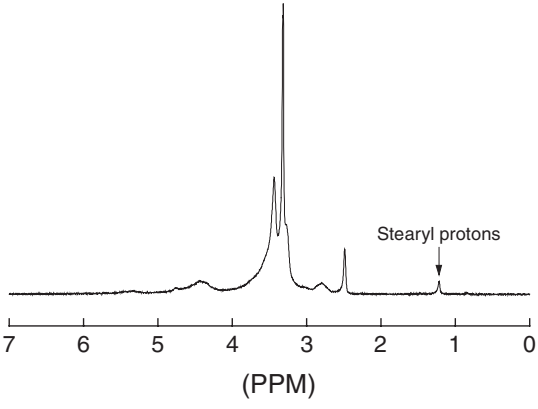
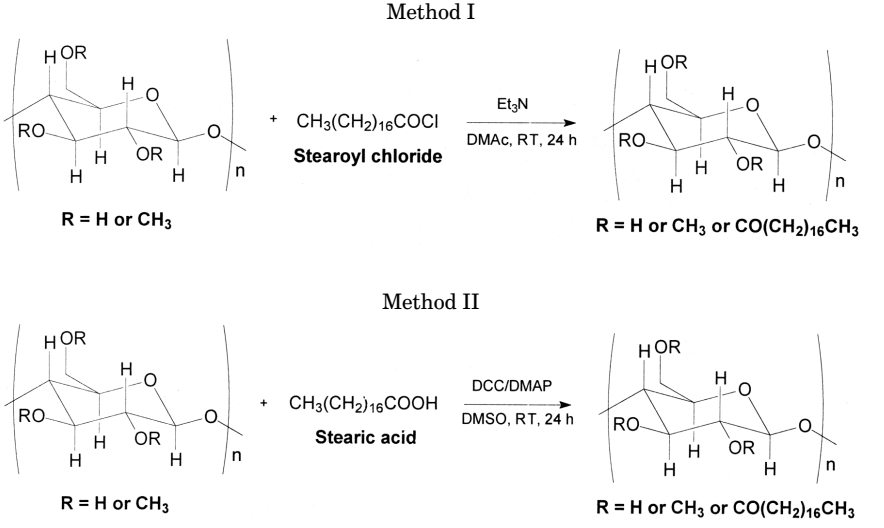


Figure 1. ^{1}H NMR spectrum of HMMC14000-1.

MC in the presence of TEA and the other method involved DCC-mediated coupling reaction between MC and stearic acid. For HMMC14000-1, the stearoyl groups were attached to MC14000 using stearoyl chloride. HMMC14000-2 and HMMC40000 were prepared by the DCC-mediated reaction. The characteristics of HMMC, such as a modification degree and the average number of stearoyl groups per chain, were estimated by analyzing the ^{1}H NMR spectra. In Table 1 is a summary of the characterization of HMMC14000-1, HMMC14000-2 and HMMC40000. HMMC14000-1 contained 2.32 wt% stearate groups, which corresponds to 1.2 modifiers/chain. On the other hand, HMMC14000-2 and HMMC40000 had 0.43 wt% stearate groups, corresponding to 0.2 and 0.6 modifiers/chain, respectively. The HMMC14000-1 grafted stearate corresponded to a modification degree



Scheme 1. Synthetic routes to HMMC.

Table 1. Characteristics of HMMC.

Polymer	Modification degree[a] (mol%)	Stearate modifier[a] (wt%)	Average number of modifiers/chain[b]
HMMC14000-1	1.6	2.32	1.2
HMMC14000-2	0.3	0.43	0.2
HMMC40000	0.3	0.43	0.6

[a]Estimated by ^{1}H NMR analysis.
[b]Average number of modifiers per chain = (wt%/100) × (MW of polymer)/(MW of modifier).

of 1.6 mol%, based on anhydroglucose units in the cellulose backbone. HMMC14000-2 and HMMC40000 have a stearate modification degree of 0.3 mol%. Modification of MC with stearate groups was more effective with stearoyl chloride than with stearic acid. The compositions of the thermogelling HMMC solutions are listed in Table 2. As the modification degree or the molecular weight of MC decreased, the concentrations of HMMC solutions had to be increased.

Thermogelling Properties

The thermogelling properties of aqueous solutions of the HMMC were evaluated and the results are shown in Table 3. The rate of thermogelation and the gelation temperature of HMMC were controlled by adding NaCl. As shown in Table 3, the HMMC14000-1a solution (2.5 wt%), containing 0.8 M NaCl, underwent sol-to-gel transition at 33°C in 15 min. At 37°C, a solution with an identical composition

Table 2. HMMC and NaCl composition of aqueous solutions.

Composition	Polymer concentration (wt%)	NaCl (M)
HMMC14000-1a	2.5	0.8
HMMC14000-1b	2.5	0.9
HMMC14000-1c	2.7	0.8
HMMC14000-2a	3.5	0.7
HMMC14000-2b	4.0	0.65
HMMC14000-2c	4.5	0.65
HMMC40000-a	2.0	1.0

Table 3. Rate control of reversible thermogelation of various aqueous compositions.

	sol-to-gel[a] (min)		gel-to-sol[b] (min)		
Composition	33°C	37°C	4°C	20°C	25°C
HMMC14000-1a	15.0	2.5	< 0.5	< 1.0	10
HMMC14000-1b	5.0	1.5	< 0.5	< 1.0	12
HMMC14000-1c	6.0	2.0	< 1.0	< 1.5	15
HMMC14000-2a	7.0	1.5	< 0.1	< 0.5	8
HMMC14000-2b	4.5	0.5	< 0.1	< 0.5	15
HMMC14000-2c	3.5	0.5	< 0.1	< 0.5	*
HMMC40000-a	5.0	1.0	< 0.1	< 1.5	*

[a]Time for the sol-to-gel transition.
[b]Time for the gel-to-sol transition, which was examined after equilibration at 33°C for 15 min.
*Not observed in 30 min.

became an immobile gel within 2.5 min. It was interesting to note that HMMC solutions formed a gel structure even at a fixed temperature and concentration, whereas thermogelation was not observed with any of the unmodified MC solutions. For example, HMMC14000-1a showed a sol-to-gel transition at 33°C but the unmodified MC14000 solution with the identical composition (e.g., 2.5 wt% polymer and 0.8 M NaCl content) did not form a gel even at 37°C. This indicated that the hydrophobic modification of MC with the stearate groups triggers thermogelation at lower temperatures due to the increased hydrophobic interaction between polymer chains in solutions. It was also noted that HMMC solutions gelled much faster than MC solutions at given polymer and NaCl concentrations. HMMC14000-1a formed a gel in less than 1 min at 40°C while MC14000 became a gel in 3 min under identical conditions. It appears that the increased hydrophobicity of HMMC, due to the stearate groups, facilitated the formation of gels in shorter time periods. The gelation rate was controlled by varying the HMMC concentration as well as the NaCl concentration at a given temperature. As the concentration of HMMC or NaCl increased, the sol-to-gel transition rate became faster. Most of the HMMC solutions exhibited reversible gel-to-sol transitions on cooling below 25°C. The HMMC14000-1a gel (2.5 wt%) containing 0.8 M NaCl became a sol within 30 s and 1 min at 4°C and 20°C, respectively. As the cooling temperature decreased, the transition rate from gel to sol became faster.

CONCLUSIONS

Aqueous compositions with inverse thermogelling properties were developed based on hydrophobically-modified methyl cellulose (HMMC) and NaCl. The temperature and the sol-to-gel rate, as well as the reversible sol-to-gel transition, were readily controlled by adjusting the polymer and NaCl concentrations. HMMC solutions underwent thermogelation more readily than unmodified MC solutions and developed a gel structure under conditions where gelation was not observed with unmodified MC solutions. The HMMC/NaCl compositions are simple systems that provide fast gelling properties as well as control of the thermogelation temperature and the thermogelation kinetics.

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