CHAPTER II
LITERATURE REVIEWS

2.1 Modification of Biopolymer

Tsukada *et al.* (1992) modified silk fibers with aromatic acid anhydrides, such as phthalic and o-sulfobenzoic anhydrides through acylation reaction. They examined the reactivity of these modifying agents toward silk fibers, the physical and thermal properties, and the dyeing behavior with acid and cationic dyes. They found that the o-sulfobenzoic anhydride was more reactive toward silk fibroin than phthalic anhydride. The modification of silk with o-sulfobenzoic anhydride caused a drastic reduction of acid dye uptake and enhanced the affinity of silk for cationic dye. Silk fibers modified with phthalic anhydride showed on differential scanning calorimetry (DSC) curves a minor and broad endothermic peak at around 210°C, attributed probably to the breaking of the crosslink formed between adjacent fibroin molecules.

Gotoh *et al.* (1993) modified silk fibroin (SF) with 2-*O*-[methoxy(polyethylene glycol)]-4,6-dichloro-8-triazine (actPEG1) at 4°C. The nucleophilic ε-amino group of the lysine residue, the imidazole group of the histidine residue, and the phenolic hydroxyl group of the tyrosine residue in SF reacted with actPEG1. The conformation of PEG1-SF in a solid state was examined by IR and X-ray measurement. They found that the PEG molecules covalently bonding to SF narrowed the spacing of the interchain periodicity and promoted the formation of the interchain β-sheet in the solid state.

Tsukada *et al.* (1994) investigated the physical properties of silk fibers modified with dibasic acid anhydride. Silk fibroin fibers were treated with 10% (w/v) succinic and glutaric anhydrides in dimethylformamide (DMF) for different times. They found that the amount of the basic amino acid residues slightly decreased following modification with both succinic and glutaric anhydrides.

Freddi *et al.* (1999) prepared silk fibroin (SF)/polyacrylamide (PAM) blend films by using the conventional casting method. Physical properties and morphology of the blend films were studied by means of various thermoanalytical techniques

(DSC, TG, TMA, and DMA), tensile measurements, and scanning electron microscopy (SEM). Moreover, FTIR was used to study the specific molecular interactions between SF and PAM. They found that the formation of intermolecular hydrogen bonds between the primary amide group of PAM and the amide and various side chain groups of SF. Then the blend films with low PAM content (≤25%) exhibited increased thermal stability and slightly improved mechanical properties compared to the pure SF film.

Cho *et al.* (2003) prepared silk sericin (SS) which reacted with activated polyethylene glycol (PEG). They obtained self-assembled SS nanoparticles which were prepared by the diafiltration method. They used amino acid analysis and H^1 NMR spectroscopy to clarify residues as the reaction site in SS. IR spectroscopy and circular dichroism (CD) measurements showed introduction of PEG into SS that changed the conformation from random coil to β-sheet. DSC thermogram is used for miscibility between PEG and SS chains. For nanoparticles of sericin–PEG conjugate with sizes and shape are observed by dynamic light scattering and scanning and transmission electron microscopes, respectively. The results suggest that sericin–PEG conjugates are self-associated to form spherical nanoparticles through hydrophobic interaction.

Teramoto *et al.* (2004) modified silk sericin with 4-cyanophenyl isocyanate using the LiCl/DMSO solvent system. FTIR analysis indicated that most 4-cyanophenyl groups were connected with sericin molecules at hydroxyl side chains through urethane linkages. They investigated solubility characteristic, hygroscopic property, and thermal stability of the modified sericins. They found that LiCl/DMSO is a good solvent of silk sericin. The modification rendered sericin less hygroscopic and thermally unstable than before modification.

Gotoh *et al.* (2004) prepared chemical modification of silk fibroin (SF) with lactose using cyanuric chloride (CY) as a coupling spacer. The covalent immobilization of lactose into SF was investigated by using ^{1}H-NMR measurement. The ^{1}H-NMR spectrum of Lac-CY-SF showed new board peaks attributed to methane and methylene protons of lactose. Their product was used in a scaffold for hepatocyte attachment.

2.2 Nanoparticles and Nanofibers of Biopolymer

Cho *et al.* (2003) prepared self-assembled SS nanoparticles which were prepared by the diafiltration method. For nanoparticles of sericin–PEG conjugate with sizes and shape were observed by dynamic light scattering and scanning and transmission electron microscopes, respectively. The results suggest that sericin–PEG conjugates were self-associated to form spherical shape, and size ranged about 200-400 nm in diameter.

Ohgo *et al.* (2003) prepared non-woven nanofibers of *Bombyx mori* and *Samia cynthia ricini* silk fibroins, and of the recombinant hybrid fiber involving the crystalline domain of *B. mori* silk and non-crystalline domain of *S. c. ricini* silk from hexafluoroacetone (HFA) solution using electrospinning method. In the electrospinning process, they used Pt wire as an electrode and applied voltage at 15-30 kV to the wire. The collecting mesh was placed at distance of 10-15 cm from the capillary tip. They found that *B. mori* silk solution of 7, 5 and 3 wt% formed a stable jet, and 1.0-1.6 kV/cm electronic field was condition for the fiber formation. SEM was used to determine the diameters and their distributions of the fibers. They obtained thin and rod-like fibers with diameters ranged from about 100-1000 nm. The best condition for nanoscale non woven *B. mori* fiber formation was 3 wt% concentration and 1 kV/cm electronic field.

Zarkoob *et al.* (2004) produced nanoscale fibers of *Bombyx mori* and *Nephila clavipes* in hexafluoro-2-propanol. The concentration of *B. mori* and *N. clavipes* were prepared at 0.74 and 0.72-1.2 %wt, respectively. In electrospinning process, they set the collecting mesh from the syringe tip about 15 cm and applied voltage at 24-30 kV. The electrospun fibers were observed by optical, scanning electron, and transmission electron microscope. The overall size range was approximately 6.5-100 nm with 25 nm being the most frequent.

Huang *et al.* (2004) investigated electrospinning of biopolymer, gelatin, and the mass concentration-mechanical property relationship of the resulting nanofiber membranes. In the electrospinning process, they applied voltage in a range of 0-50 kV and used a constant mass flow rate of 0.8 ml/h. The resulting solution with a

mass concentration in between 5 and 12.5% can be successfully elctrospun into nanofibers of a diameter in a range from 100 to 340 nm.

Park *et al.* (2004) blended silk fibroin (SF) and chitosan (CS) in different composition ratios and could be electrospun into the continuous fibrous structure. They prepared 12 and 3.6 wt% of the concentration of SF and CS solution in formic acid, respectively. The SF/CS blend solutions with different mixing ratios (100/0, 90/10, 80/20, 70/30, 60/40, 50/50, and 0/100 SF/CS, w/w) were prepared for electrospinning. In the electrospinning process, they applied voltage 16 kV and used flow rate of polymer solution at 1.0 ml/h. They found that the SF/CS blends containing up to the CS content of 30% could be electrospun into the continuous fibrous structure and their diameters gradually decreased up to 100 nm with the addition of CS.