

## CHAPTER III

### RESEARCH METHODOLOGY

The aim of this work is to prepare semi-interpenetrating polymer networks (semi-IPNs) of chitosan/carboxymethylchitosan (CMC) hydrogels modified with polysiloxane (PDMS)/poly(ethylene glycol) (PEG). The effects of PDMS/PEG contents (1, 5, 10 and 20 wt%) and their  $\overline{M}_n$ 's (2,000 and 8,000 g/mol) in the hydrogels on the properties, e.g. water swelling behavior, percent of crosslinking, water contact angles, water vapor permeability, surface morphology, and tensile strength, were investigated. This chapter thus presents the experimental and characterization of such hydrogels.

#### Materials

##### 1. Solvents and chemicals

- 1.1 Chitosan from crabs (Taming Enterprise, Co.) with 98% deacetylation
- 1.2 Octamethylcyclotetrasiloxane (D<sub>4</sub>), 99% (Fluka)
- 1.3 1, 1, 3, 3-Tetramethyldisiloxane, 97% (Acros)
- 1.4 Allyl alcohol, 99% (Acros)
- 1.5 Karstd's catalyst (Platinum (0)-1, 3-divinyl-1, 1, 3, 3-tetramethyldisiloxane complex solution in poly(dimethylsiloxane), vinyl terminated) (Acros)
- 1.6 Trifluoromethanesulfonic acid (Triflic acid), 98% (Aldrich)
- 1.7 Poly(ethylene glycol) (PEG) with  $\overline{M}_n$  2,000 and 8,000 g/mol (Acros)
- 1.8 1, 6-Hexamethylene diisocyanate (HDI), 99% (Acros)
- 1.9 Sodium metabisulphite (Acros)
- 1.10 Chitosan powder from crabs, oligomer type 100 meshes (Taming Enterprise, Co.)
- 1.11 Monochloroacetic acid (CH<sub>3</sub>COOCl), 99% (Acros)

## Experimental

To prepare semi-IPN of chitosan/CMC hydrogels, the procedure involved three major steps as following:

1. Hydroxyl-terminated PDMS (PDMS diol) was synthesized *via* cationic ring-opening polymerization. Two different  $\overline{M}_n$ 's were targeted (2,000 and 8,000 g/mol). Two different  $\overline{M}_n$ 's of PEG were purchased and used as received (2,000 and 8,000 g/mol) from vendor. Abbreviated names of PDMS/PEG are illustrated in Table 3.1.

2. Chitsan/CMC hydrogels were modified with PDMS or PEG and hexamethylene-1, 6-di-(aminocarboxysulfonate) (HDA) was used a crosslinking agent.

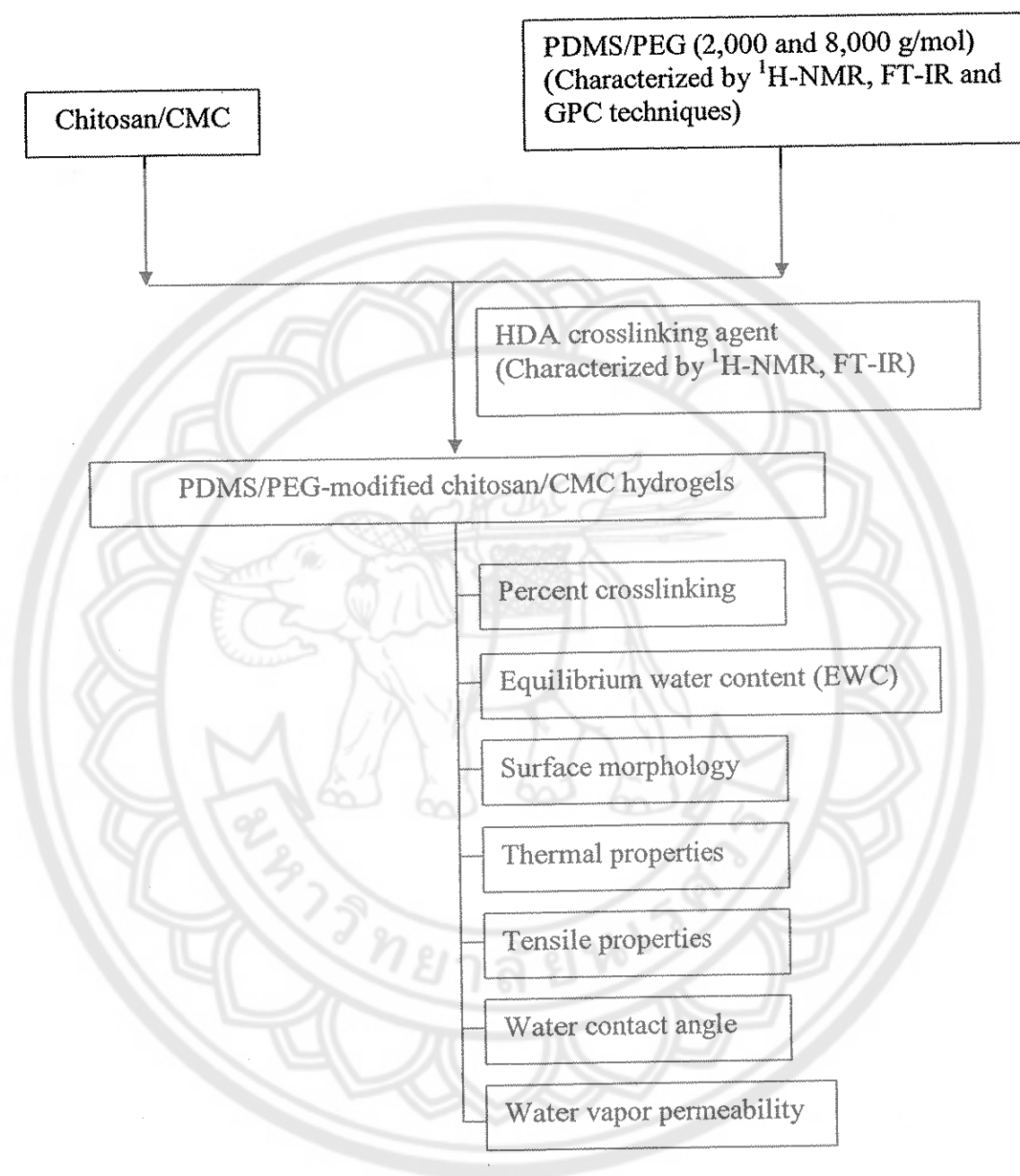
3. The properties of interest of the modified hydrogels were investigated. Such properties include;

- 3.1 Percent crosslinking
- 3.2 Water swelling properties
- 3.3 Surface morphology
- 3.4 Thermal properties
- 3.5 Tensile properties
- 3.6 Water contact angles
- 3.7 Water vapor permeability

**Table 6** Polymers used to modify chitosan/CMC hydrogels

Type of polymer used	Molecular weight (g/mol)	Abbreviated name	Number of repeating units (n)
PDMS	2,000	2K	27
	8,000	8K	108
PEG	2,000	2K	45
	8,000	8K	181

The overview of the current work is depicted in Figure 14.

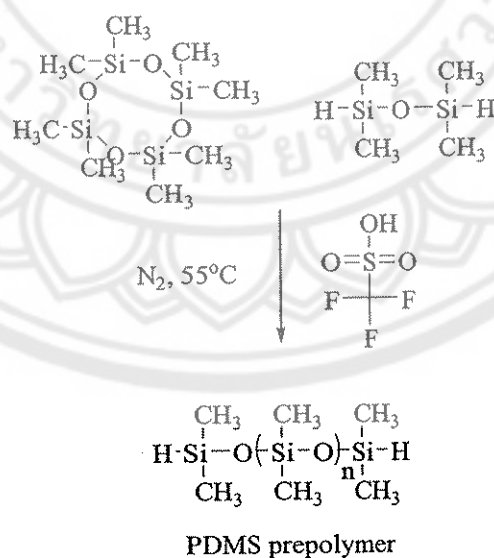


**Figure 14 Research overview for preparing PDMS/PEG-modified chitosan/CMC hydrogels and their characterization**

## 1. Synthesis of polydimethylsiloxane (PDMS)

### 1.1 Synthesis of dihydrogen-terminated polydimethylsiloxane (PDMS prepolymer)

PDMS prepolymer with targeted  $\overline{M}_n$  of 2K is provided (Figure 15). The PDMS prepolymer was synthesized *via* a ring-opening polymerization of octamethylcyclotetrasiloxane (D<sub>4</sub>) (20 g, 0.068 mol) with 1, 1, 3, 3-tetramethyl-disiloxane (1.77 ml, 0.01 mol) as an endcapping agent to control the molecular weights and obtain PDMS with Si-H terminal. When the temperature increased to 55 °C, triflic acid (0.13 ml, 0.65 wt% based on siloxane monomers) was slowly added *via* a syringe, under filled N<sub>2</sub> for 72 h. After the reaction, diethyl ether was added into the solution and it was then washed with distilled water repeatedly to neutralize the mixture. Anhydrous magnesium sulfate as a drying agent was added into the solution, then it was filtered through a filter paper. The solvent was removed by rotavapor evaporator and an excess D<sub>4</sub> monomers remaining from the equilibrium were removed under reduced pressure at 100 °C overnight. PDMS 8K prepolymer was synthesized in the similar fashion but using an appropriate molar ratio of D<sub>4</sub> monomer to the endcapper.



**Figure 15 Synthetic scheme of dihydrogen-terminated polydimethylsiloxane (PDMS prepolymer)**

## 1.2 Synthesis of dihydroxy-terminated polydimethylsiloxane (PDMS diol)

PDMS diol was synthesized via **hydrosilylation** reaction of PDMS **prepolymer** with allyl alcohol (Figure 16). PDMS **prepolymers** 2K (20 g, 0.01 mol) were slowly added into an excess of allyl alcohol (2.04 ml, 0.03 mol) in the presence of Karstedt's catalyst (0.08 ml) with **stirring** at 65°C, under filled N<sub>2</sub> for 2 h. Excess allyl alcohol was removed under reduced pressure at 100 °C for 4 h. <sup>1</sup>H NMR and FT-IR were used to verify its chemical structure and functional groups.



Figure 16 Synthesis of **dihydroxy-terminated** polydimethylsiloxane (PDMS diol)

## 2. Synthesis of hexamethylene-1,6-di-(aminocarboxysulfonate) (HDA) as a crosslinking agent

In a round-bottom flask containing a magnetic bar, 1,6-hexamethylene diisocyanate (HDI) was added to Na<sub>2</sub>S<sub>2</sub>O<sub>5</sub> dissolved in water and stirred for 24 h at room temperature (Figure 17). The product was precipitated in acetone and filtered. Insoluble polymeric by-product was removed by dissolving the HDA product in water, followed by filtration. The product was re-precipitated in acetone, dried in vacuum, and resulted in white powder.

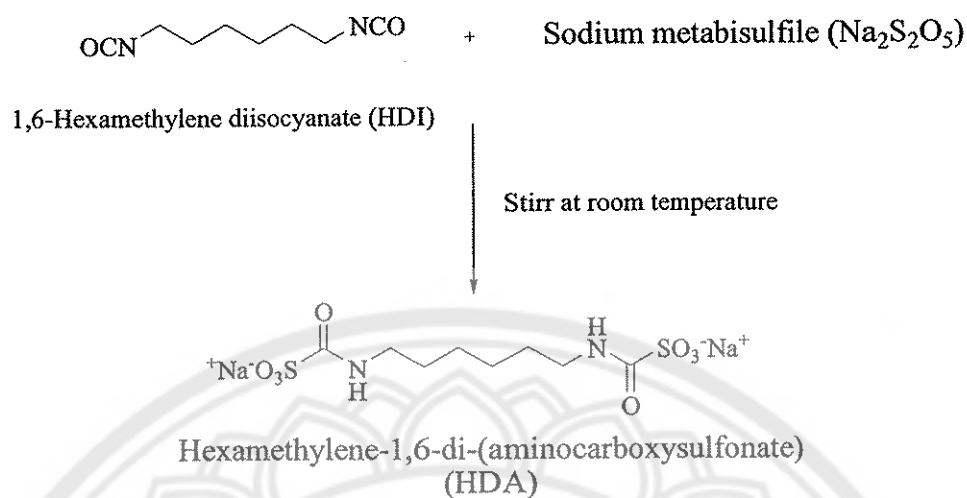


Figure 17 Synthesis of hexamethylene-1,6-di-(aminocarboxysulfonate) (HDA)

### 3. Synthesis of carboxymethylchitosan (CMC)

Carboxymethylchitosan (CMC) oligomer from crap (40 g) was prepared first immersed into isopropanol (500 ml) for 24 h. The swollen CMC was then submerged in NaOH solution (40.32 g in 100.8 ml  $\text{H}_2\text{O}$ ) for 75 min. Subsequently, it was reacted with monochloroacetic acid (48 g in 100 ml  $\text{H}_2\text{O}$ ) at  $60^\circ\text{C}$  for 5 h (Figure 18). After the reaction, the product was precipitated in an excess of methanol, washed with methanol: $\text{H}_2\text{O}$  solution (70:30 and 80:20 v/v, respectively) to remove salt, filtered and dried at  $40^\circ\text{C}$ . The final product appeared as a dried yellow powder and was characterized by FT-IR.

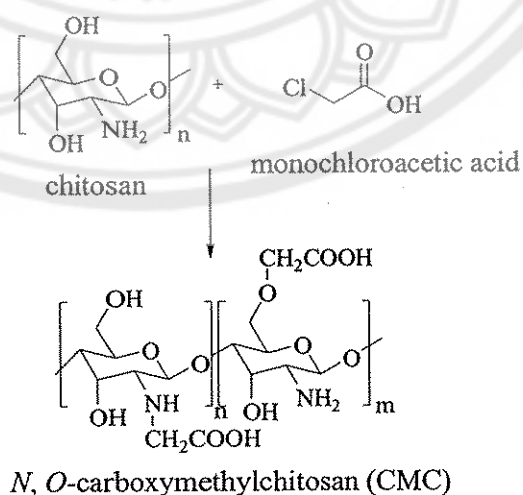


Figure 18 Synthesis of carboxymethylchitosan (CMC)

#### 4. Formation of PDMS/PEG-modified chitosan/CMC hydrogels

##### 4.1 PDMS-modified chitosan/CMC hydrogels

In a 100 ml round-bottom flask containing an excess of HDA dissolved in a mixed solvent of THF:H<sub>2</sub>O (50:50 v/v), a given amount of PDMS diol solution (THF:H<sub>2</sub>O 50:50 v/v) was slowly added and stirred at 40 °C for 1 h. To form Semi-IPN structure, the PDMS/HDA mixture was added into acidic aqueous solution of chitosan or CMC in DI water and stirred 5 min. The solution was then cast onto a glass mold and dried at 30°C for 2 days.

##### 4.2 PEG-modified chitosan/CMC hydrogels

In a 100 ml round-bottom flask containing an excess of HDA dissolved in a water, a given amount of PEG was slowly added and stirred at 40 °C for 1 h. To form semi-IPN structure, the PEG aqueous solution was added into acidic aqueous solution of chitosan or CMC in DI water and stirred 5 min. The solution was then cast onto a glass mold and dried at 30°C for 2 days.

#### Characterization

##### 1. Characterization of polymers

1.1 Proton Nuclear Magnetic Resonance (<sup>1</sup>H NMR) was performed on a 400 MHz Bruker NMR spectrometer using CDCl<sub>3</sub> as a solvent.

1.2 Fourier Transform Infrared Spectroscopy (FT-IR) spectra were measured using a KBr disk method *via* a Perkin-Elmer Spectrum GX0 Series FT-IR Spectrophotometer.

1.3 Gel Permeation Chromatography (GPC) was conducted on PLgel 10 mm mixed B2 columns with a refractive index detector. THF was used as a solvent with a flow rate of 1 ml/min at 30 °C.

1.4 Differential Scanning Calorimetry (DSC) was performed on a Perkin-Elmer Pyris-1 DSC. Rate of scan 10 °C/min, between -140 °C to 90 °C.

## 2. Determination of percent crosslinking [37]

To determine percent crosslinking, sample films with the dimension of 1 x 1 cm<sup>2</sup> were submerged into 1 M acetic acid in the case of chitosan or distilled water in the case of CMC and stirred at room temperature for 48 h to dissolve uncrosslinked chitosan/CMC and HDA. The undissolvable hydrogels were filtered and washed with distilled water and acetone for remove uncrosslinked portion. The swollen gels were then dried at 30 °C for 24 h. Percent crosslinking was calculated as following:

$$\text{Percent crosslinking (\%)} = (W_2 / W_1) \times 100$$

where  $W_1$  and  $W_2$  are the dried weights of the samples before and after the dissolution, respectively. The reported values are the average of at least three different measurements.

## 3. Equilibrium water content measurement [37]

The pre-dried samples were precisely weighed and immersed in 1 M NaOH aqueous solution in the case of chitosan or 1 M acetic acid in the case of CMC for 1 h and then submerged in distilled water at room temperature for 48 h. The swollen samples were then removed from water, wiped off excess water on their surface and weighed. Percent of equilibrium water content was calculated by:

$$\text{Percent of equilibrium water content (\%EWC)} = \frac{W_s - W_d}{W_d} \times 100$$

where  $W_s$  and  $W_d$  are the weights of the swollen and dried samples, respectively.

## 4. Surface morphological studies

Morphological studies of sample surface were carried out through LEO 1455 VP scanning electron microscopy (SEM) with an accelerating voltage of 5 kV. Sample films were dried at 30°C for 1 day. They were cut into 1 x 1 cm<sup>2</sup> in size, adhered onto an aluminum stub and coated with gold.



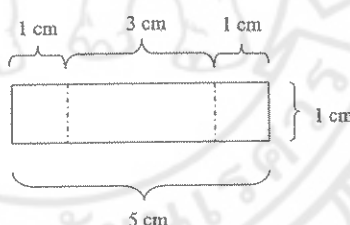
### 5. Determination of tensile properties

Tensile strength and elongation properties were performed on a Universal Testing Machine (Testing machine model WDW-5E). The film samples were cut into a rectangular shape with 1.0 cm width (ASTM D882) (Figure 19). The samples were performed at the 30 mm gage length with the crosshead rate of 10 mm/min and 1 kN load cell. The reported values are the average of five different measurements. Tensile strength and percent elongation at break were calculated as follows:

$$\text{Tensile strength (N/mm}^2\text{)} = \frac{\text{Breaking force (N)}}{\text{Cross-section area of the sample (mm}^2\text{)}}$$

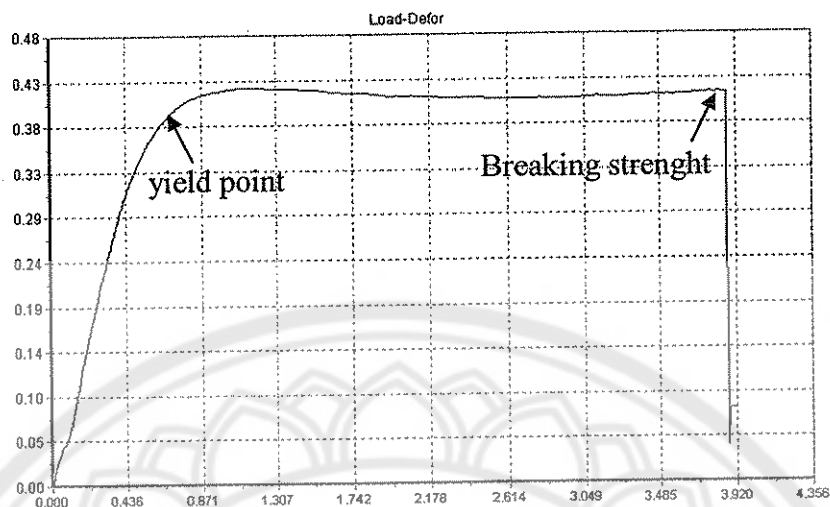
$$\text{Percent elongation at break (\%)} = \frac{\text{The increase in length at breaking point (mm)}}{\text{Original length (mm)}} \times 100$$

A rectangular shape of the film samples was cut with 1.0 cm width and 5.0 cm length for tensile strength and elongation properties measurement.



**Figure 19** The rectangular shape of the film samples for tensile and elongation measurement

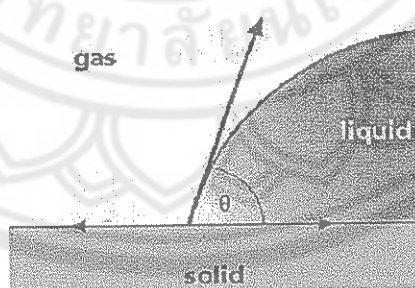
A typical stress-strain curve is illustrated in Figure 20.



**Figure 20** A representative stress-strain curve of 20% PEG 2K-modified chitosan hydrogel

#### 6. Water contact angle measurement

Contact angles ( $\theta$ ) between water and sample films were measured at room temperature [37] using the sessile method on a STANDARD GONIOMETER model 200-F1. A drop of water was carefully applied on a sample film and a contact angles was quickly determined before the film commenced to swell. The reported values are the average of five different measurements.



**Figure 21** Measurement of a contact angle ( $\theta$ ) between a chitosan/CMC film and a water drop

### 7. Determination of water vapor permeability

A measurement procedure of water vapor permeability was described by Khan [5]. A Glass vial was filled with precise weight of anhydrous calcium chloride ( $\text{CaCl}_2$ ). The sample films were cut in a round shape and tightly adhered onto the top of vials with an approximate volume about  $25 \text{ cm}^3$ , while glass vials for control contained glass bead of the weight approximately identical weights to that of the sample vials. The vials were kept in a close desiccator containing saturated NaCl solution at  $30 \pm 3 \text{ }^\circ\text{C}$  and  $70 \pm 5\% \text{ RH}$  for 14 days in close desiccator. Rate of water vapor permeability was reported in gram/day/litre unit and calculated as follows:

$$\text{Rate of water vapor permeability (g/d/L)} = \frac{[(S_f - S_i) - (C_f - C_i)]}{14\nu} \times 1000$$

where  $S_i$  and  $S_f$  are initial and final weights (g) of sample vials.  $C_i$  and  $C_f$  are initial and final weights of control vials.  $\nu$  is the volume of glass vial. The reported values are average of three different measurements.