Journal of Applied Polymer Science

Synthesis of a Biocompatible Poly(2-hydroxyethyl methacrylate)–Chitosan Core–Shell Hydrogel Latex

Somkieath Jenjob,[1,2] Montri Ratanajanchai,[1] Natshisa Mahattanadul,[1,2] Sunhapas Soodvilai,[3] Panya Sunintaboon[1,2]

[1]Department of Chemistry, Faculty of Science, Mahidol University, 999 Phuttamonthon 4 Road, Salaya, Nakhon Pathom 73170, Thailand

[2]Center of Excellence for Innovation in Chemistry, Department of Chemistry, Faculty of Science, Mahidol University, Rama 6 Road, Bangkok 10400, Thailand

[3]Department of Physiology, Faculty of Science, Mahidol University, Rama 6 Road, Bangkok 10400, Thailand

Correspondence to: P. Sunintaboon (E-mail: panya.sun@mahidol.ac.th)

ABSTRACT: Core–shell hydrogel latexes, composed of a poly(2-hydroxyethyl methacrylate) (PHEMA) core chemically coated with chitosan (CS) shell, were synthesized via an emulsifier-free emulsion polymerization, free radically initiated by a redox couple of *tert*-butyl hydroperoxide and amine groups on CS itself. The variation of some polymerization parameters [e.g., polymerization time, CS/2-hydroxyethyl methacrylate (HEMA) weight ratio, and content of crosslinker] was systematically investigated in this study. We found that the weight ratios between CS and the HEMA monomer influenced the course of polymerization, which was traced by the change in percentage monomer conversions, and the colloidal stability of the PHEMA–CS hydrogel latexes obtained. Moreover, the polymerization time affected their particle sizes and surface charges. For the colloidally stable PHEMA–CS hydrogel latexes, their sizes and charges ranged from 600 to 689 nm and from 32 to 51 mV, respectively. *N,N′*-Methylene bisacrylamide was used as a crosslinking agent for the core component; this was found to be able to enhance the hydrogels' thermal stability and water uptake. Moreover, the 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide assay showed that 100% cell viability was achieved during the treatment of the PHEMA–CS latex (0.2–2.5 mg/mL) with Caco-2 cells. © 2013 Wiley Periodicals, Inc. J. Appl. Polym. Sci. **2014**, *131*, 40003.

KEYWORDS: biocompatibility; colloid; emulsion polymerization; latexe; microgel

Received 23 July 2013; accepted 24 September 2013

DOI: 10.1002/app.40003

INTRODUCTION

Hydrogel is a polymeric network with chemical or physical crosslinks that can be swollen in water. The amount of water absorbed in the polymeric hydrogel matrix is at least 20% and can reach a value of 99 %.[1] Hydrogels based on both natural and synthetic polymers are widely used in several biomedical applications, including soft contact lenses,[2] endovascular occlusion of blood vessels,[3] and drug-delivery systems.[4] In addition, they have continued to be of interest for cell encapsulation and tissue engineering technology.[5] Among synthetic polymers, poly(2-hydroxyethyl methacrylate) (PHEMA) is well known and has been used as a hydrogel because of its intrinsically greater water absorption ability, mechanical strength, and excellent biocompatibility.[6] Sahiner et al.[7] prepared cationic hydrogels of PHEMA, acrylamide, and (3-acrylamidopropyl) trimethyl ammonium chloride to complex with anionic DNA for gene-delivery vehicles. Danisman et al.[8] prepared a membrane from poly(2-hydroxyethyl methacrylate–glycidyl methacrylate) as a support for invertase immobilization.

Chitosan (CS), a natural copolymer derived from chitin, has been widely used in many applications (pharmaceutical and biomedical) because it is nontoxic, biodegradable, and biocompatible.[9] These properties render CS as a good candidate for the development of nutraceutical-delivery systems in the food industry.[10] Furthermore, because of its polycationic nature, CS has been investigated extensively for carriers and delivery systems of negatively charged agents, such as DNA, drugs, and cosmetic ingredients.[11,12] Moreover, the positive charge of CS can interact with the tight junction of epithelial cells, through which a capacity of drug transport can be improved.[13]

Usually, biocompatible, biodegradable, and water absorption properties are important characteristics for uses in biomedical

Additional Supporting Information may be found in the online version of this article.

applications, such as controlled release carriers or tissue engineering scaffolds. Hence, the combination of CS and PHEMA would be useful for providing these properties altogether. Many researchers have found that the graft copolymers of PHEMA and CS exhibited improved hydrophilicity and biocompatibility.[14] For example, Moghaddam et al.[15] prepared CS–PHEMA core–shell nanoparticles via radical polymerization with cerium(IV) ammonium nitrate as a redox initiator for the encapsulation of fluorescein isothiocyanate dextran. Kim et al.[16] prepared a semi-interpenetrating polymer network hydrogel of CS and PHEMA with azobisisobutyronitrile as an initiator and ethylene glycol dimethacrylate as a crosslinking agent. Casimiro et al.[17] fabricated CS–PHEMA membranes by γ-irradiation from a ^{60}Co source. In recent years, there has been increasing interest in research of both the syntheses and applications of hydrogels that are in the form of nanometer or submicrometer-sized particles. Such particles have a larger surface-to-volume ratio because of their small size; this leads to more exposure and a fast response to the surroundings. Their surface and bulk properties can be tailored so that multifunctional or intelligent characteristics can be achieved. Herein, we report the synthesis of a core–shell hydrogel latex having a CS shell and a PHEMA core through an emulsifier-free emulsion polymerization induced by a small amount of *tert*-butyl hydroperoxide (TBHP). The systematic variation of polymerization parameters, such as the amounts of CS and 2-hydroxyethyl methacrylate (HEMA) monomer and the polymerization time, on the course of polymerization was investigated. Then, their extensive physicochemical properties, such as their particle size and ζ-potential measurement, scanning electron microscopy, transmission electron microscopy (TEM), atomic force microscopy (AFM), and thermogravimetric analysis (TGA) were examined. The effect of *N,N′*-methylene bisacrylamide (MBA) crosslinking agent was also evaluated; it was found to influence the PHEMA–CS's thermal stability and water uptake. Moreover, the cytotoxicity of such hydrogels was evaluated by a 3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide (MTT) assay.

EXPERIMENTAL

Materials

CS was purchased from Seafresh Chitosan Laboratory (Thailand). The degree of deacetylation was about 90%, and its average molecular weight was about 150 kDa. HEMA and MBA were purchased from Aldrich. HEMA was purified with a column packed with alumina adsorbents. TBHP was obtained from Fluka and used without further treatment. MTT, Dulbecco's phosphate-buffered saline (PBS), Dulbecco's Modified Eagle's Medium (DMEM), fetal bovine serum, and trypsin were purchased from Invitrogen (Carlsbad, CA). Deionized water was used throughout this study. All other chemicals were commercially available and were analytical grade.

Preparation of the PHEMA–CS Hydrogel Latex

The PHEMA–CS hydrogel latex was synthesized via an emulsifier-free emulsion polymerization, free radically triggered by a redox-initiating system consisting of TBHP and amine groups from CS. A batch process of emulsifier-free emulsion polymerization was carried out in a 100-mL, water-jacketed glass reactor equipped with a nitrogen inlet, reflux condenser, water cooling system, and magnetic stirrer.[18] Deionized water (23 g), a CS solution (predissolved in a 1 wt % acetic acid aqueous solution, 25 g), and the MBA crosslinker with a predetermined amount (0.01–0.1 g) were first charged into the reactor. After the reactor was purged with nitrogen gas for 30 min, HEMA monomer was added to the reactor. The polymerization reaction was then started after the addition of a TBHP aqueous solution (1 g, 5×10^{-3} *M*) and was continued at 80°C for up to 2 h under continuous stirring. The conversion percentages and solid contents were determined by a gravimetric method on basis of the weight of HEMA used and that of polymerized HEMA [with eqs. (1) and (2), as shown in the Supporting Information]. The number of particles per unit volume of latex (N_p) was calculated with the method described in ref. 19.

To study the effect of the HEMA-to-CS ratio, the weight ratio of the HEMA monomer to the CS solution (1 wt % in acetic acid), was varied at 1:10, 1:15, 1:20, and 1:25 at a fixed amount of MBA (0.01 g). Each polymerization reaction underwent at 80°C for 2 h. Upon the variation of the polymerization time (0–120 min), some data of the resulting products were acquired, including the percentage conversion, hydrodynamic diameter, ζ potential, N_p, and core size of the particle. The effect of the MBA crosslinker was also investigated by the variation of its content at 0.01, 0.03, 0.05, and 0.1 g.

Particle Size and ζ-Potential Measurements

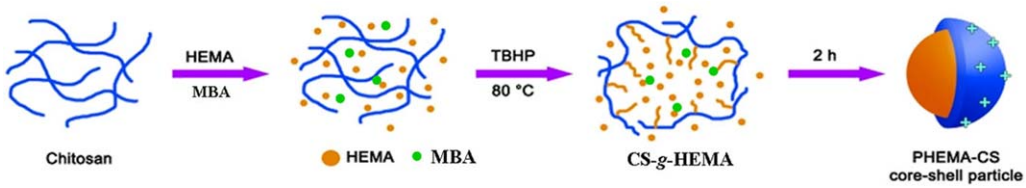
Before characterizations, the resulting latex of the PHEMA–CS hydrogels was cleaned to remove the free CS and unreacted HEMA monomer by a few centrifugation–redispersion cycles at a centrifugation speed of 30,000 rpm for 30 min in each cycle. The particle size and ζ-potential of the hydrogel latex were measured by electrophoretic light scattering with a Zetasizer 3000 (Malvern Instruments, United Kingdom) in a 1 m*M* NaCl solution at 25°C.

Morphological Analysis

The size and morphology of the PHEMA–CS hydrogel latex was obtained by TEM. To prepare a sample for TEM, a drop of the latex sample diluted with deionized water was dried on a Formva-coated copper grid, which was then stained with a 1.5 wt % phosphotungstic acid aqueous solution before it was imaged through a transmission electron microscope (JEM-1230, JEOL, accelerating voltage = 100 kV). An AFM instrument (model Ns3a, Digital Instruments, Inc.) equipped with a Nanoscope IIIa controller via tapping mode was also used to gain additional information about the particle's size and morphology. To prepare a sample for AFM, a diluted latex dispersion was dropped onto the 10×10 mm^2 cover glass and dried under ambient temperature for 2 days before it was visualized.

Chemical Functional Group Analysis

After it was air-dried for 2 days, the PHEMA–CS25 powder was extracted by a Soxhlet apparatus with ethanol as a solvent for 48 h to remove uncrosslinked homo-PHEMA. The remaining solids were then dried in an oven at 70°C for 24 h and then characterized by Fourier transform infrared (FTIR) spectroscopy (PerkinElmer, PE 2000) with KBr disks. The scanning for each spectrum was attained in a range of 4000–370 cm^{-1} at a



Scheme 1. Formation of the PHEMA–CS core–shell hydrogel latex by emulsifier-free emulsion polymerization. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

resolution of 4 cm^{-1} and with 32 scans. KBr powder was used as a reference background.

TGA

TGA (TGA/SDTA851, Mettler, Switzerland) was used to study the effect of the crosslinker on the hydrogel thermal stability. The sample was accurately weighed (8–12 mg) in an aluminum pan. The measurement was done in a temperature range of 40–600°C at a heating rate of 10°C/min and under a nitrogen and oxygen purge at a flow rate of 60 cm^3/min.

Water Uptake Measurement

The water uptake was one of the intrinsic properties of the prepared hydrogel. Before measurement, a freeze-dried PHEMA–CS hydrogel powder (ca. 0.05 g) was pressed with a 2-ton mold to form a disc with 6.5 mm in radius and 1.0 mm in thickness. The water uptake was then determined gravimetrically. To do so, each of the discs was submerged in a 100-mL beaker containing deionized water. At each predetermined time (0, 5, 10, 20, 30, 60, and 120 min), the disc was removed from the medium, blotted to remove excess water, and immediately weighed. The water uptake was then calculated according to the following equation:

$$\text{Water uptake}\,(\%) = [(W_t - W_0)/W_0] \times 100 \qquad (1)$$

where W_0 and W_t are the weights of the dry and swollen discs, respectively, measured at different time periods. The value reported was the average of three measurements.

Cytotoxicity Measurement

The cytotoxicity of the PHEMA–CS core–shell hydrogel latex was evaluated by MTT assay. The MTT assay is a method of determining the viable cell number in cytotoxicity studies. This assay is based on the cleavage of the yellow tetrazolium salt (MTT) to form a soluble purple formazan product by mitochondrial enzymes, and the amount of formazan product is directly proportional to the number of living cell. Caco-2 cells, human intestinal epithelial cells, were used as a model to study the cytotoxicity. Caco-2 cells (passage 39) were seeded at 20,000 cells/well in a 96-well plate and maintained at 37°C in an atmosphere of 5% CO_2. The DMEM was removed; then, the cells were washed in PBS and treated with 100 μL of sample (0.2–2.5 mg/mL) dissolved in DMEM for 24 h. Afterward, the solution was removed, and the cells were washed with PBS followed by the addition of MTT solution (5 mg/mL in DMEM) for a further 3-h incubation. The solution was then removed, and 150 μL of dimethyl sulfoxide was added. The optical density at 540 nm was measured. MTT is a tetrazolium salt that is oxidized by mitochondrial dehydrogenase in living cells to give a dark blue formazan product. Damaged or dead cells show reduced or no dehydrogenase activity. The relative cell viability (%) was calculated as follows:

$$\text{Cell viability}\ (\%) = \left(\text{Absorbance}_{\text{sample}}/\text{Absorbance}_{\text{control}}\right) \times 100 \qquad (2)$$

RESULTS AND DISCUSSION

Formation of the PHEMA–CS Hydrogels

The emulsifier-free emulsion polymerization was accomplished with the aid of an amine/TBHP initiating system. Because CS has a plenty of amine groups on its backbone, it could act as a co-initiator for this polymerization system. Moreover, CS could also behave as a colloidal stabilizer for the formed particles because it had a tendency to be located in the outer layer because of its hydrophilicity. The mechanism for this polymerization system was proposed as follows: the amphiphilic graft copolymers consisting of a hydrophilic CS backbone and relatively less water-soluble PHEMA side chains were first formed.[18] Then, they spontaneously self-assembled to core–shell particles in an aqueous medium with CS as the shell and PHEMA as the core, as shown in Scheme 1. In this study, a systematic investigation of the effects of some of the polymerization parameters was conducted.

Effect of the Polymerization Time

In this part, TEM images of the latex samples collected from each polymerization time interval were acquired; these were a source of information on the particle sizes, numbers, and morphology, as shown in Figure 1. At the early stage of polymerization (<30 min polymerization time), the core–shell morphology of the latex sample could not be clearly observed [Figure 1(a–c)]. However, after 30 min, we observed the formation of PHEMA–CS hydrogels with a well-defined core–shell morphology, in which the PHEMA core was coated with the CS shell, indicated by the dark periphery [Figure 1(d–i)]. From the TEM micrographs, the core diameter of the particles and N_p were calculated [eq. (3)]; they are shown in Figure 2. The PHEMA core tended to increase with the polymerization time. In the first stage of polymerization (5–10 min), many particles were already formed, as confirmed by a gradual increase in N_p from 10.9×10^{10} to 23.3×10^{10} particles/mL. Then, N_p increased from 27.7×10^{10} to 35.1×10^{10} particles/mL within 60–70 min and remained almost constant (ranging from 35.1×10^{10} to 34.3×10^{10} particles/mL) after 80 min of polymerization time (Figure 2). In this polymerization system, the free-radical initiation and subsequent polymerization seemed very effective because an induction period was not observed and the change in the monomer conversion was quite rapid in the first 40 min of polymerization. Similarly to the mechanism proposed for emulsifier-free emulsion polymerization, the particle nucleation

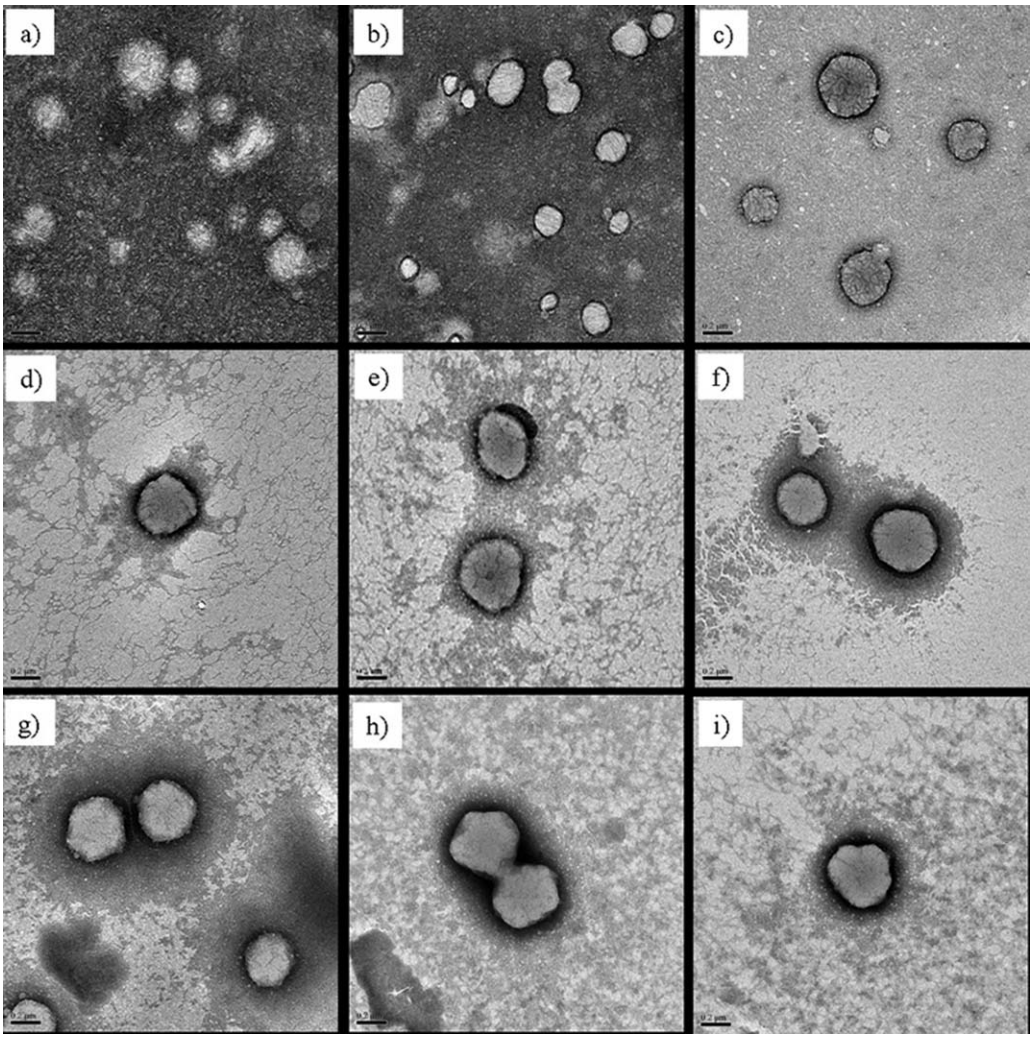


Figure 1. TEM images of the PHEMA–CS25 (HEMA 1 g, CS 25 g) hydrogel latexes at different polymerization times: (a) 5, (b) 10, (c) 20, (d) 30, (e) 40, (f) 60, (g) 80, (h) 100, and (i) 120 min (scale bar = 0.2 μm).

in this system occurred as follows. Once the HEMA monomers were grafted from the CS chains to a critical length, such grafted copolymers became surface-active. Then, these surface-active species aggregated to micellelike microdomains, having the grafted PHEMA and/or possible homo-PHEMA located inside the micelles, with the CS chains pointing toward the aqueous medium and bringing colloidal stabilization to the micelles and the polymer particles. Normally, PHEMA is quite water-soluble, but with the aid of the MBA crosslinker, the surface-active properties were enhanced. With regard to the particle number (as shown in Figure 2), it increased rapidly in the first 40 min; this indicated that the nucleation process was occurring during this period of time. Then, it slowly increased and became constant through the end of polymerization (120 min). Such a short nucleation process without an induction period confirmed the effective free-radical formation and polymerization with TBHP/amine groups (from CS).

During the course of polymerization, hydrodynamic diameters and ζ-potentials were also observed. The results revealed that the diameters of the particles increased from 374 to 535 nm with increasing polymerization time, whereas the ζ-potential values did not significantly change with the polymerization time (ca. +60 mV), as displayed in Figure 3. However, the ζ-potential value of PHEMA–CS25 in Table I was lower than the value appearing in Figure 3 because the result in Table I was

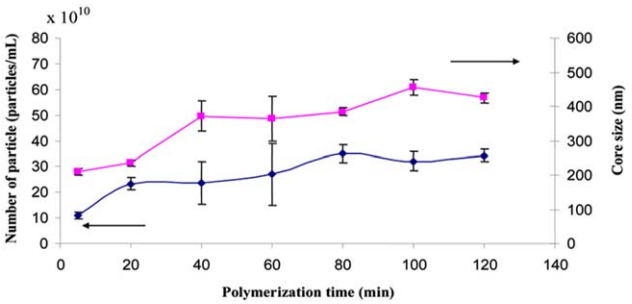


Figure 2. N_p values and core sizes of the PHEMA–CS25 hydrogel latexes at different polymerization times. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

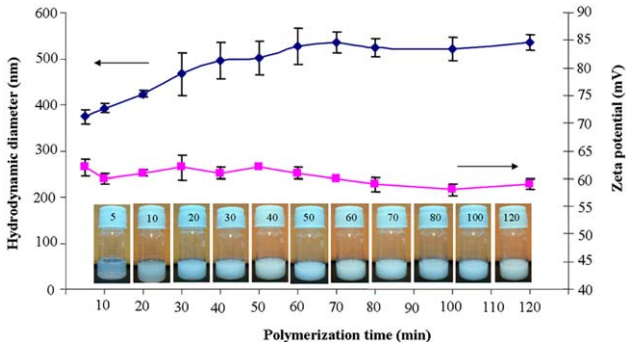


Figure 3. Hydrodynamic diameters and ζ potentials of the PHEMA–CS25 hydrogel latexes at different polymerization times. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

obtained from the latex after cleaning by centrifugation, whereas the result in Figure 1 was measured without centrifugation.

Effect of the HEMA/CS Ratios

Table I summarizes the monomer conversions, solid contents, particle sizes, and ζ-potentials after 2 h of polymerization. We observed that the monomer conversions increased from 62 to 82% with increasing amount of CS; this also increased the solid content of the latexes, which ranged from 1.4 to 2.1%. Because CS could provide amine groups that can generate propagating radicals in the presence of TBHP, the increase in CS led to an increase in propagating radicals, which accelerated polymerization. As a result, higher monomer conversions were increased. The ζ-potential values (32–51 mV) seemed to increase with the amount of CS added. The positive surface charges also implied that CS was located at the particle's surface, according to the mechanism mentioned previously. The average diameters of the stable PHEMA–CS systems were in the ranges 600–689 and 403–627 nm from electrophoretic light scattering (ELS) and TEM, respectively. In addition, from TEM, we observed that the sizes of the PHEMA–CS latexes appeared to be polydisperse.

The effect of the HEMA monomer feed on the conversion was also investigated as the amount of 1 wt % CS solution was kept constant at 25 g (Table S1, Supporting Information). The weight of the HEMA monomer was varied from 1 to 4 g. We was found that after the polymerization reaction was complete, coagula were observed in the latex products when 2, 3, and 4 g of HEMA monomer were fed (Figure S1, Supporting Information). The increase in the HEMA monomer may have resulted in a higher percentage conversion of HEMA to PHEMA and led to a larger PHEMA core component. Because of this, the CS, which was located on the particle shell layer, the electrostatic colloidal, might not have been sufficient to provide stabilization to the particles, and this resulted in the formation of coagula. Thus, the optimum amounts of HEMA monomer and CS solution for the colloidally stable PHEMA–CS hydrogel latex were 1 and 25 g, respectively.

The core–shell nanostructure of the PHEMA–CS hydrogel latexes with different amounts of CS added was clearly observed, as shown in Figure 4. We observed that the PHEMA core was coated with a CS shell. However, although the amount of CS increased, their sizes were not significantly different and were around 400–600 nm.

The chemical functional groups of PHEMA–CS25 were qualitatively analyzed through FTIR spectroscopy. Figure 5 represents the FTIR spectra of the PHEMA-CS solid obtained after Soxhlet extraction and that of the original CS. In the spectrum of CS [Figure 5(a)], the broad peak appearing in the range 3178–3654 cm^{-1} corresponded to O—H and N—H stretching. The peaks at 1654 and 1595 cm^{-1} were assigned to amide I and amide II (due to the remaining acetamide groups in the CS backbone), respectively. The peak at 1151 cm^{-1} was assigned to C—O, and those at 2886 and 1460 cm^{-1} were assigned to the C—H band. For PHEMA–CS25 [Figure 5(b)], all bands mentioned previously, including the band at 1725 cm^{-1} for C═O stretching, occurred. This indicated that PHEMA was grafted from the CS backbone.

Effect of the MBA Crosslinker

In this part, the effect of MBA on the formation and properties of the PHEMA–CS hydrogel latexes was illustrated. We found that upon the variation of MBA (0.01–0.1 g), the hydrodynamic size and ζ-potential of the resulting PHEMA–CS latexes were not much different. However, the comparison between the uncrosslinked and crosslinked PHEMA–CS25 latexes was also clarified with the AFM technique. As shown in Figure 6, the uncrosslinked PHEMA–CS latex appeared to be fused and aggregated together, whereas the crosslinked particles exhibited a more uniform spherical shape, which was less aggregated. We observed that the inner core provided the phase with a color similar to that of the background (cover slide) because it was crosslinked by the MBA crosslinking agent; this made it relatively harder than the shell of the particles.

TGA was used to study the thermal degradation of the crosslinked hydrogels compared to that of the uncrosslinked

Table I. Monomer Conversions, Solid Contents, Particle Sizes, and ζ-Potentials of the PHEMA–CS Core–Shell Hydrogel Latexes with Different Amounts of Added CS

Sample	HEMA/CS (w/w)[a]	Conversion (%)	Solid content (%)	Size by ELS (nm)	Size by TEM (nm)[b]	ζ (mV)
PHEMA-CS10	1:10	62 ± 1	1.4 ± 0.1	630 ± 8	403 ± 102	32 ± 2
PHEMA-CS15	1:15	68 ± 2	1.5 ± 0.1	600 ± 16	627 ± 248	42 ± 2
PHEMA-CS20	1:20	70 ± 2	1.7 ± 0.2	648 ± 9	432 ± 122	46 ± 1
PHEMA-CS25	1:25	82 ± 2	2.1 ± 0.1	689 ± 1	463 ± 138	51 ± 1

[a] 1 wt % CS.
[b] The *Np* values measured to determine the TEM diameters were approximately 20–70.

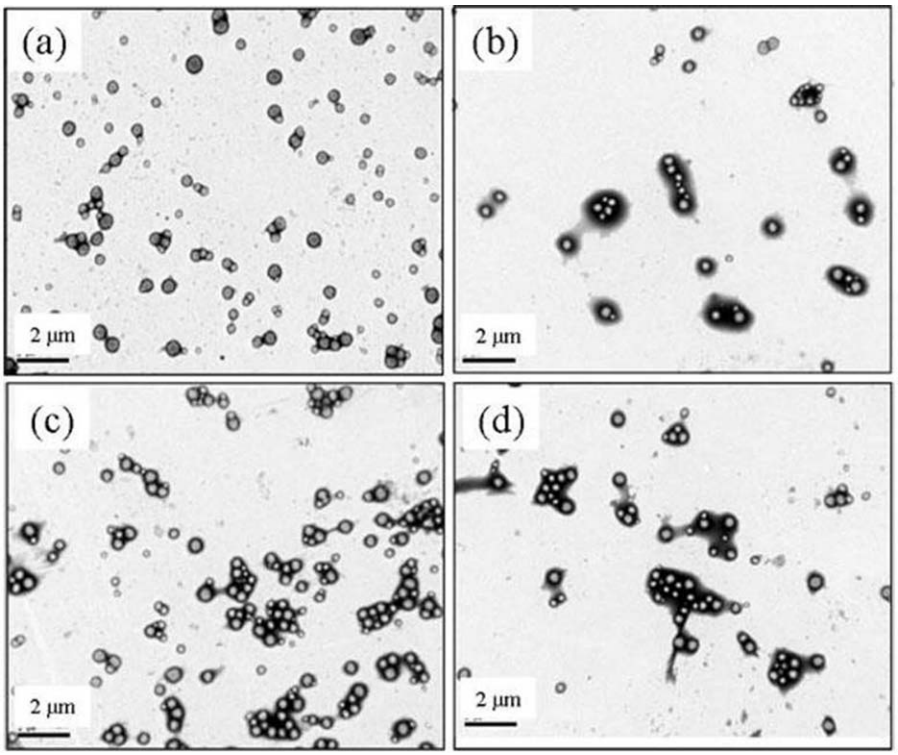


Figure 4. TEM images of the (a) PHEMA–CS10, (b) PHEMA–CS15, (c) PHEMA–CS20, and (d) PHEMA–CS25 hydrogel latexes.

PHEMA–CS25 hydrogels and pure CS. In the case of CS, a two-stage weight loss was observed and is illustrated in Figure 7. The first stage ranged between 40 and 170°C and had about a 12.1% weight loss. This may have corresponded to the loss of bound water. The second stage started at 250°C and continued up to 600°C with a 51.8% weight loss; this was probably due to the degradation of the CS chains. However, the TGA results showed that the destruction of the uncrosslinked PHEMA–CS25 hydrogels started at 158°C and ended at 450°C. The total weight loss of 99.7% was recorded. In contrast, the destruction temperature of the crosslinked PHEMA–CS25 hydrogels was shifted to 315°C, and degradation ended at 450°C. The total weight loss of PHEMA–CS25 was 97.4%. The TGA of the crosslinked PHEMA–CS25 hydrogels also showed a two-stage weight loss (excluding a loss of moisture at 100°C) in the temperature range 130–600°C. The first step, at 130–388°C, corresponded to a random chain scission of weak bonds between PHEMA and CS. The second stage, in the temperature range of 388–450°C, was associated with a chain scission within the PHEMA chains,[20] as shown in Figure 7. Therefore, it was clear that the crosslinking agent added to the system enhanced the particle's thermal stability.

The crosslinker affected not only the thermal stability of the resulting particle hydrogels but also their water uptakes. The water uptake measurement of the PHEMA–CS25 hydrogels with various MBA contents and at various immersion times was examined, and the results are shown in Figure 8. We observed that the water uptakes of the hydrogel without MBA abruptly increased from 0 to 76% within 30 min, then slowly increased from 76 to 89% within 60 min, and remained constant until 120 min of immersion time passed. In contrast, when the MBA crosslinker was charged into the system, the water uptakes exhibited abrupt change from 0 to around 100% in just 20 min of immersion time before remaining constant. However, the varied amount of MBA (from 0.01 to 0.1 g) did not result in a significant difference in the water uptake. The highest water uptake of up to 107% was obtained; this confirmed their water absorption ability, an important characteristic for uses in biomedical applications, such as controlled release carriers or tissue engineering scaffolds.

Cytotoxicity Measurement (MTT Assay)

Caco-2 cells, human intestinal epithelial cells, were used as a model for studying the cytotoxicity. The dispersions of such

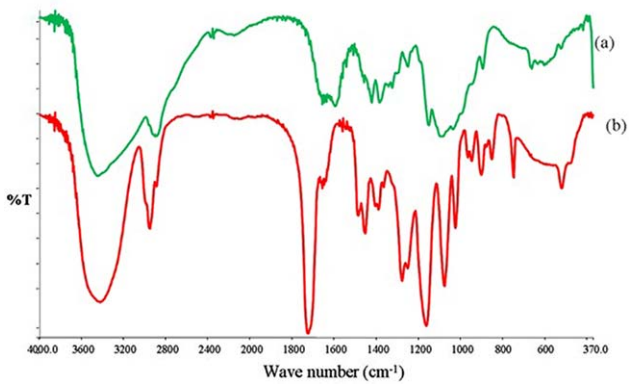


Figure 5. FTIR spectra of the (a) pure CS and (b) PHEMA–CS25 (HEMA 1 g, CS 25 g) hydrogel latex. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

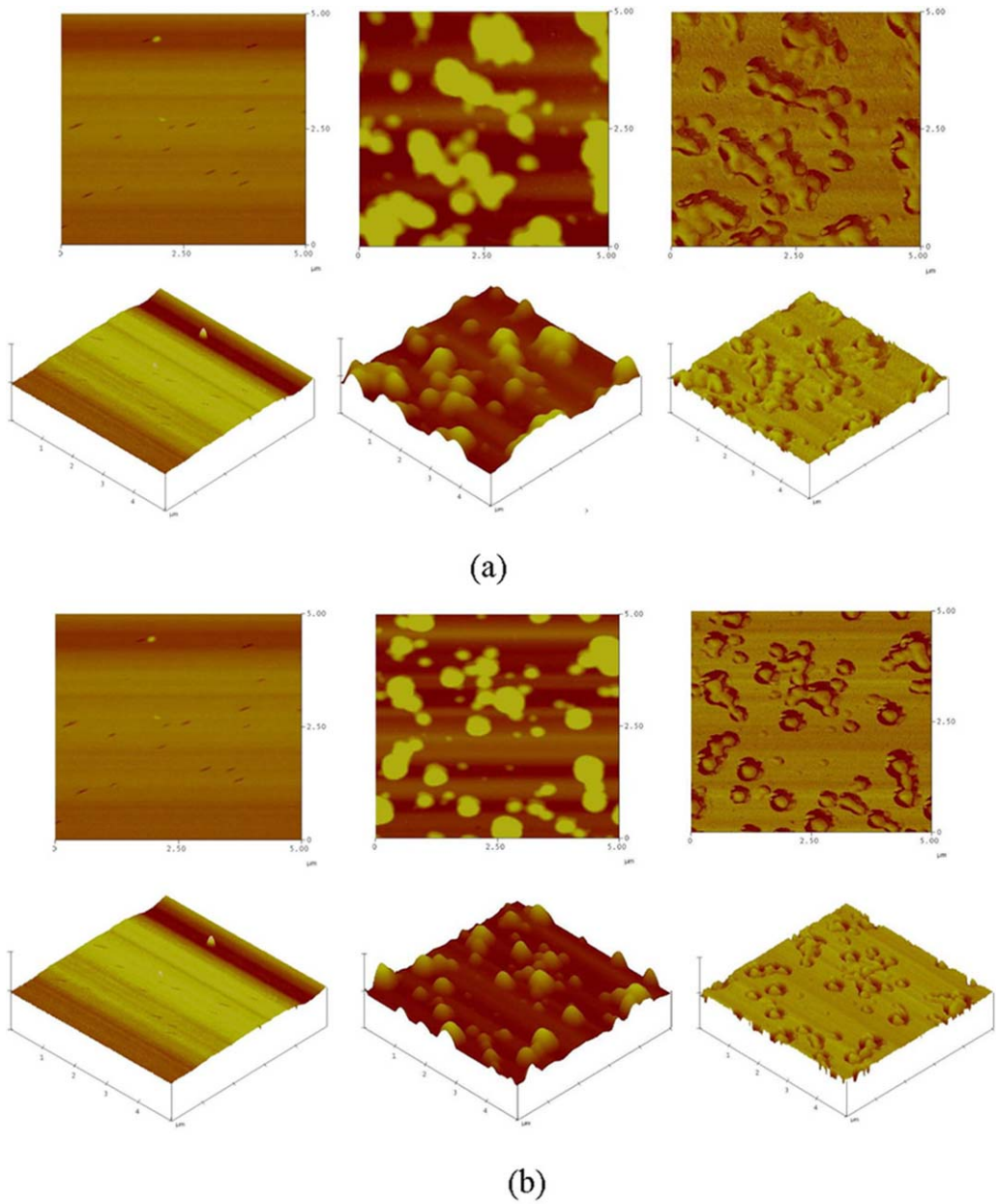


Figure 6. AFM micrographs of the PHEMA–CS25 hydrogel latexes: (a) MBA-free and (b) with 0.01 MBA (scale bar = 5 μm). The left panels show the glass blank, the middle panels show the height, and the right panels show the phase. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

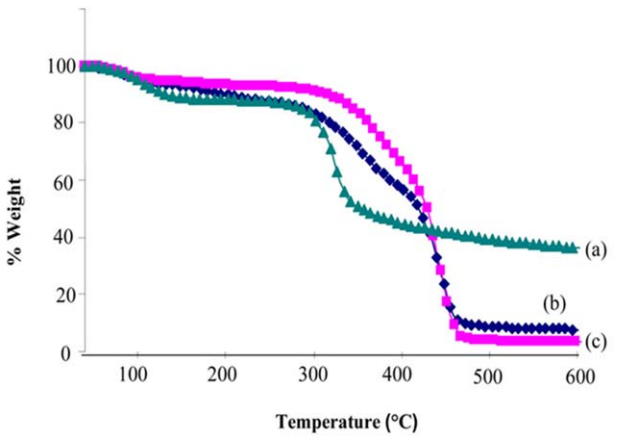


Figure 7. TGA thermograms of the (a) pure CS, (b) PHEMA–CS25 (MBA-free), and (c) PHEMA–CS25 hydrogel latexes. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

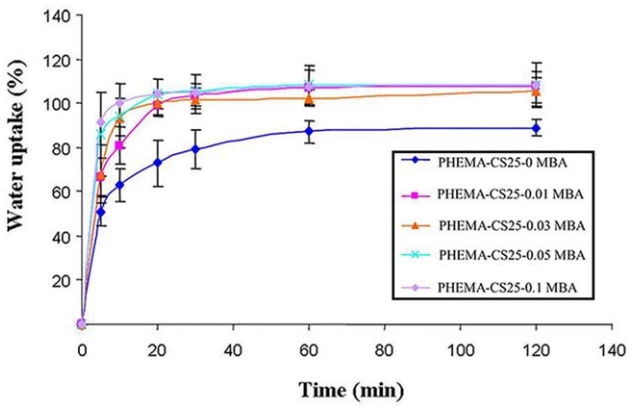


Figure 8. Water uptake (%) of 0, 0.01, 0.03, 0.05, and 0.1 MBA in the PHEMA–CS25 hydrogel latexes. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

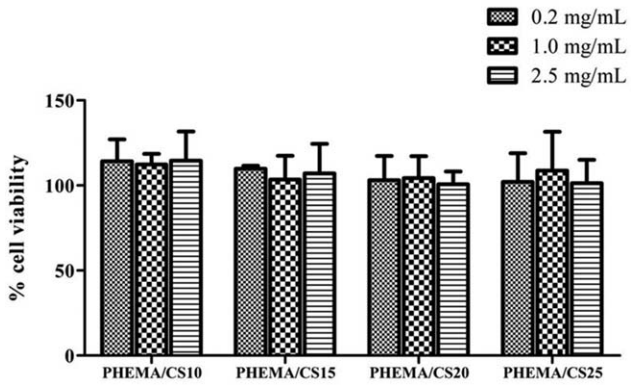


Figure 9. Cell viability (%) of Caco-2 cells in the presence of the PHEMA–CS hydrogel latexes prepared with different amounts of CS ($n = 3$).

hydrogels prepared from various amounts of CS in the concentration range of 0.2–2.5 mg/mL were subjected to MTT assay against the Caco-2 cells. The cell viability was then determined and is displayed in Figure 9. All of the PHEMA–CS samples in the whole concentration range (0.2–2.5 mg/mL) exhibited 100% cell viability. The results confirmed that the PHEMA–CS hydrogel latexes were biocompatible against the Caco-2 cells. Thus, theses hydrogel latexes consisting of biocompatible PHEMA and CS may be useful for various biomedical applications.

CONCLUSIONS

PHEMA–CS core–shell hydrogel latexes were prepared via an emulsifier-free emulsion polymerization. Their physicochemical properties were confirmed through particle size and ζ-potential measurement, TEM, AFM, and FTIR spectroscopy. For the synthesis, we found that polymerization time had a dramatic effect on the course of polymerization and the evolution of hydrogel latexes, whereas the HEMA/CS ratios also affected their formation and colloidal stabilization. The presence of the MBA crosslinking agent influenced the latexes' thermal stability and water uptake. The biocompatibility of the PHEMA–CS hydrogel latexes through MTT assay against Caco-2 cells was also illustrated.

ACKNOWLEDGMENTS

This research project is supported by Mahidol University. Also, financial support from the Center of Excellence for Innovation in Chemistry, Office of the Higher Education Commission, Ministry of Education is gratefully acknowledged.

REFERENCES

1. Deligkaris, K.; Tadele, T. S.; Olthuis, W.; Berg, A. V. D. *Sensors Actuators B* **2010**, *147*, 765.
2. Wichterle, O.; Lim, D. *Nature* **1960**, *185*, 117.
3. Horak, D.; Svec, F.; Kalal, J.; Gumargalieva, K.; Adamyan, A.; Skuba, N.; Titova, M.; Trostenyuk, N. *Biomaterials* **1986**, *7*, 188.
4. Roberts, C. C. R.; Buri, P. A.; Peppas, N. A. *J. Controlled Release* **1987**, *5*, 151.
5. Nguyen, K. T.; West, J. L. *Biomaterials* **2002**, *23*, 4307.
6. Weaver, J. V. M.; Bannister, I.; Robinson, K. L.; Bories-Azeau, X.; Armes, S. P. *Macromolecules* **2004**, *37*, 2395.
7. Sahiner, N.; Godbey, W. T.; McPherson, G. L.; John, V. T. *Colloid Polym. Sci.* **2006**, *284*, 1121.
8. Danisman, T.; Tan, S.; Kacar, Y.; Ergene, A. *Food Chem.* **2004**, *85*, 461.
9. Xu, Y.; Du, Y. *Int. J. Pharm.* **2003**, *250*, 215.
10. Chen, L.; Subirade, M. *Biomaterials* **2005**, *26*, 6041.
11. Pimpha, N.; Ruktanonchai, U.; Surassmo, S.; Opanasopit, P.; Rattanarungchai, C.; Sunintaboon, P. *Colloid Polym. Sci.* **2008**, *286*, 907.
12. Sang, Y. H.; Eun, L. J.; Chung, H.; Chan, K. I.; Young, J. S. *J. Controlled Release* **2005**, *103*, 235.
13. Mi, F.-L.; Wu, Y.-Y.; Lin, Y.-H.; Sonaje, K.; Ho, Y.-C.; Chen, C.-T.; Juang, J.-H.; Sung, H.-W. *Bioconjugate Chem.* **2008**, *19*, 1248.
14. El-Tahlawy, K.; Hudson, S. M. *J. Appl. Polym. Sci.* **2001**, *82*, 683.
15. Moghaddam, F. A.; Atyabi, F.; Dinarvand, R. *Nanomed.-Nanotechnol.* **2009**, *5*, 208.
16. Kim, S. J.; Shin, S. R.; Shin, D. I.; Kim, I. Y.; Kim, S. I. *J. Appl. Polym. Sci.* **2005**, *96*, 86.
17. Casimiro, M. H.; Gil, M. H.; Leal, J. P. *Nucl. Instrum. Methods Phys. Sect. Res. B* **2007**, *265*, 406.
18. Inphonlek, S.; Pimpha, N.; Sunintaboon, P. *Colloid Surf. B* **2010**, *77*, 219.
19. Ho, K. M.; Li, W. Y.; Lee, C. H.; Yam, C. H.; Gilbert, R. G.; Li, P. *Polymer* **2010**, *51*, 3512.
20. Huacai, G.; Wan, P.; Dengke, L. *Carbohydr. Polym.* **2006**, *66*, 372.