

CHAPTER IV

RESULTS AND DISCUSSION

4.1 Characterization of Chitosan

Shrimp shells generally compose of three major components, which are chitin, calcium carbonate, and protein. By solvent extraction, calcium carbonate and protein can be removed and leaving behind chitin as a white flaky portion.

In this research, chitin was prepared from shells of *Penaeus merguensis* shrimp by decalcification with hydrochloric acid solution and deproteinization with aqueous sodium hydroxide solution in order to remove calcium carbonate and protein, respectively. Because of the difficulty to dissolve chitin in common solvents, chitin is preferably derived further to chitosan in order to achieve the ease of dissolution and chemical modification. Chitin was deacetylated in 50% w/w of sodium hydroxide solution and chitosan with mainly reactive amino groups would be obtained.

The FTIR spectrum of chitosan is shown in Figure 4.1 and the absorption frequencies of characteristic bands of chitosan are summarized in Table 4.1.

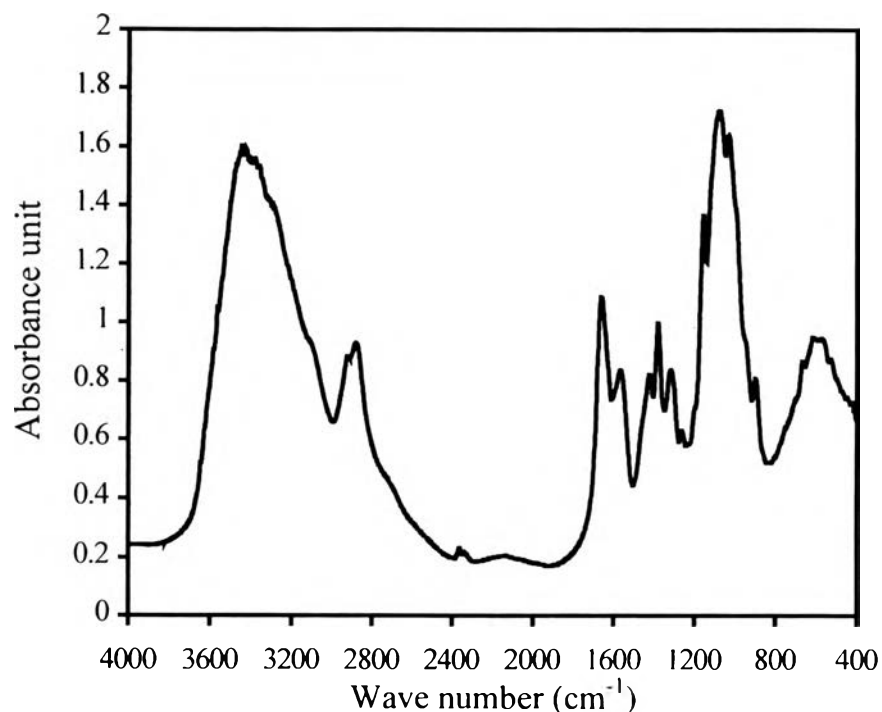


Figure 4.1 FTIR spectrum of chitosan.

Table 4.1 FTIR characteristic Absorption bands of chitosan.

Frequencies (cm ⁻¹)	Assignment and remarks	Reference(s)
1659	C=O stretching	Muzzarelli, 1977;
1561	NH deformation	Kurita <i>et al.</i> , 1993;
1317	CN band and CH ₂ wagging	Kim, <i>et al.</i> , 1996
1080 and 1031	C-O stretching vibration	Kim, <i>et al.</i> , 1994

Chitosan normally has greater extent of amino groups than acetamide groups at C₂ position of N-acetyl glucosamine units. The degree of deacetylation of chitosan depends on the severity of the deacetylated condition. According to the method of Sannan (1978), the degree of deacetylation of chitin can be calculated from FTIR to obtain to value of 80%.

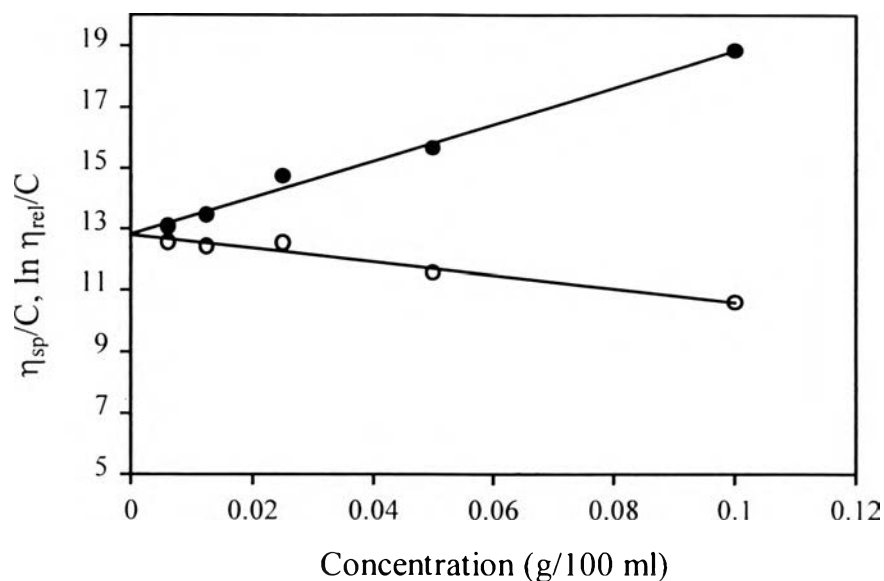


Figure 4.2 The plot of reduced viscosity $\left(\frac{\eta_{sp}}{C}\right)$ and $\left(\ln \frac{(\eta_{rel})}{C}\right)$ versus concentration of chitosan solution. ● $\frac{\eta_{sp}}{C}$ ○ $\ln \frac{(\eta_{rel})}{C}$

Chitosan is a biopolymer. The molecular weight yield a wide range of chitosan varies from 1.0×10^5 to 1.2×10^6 depending on the nature of chitosan resources and the severity of the deacetylation process. In this study, chitosan was prepared from chitin, which was obtained from *Penaeus merguensis* shrimp shells. The molecular weight of chitosan was determined by viscometric method. By following the method of Lee (1978), the molecular weight of chitosan was derived from its intrinsic viscosity. The plot of reduced viscosity $\left(\frac{\eta_{sp}}{C}\right)$ and $\ln \frac{(\eta_{rel})}{C}$ versus concentration of chitosan solution is shown in Figure 4.2. This plot shows the extrapolated value of each line reaches the same position and this value was referred to intrinsic viscosity of chitosan. From Mark-Houwink equation, when a is 0.71 and k is 8.93×10^{-4} , the viscosity-average molecular weight of chitosan obtained from the calculation was 7.12×10^5 .

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4.2 Characterization of Chitosan/Silk Fibroin Blend Films

4.2.1 FTIR Spectra Characterization

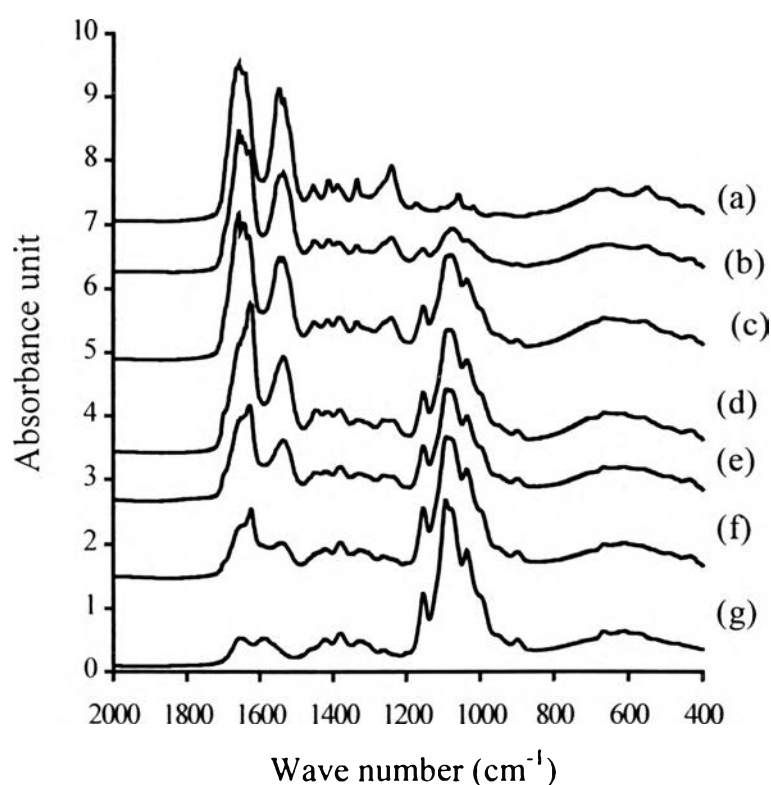


Figure 4.3 FTIR spectra of pure and blend films at various compositions of chitosan to silk fibroin with the addition of 0.01 mole of glutaraldehyde /glucosamine unit of chitosan. Chitosan:silk fibroin (a) 0/100:(b) 20/80: (c) 4/6: (d) 50/50: (e) 60/40: (f) 80/20: (g) 100/0

The molecular character of chitosan/silk fibroin blend films was examined by infrared spectroscopy. The FTIR spectrum of pure silk fibroin film is shown in Figure 4.3(a). The characteristic peaks of silk fibroin at 1663 cm^{-1} , 1547 cm^{-1} and 1242 cm^{-1} were attributed to amide I, amide II, and

amide III of random coil conformation. While the characteristic absorption bands of chitosan at 1589 cm^{-1} belonged to amide I band and the others occurred at 1153 cm^{-1} and 898 cm^{-1} were the characteristic peaks of polysaccharide. For the spectrum of the blend film with 80 % silk fibroin content, the amide I band of silk fibroin showed another peak at 1625 cm^{-1} that was assigned to the β -sheet structure of silk fibroin (Yoshimizu and Asakara, 1990). As seen in Figure 1, the absorption band at 1625 cm^{-1} became stronger with increase of chitosan content. This agrees with the results reported by Chen *et al.* (1997) who concluded that the increasing of β -sheet conformation of silk fibroin was caused by the hydrogen bond formation between amino groups of chitosan and amide groups of silk fibroin. These results reveal that in the blend films some originally random coil of silk fibroin chains changes to β -sheet structure during blending with chitosan. The occurrence of β -sheet structure leads to more regular structure of silk fibroin chains.

4.2.2 Crystallinity

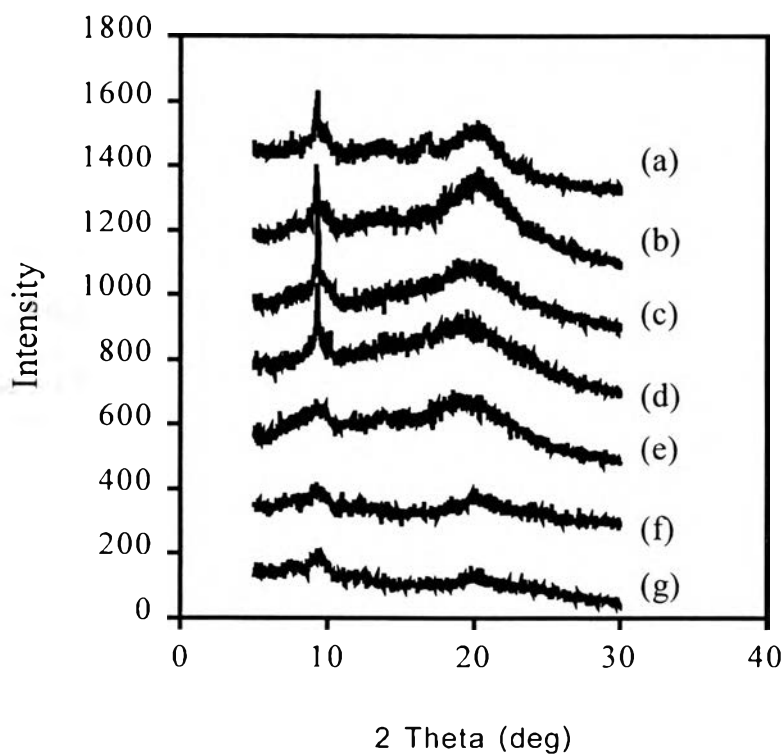


Figure 4.4 Wide-angle X-ray diffraction profile of pure and blend films. Chitosan : silk fibroin (a) 100/0: (b)80/20: (c) 60/40: (d) 50/50: (e) 40/60: (f) 20/80: (g) 0/100

The X-ray diffraction patterns of pure and the blend films are shown in Figure 4.4. The pure chitosan film exhibited the diffraction pattern of partial crystalline structure with 2θ peaks occurring at 9.35° and 20° . The silk fibroin film showed a non-crystalline structure. According to Freddi *et al.*, (1995), the dissolution of silk fibroin was caused by reagent penetrating into the adjacent chains and breaking hydrogen bonds between them. This led to the decrease in crystallinity of silk fibroin films as compared to the original silk fibroin fiber. The pattern of the blend films exhibited a gradual transformation from characteristic crystalline peaks of chitosan to the completely amorphous pattern of silk fibroin with diffraction pattern increase

of silk fibroin content in the blend film. Chen *et al.* (1995) studied the crystallinity of pure silk fibroin membrane without further treatment. The diffraction pattern of pure silk fibroin membrane showed no clear 2θ peak. This includes a mainly random coil conformation that is similar to present results. According to Hasegawa *et al.* (1992), the chitosan film showed the crystalline peaks at around 10° and 20° and the blend film of chitosan/cellulose showed gradual transformation between chitosan and cellulose. The analysis of Hasegawa *et al.* corresponds to our results. However, these all diffraction patterns did not give clear information about the crystallinity because the crystalline structures of chitosan and silk fibroin were remarkably frustrated during dissolution process. Therefore, the chitosan/silk fibroin blend films were mainly amorphous.

4.2.3 Thermal Stability

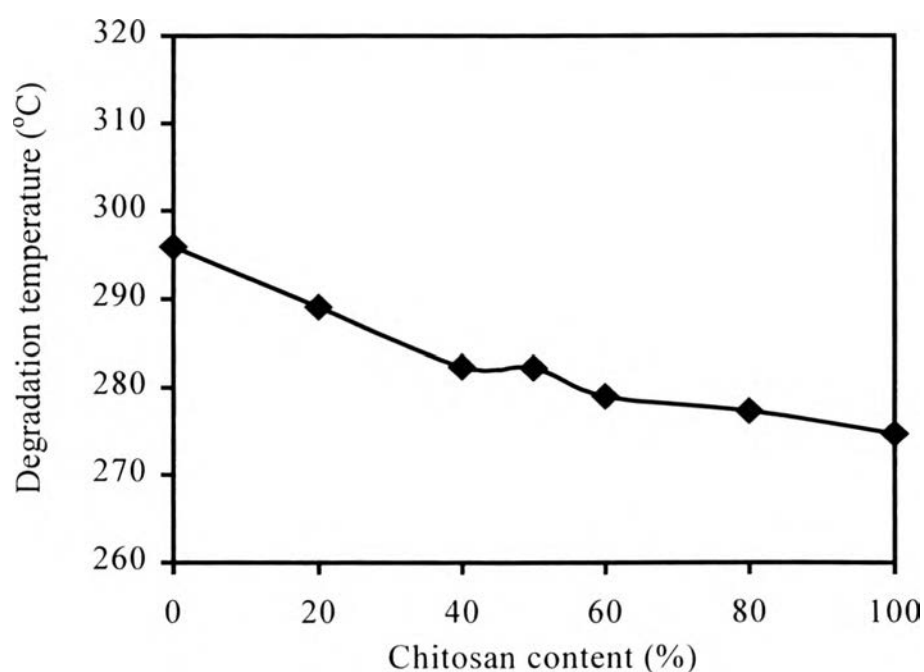


Figure 4.5 Thermal decomposition temperature of pure and blend films.

The thermal stability is an important functional property, which can definitively determine the performance of a material for practical uses. The degradation temperature of pure and the blend films as a function of chitosan content is shown in Figure 4.5. Pure chitosan and silk fibroin films were stable until at 274.68 °C and 295.87 °C, respectively. Then their weights dropped sharply resulting from the thermal decomposition of pure chitosan and pure silk fibroin. The thermal stability of the blend films falls between pure chitosan and pure silk fibroin films. The decomposition temperature of the blend films increased with increase of silk fibroin content. From this thermal behavior, the thermal stability of pure silk fibroin films was better than pure chitosan film. By blending with silk fibroin, the thermal stability of the blend film could be improved as compared to pure chitosan film. These results agree with the work of Tsukada *et al.* (1994) who carried out thermogravimetric analysis of *Antheraea pernyi/Bombyx mori* silk fibroin blend films. The thermal behavior of *Antheraea pernyi/Bombyx mori* silk fibroin blend films was between the two pure components. However, the decomposition temperature of *bombyx mori* silk fibroin films studied by Tsukada *et al.* (1994) was slightly different from the results in this study. This may be due to the difference in solvent system for dissolution and films preparation.

4.3 Swelling Study

4.3.1 Equilibrium Water Content

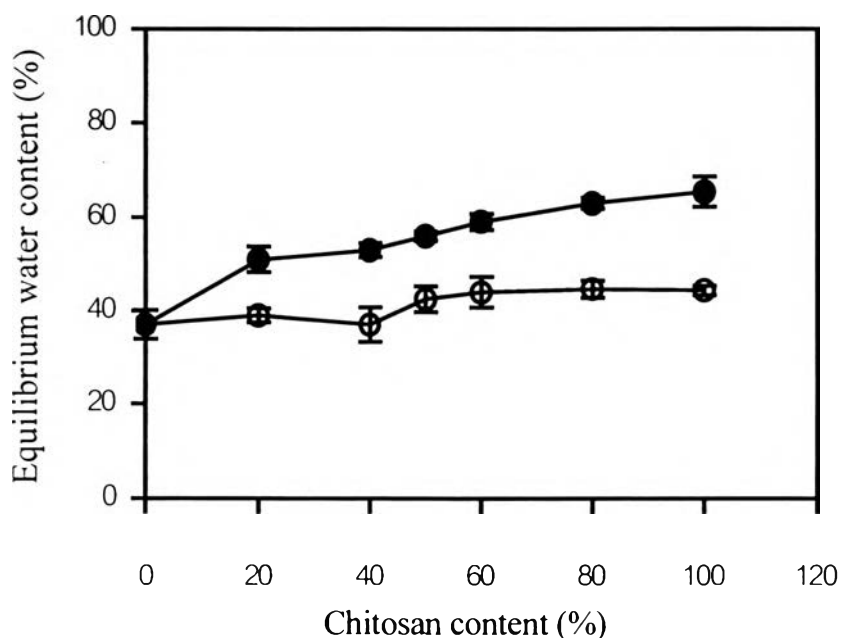


Figure 4.6 Equilibrium water content of chitosan/silk fibroin blend films.

○ blend films without crosslinking; ● crosslinked blend films.

The equilibrium water contents of the blend films with and without crosslinking are shown in Figure 4.6. The equilibrium water content increased as chitosan content increased. This may be due to the hydrophilicity of chitosan. In the case of the crosslinked blend films, they exhibited the lower in equilibrium water content as compared to the blend films without crosslinking. The reason for this is that the crosslinks restrict the swelling of the blend films because chitosan chains are held together by crosslinks. This decreases the amount of water molecules that can penetrate inside the blend films. Pure silk fibroin film showed the lowest equilibrium water content owing to its molecular structure. The major amino acid

components of *bombyx mori* silk fibroin are glycine and alanine. These amino acids have hydrophobic side chains (glycine-H, alanine-CH₃) (Shen *et al.* 1998). Therefore, the absorbability of pure silk fibroin film was low. Liang *et al.* (1992) suggested that the water absorption of pure silk fibroin film was very low and it could be improved by blending with sodium alginate. This is consistent with our results that water absorption of silk fibroin film was low and that chitosan could improve the water absorbability of silk fibroin.

4.3.2 Effect of pH

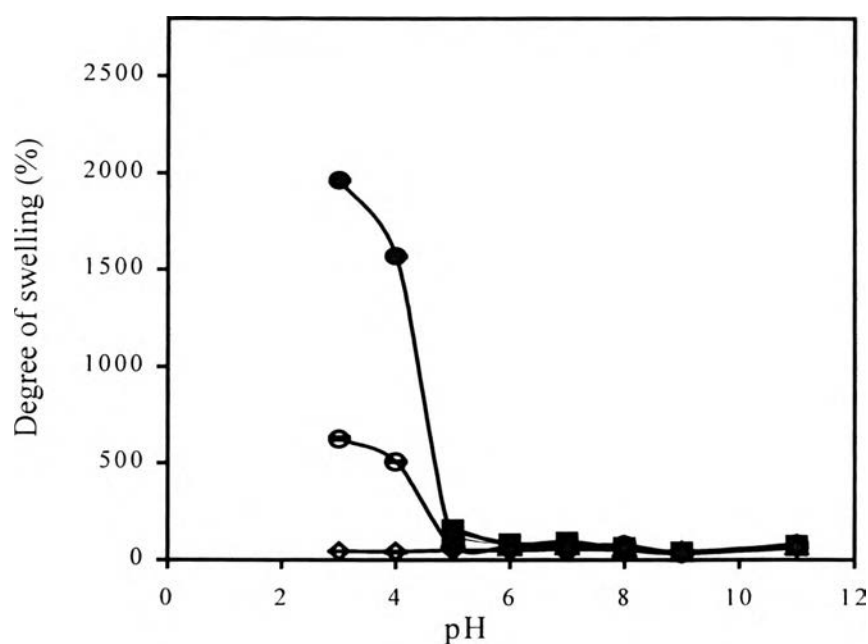


Figure 4.7 The degree of swelling of pure and the blend films without crosslinking in pH buffer solutions. Chitosan : silk fibroin ■ 100/0: □ 80/20: ▲ 60/40: △ 50/50: ● 40/60: ○ 20/80: ◇ silk fibroin

The effect of pH on the degree of swelling of pure and the blend films without crosslinking is shown in Figure 4.7. There was no change in the degree of swelling of pure silk fibroin over the whole pH range from pH 3 to pH 11. Likewise, the degree of swelling of pure chitosan film and the blend

films was nearly constant when pH is higher than 5. However, in the range of pH less than 5, the degree of swelling of the blend films with less than 50 % chitosan content increased sharply proportional to the chitosan content. This can be explained that in the acidic pH range, the amino groups of chitosan are protonated leading to the destruction of hydrogen bonding between amino group of chitosan and amide group in the main chain of silk fibroin (Chen *et al.* 1997). The disappearance of hydrogen bonds between the polymer chains results in swelling of the blend films. It can be said that chitosan caused the blend films to respond to pH change and dissolved. For the blend films with higher than 50 % chitosan content swell rapidly in the acidic medium.

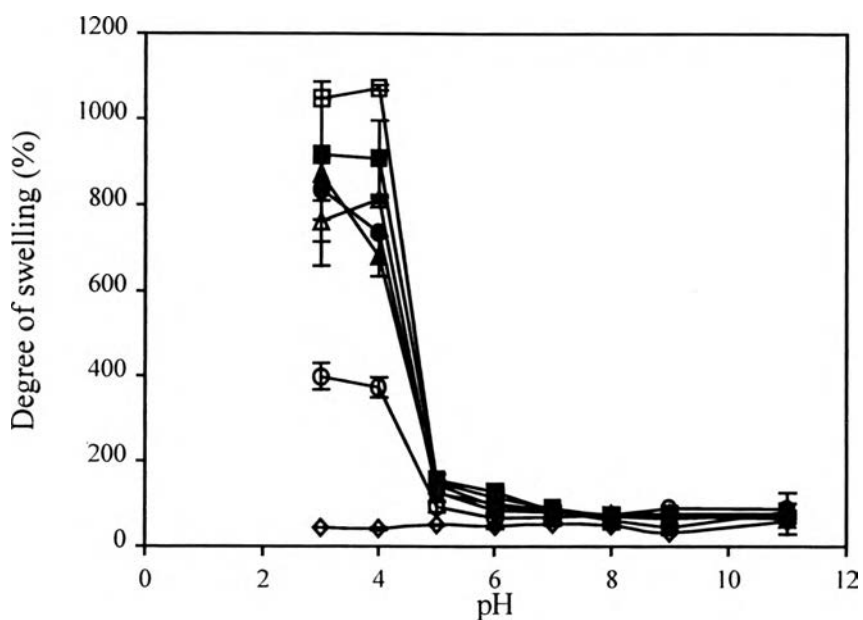


Figure 4.8 The degree of swelling of pure and blend films of chitosan to silk fibroin with crosslinking in pH buffer solution. The amount of added glutaraldehyde was 0.01 mole/glucosamine unit of chitosan

■ chitosan: □ 80/20: ▲ 60/40: △ 50/50: ● 40/60: ○ 20/80:
◇ silk fibroin

The effect of pH on the degree of swelling of pure and the blend films with crosslinking is shown in Figure 4.8. When pH was less than 5, the degree of swelling of the crosslinked blend films increased with increasing chitosan content. The maximum degree of swelling was observed for the blend film with 80 % chitosan content. This may be due to the dissociation between chitosan and silk fibroin chains caused by the protonation of amino groups of chitosan. Unlike that of the blend films without crosslinking, the crosslinked blend films with higher than 50% chitosan content could maintain their structures in acidic pH range. However, the degree of swelling of the crosslinked blend films was lower than the blend films without crosslinking. Wang *et al.* (1997) reported that the crosslinked chitosan

network could prevent the semi-interpenetrating polymer network of chitosan and poly(acrylic acid) from collapse at low pH but crosslinking could limit swelling of the films. It can be explained that the crosslinking makes chitosan chains covalently link together and form network that can prevent the dissolution of the films even though it restricts the swelling of the films. In contrast, the degree of swelling of the blend films did not exhibit any difference when pH was higher than 5. It can be explained by the fact that, at alkaline pH range, the number of protonated amino groups of chitosan is very low. The pKa of chitosan is about 6.3-6.5 (Hugerth *et al.*, 1997), which indicates that chitosan tends to be protonated in the acidic solution. Thus the degree of swelling of the blend films in alkaline solution was very low as compared to that of the blend films in acidic solution.

From the above, it is evident that the blend films of chitosan and silk fibroin are pH-responsive. This agrees with the study of Chen *et al.* (1997). Moreover, the crosslinking enabled the blend films to maintain their structure in the acidic medium but caused a lower degree of swelling.

4.3.3 Effect of Salt Type

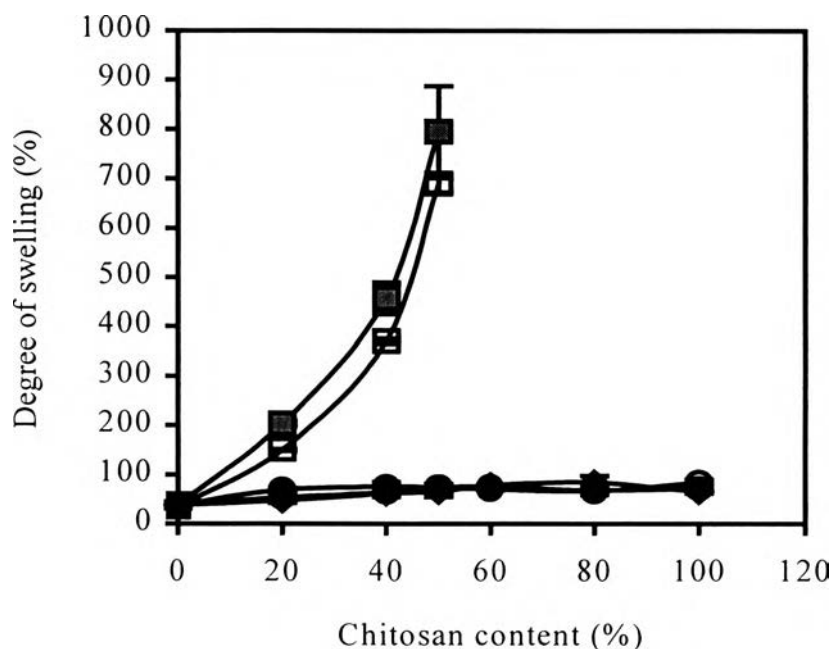


Figure 4.9 The degree of swelling of pure and blend films without crosslinking in various types of salt solutions. □ 0.25 M FeCl₃ solution: ■ 0.25 M AlCl₃ solution: ○ 0.25 M CaCl₂ solution: ● 0.25 M NaCl solution: ◇ 0.25 M LiCl solution

The degree of swelling of pure and the blend films without crosslinking in various types of salt solutions is shown in Figure 4.9. It can be seen that the films exhibited a significant increase in the degree of swelling in 0.25 M AlCl₃ and 0.25 M FeCl₃ solutions as chitosan content increased. This is believed to be due to the formation of coordinated covalent bond between nitrogen at amino group of chitosan and Al³⁺ or Fe³⁺. In solution, Al³⁺ is surrounded by six molecules of water while Cl⁻ acts as a counter ion (Gillespie *et al.*, 1989). This Al³⁺ hydrated still has empty orbital that is electron deficient. While nitrogen atom at amino group of chitosan has a lone pair electron that can coordinate to Al³⁺. So Al³⁺ can form a coordinated

covalent bond with amino group of chitosan resulting in greater degree of swelling at higher chitosan content. The transition metal ion, like Fe^{3+} , can also form a coordinate covalent bond with the amino group of chitosan. The covalent binding of Al^{3+} or Fe^{3+} has the effect of increasing the net positive charge on the chitosan. Therefore, increasing of chitosan content could enhance the degree of swelling of the blend films in both AlCl_3 and FeCl_3 solutions. At chitosan content greater than 50 %, the degree of swelling could not be reported because of overswelling leads to the disintegration of the films.

The swelling of the films in 0.25 M NaCl, 0.25 M LiCl, and 0.25 M CaCl_2 solutions were nearly the same for all blend compositions. The reason is that chitosan does not bind or bind weakly to alkali and alkali earth metal ions such as Na^+ , Li^+ , and Ca^{2+} resulting in lower degree of swelling (Wang *et al.*, 1997).

From Figure 4.9. it was observed that pure silk fibroin film had the lowest degree of swelling. By the addition of chitosan to silk fibroin film, the degree of swelling of silk fibroin could be enhanced. Moreover, the blend films of chitosan and silk fibroin were sensitive to Al^{3+} and Fe^{3+} .

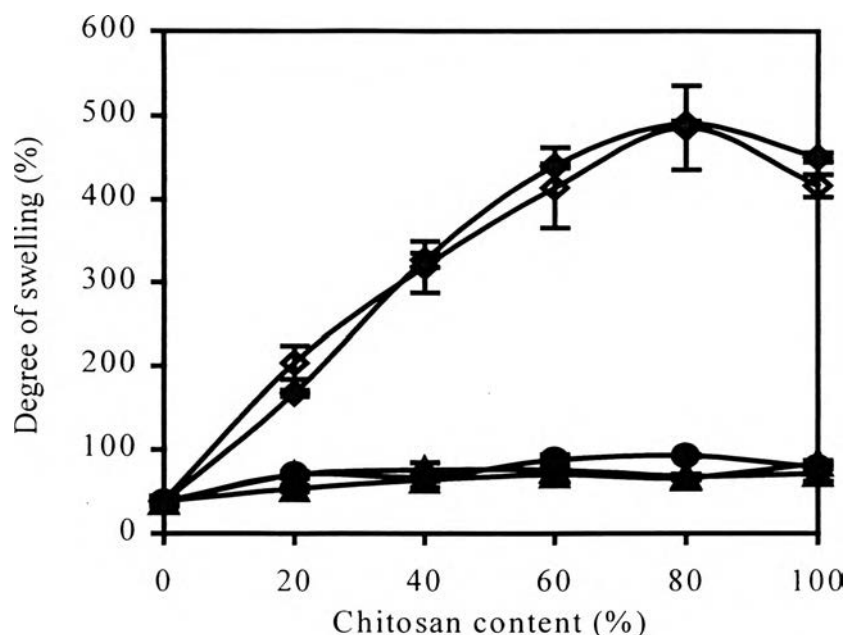


Figure 4.10 The degree of swelling of pure and blend films of chitosan and silk fibroin with crosslinking in various types of salt solutions. The amount of added glutaraldehyde was 0.01 mole/glucosamine unit of chitosan. ◆ 0.25 M FeCl₃ solution: ◇ 0.25 M AlCl₃ solution: ▲ 0.25 M CaCl₂ solution: △ 0.25 M NaCl solution: ● 0.25 M LiCl solution

The degree of swelling of pure and the blend films with crosslinking in various types of salt solutions is shown in Figure 4.10. As chitosan content increases, the degree of swelling of pure and the blend films with crosslinking increases significantly in the case of immersing the films in AlCl₃ and FeCl₃ solutions. The reason has been mentioned already with respect to the results of Figure 4.9. For the blend films with crosslinking, the degree of swelling of blend films with greater than 50 % chitosan content can be observed and the highest degree of swelling is observed at 80 % chitosan content. This indicates that the presence of crosslinks can prevent the disintegration of the films by the formation of network between the chitosan chains. However, the crosslinks limit the swelling ability of the films. The degree of swelling of the crosslinked blend films is lower than that of the

non-crosslinking blend films at the corresponding blend compositions. The degree of swelling of the crosslinked blend films is very low in 0.25 NaCl, 0.25 M LiCl, and 0.25 M CaCl₂ solutions for a whole range of chitosan content.

4.3.4 Effect of AlCl₃ and FeCl₃ Concentrations

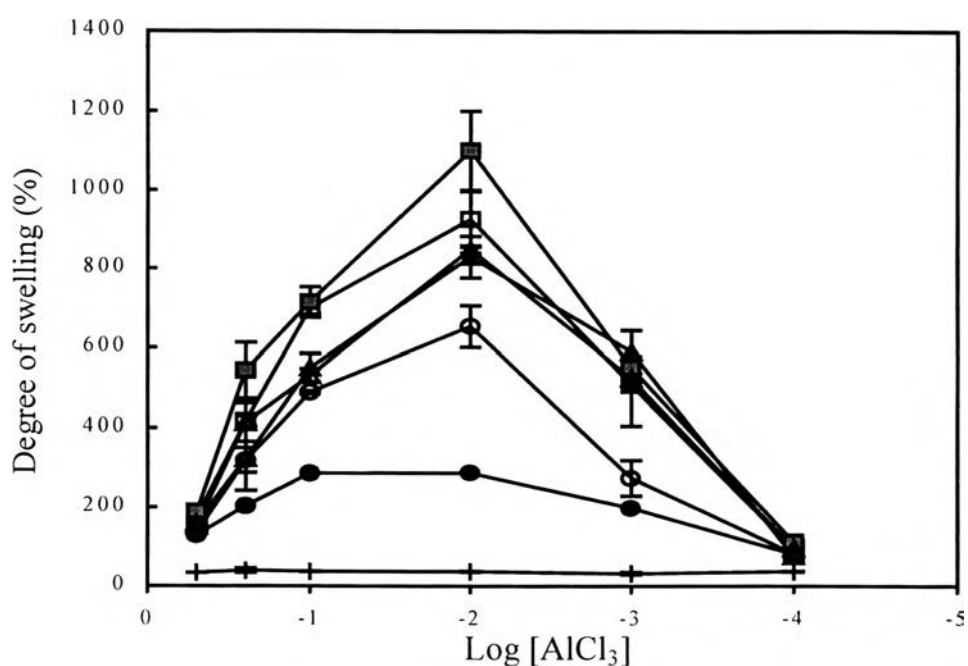


Figure 4.11 The degree of swelling of pure and blend films with crosslinking in various concentrations of AlCl₃ solution. The amount of added glutaraldehyde was 0.01 mole/glucosamine unit of chitosan. Chitosan : silk fibroin ■ chitosan: □ 80/20: Δ 60/40: ▲ 50/50: ○ 40/60: ● 20/80: ◇ silk fibroin

From previous results, chitosan and blend films with crosslinking showed strong swelling in AlCl₃ and FeCl₃ solutions. Therefore, further investigations were carried out to study the effect of AlCl₃ and FeCl₃ concentrations on the swelling behavior of pure and the blend films with crosslinking.

The effect of AlCl_3 concentration on the degree of swelling of pure and the blend films with crosslinking is shown in Figure 4.11. The AlCl_3 concentration did not affect the degree of swelling of pure silk fibroin film at all salt concentrations because of the very low interaction between Al^{3+} and silk fibroin. However, chitosan and the blend films showed a strong variation in the degree of swelling relational to the changes in salt concentration. The degree of swelling increased significantly as AlCl_3 concentration increased. When AlCl_3 concentration was 1.0×10^{-2} M, the degree of swelling of pure chitosan and the blend films showed the maximum value. The maximum degree of swelling belonged to the blend film with 80 % chitosan content. The occurrence of the highest degree of swelling at AlCl_3 concentration of 1.0×10^{-2} M can be explained by Donnan equilibrium effect (Yao *et al.*, 1993). Donnan equilibrium effect arises from the ionic osmotic pressure generated from mobile counterions, which accompany the bound ions on the network strands (Park & Hoffman, 1992). The high counterion concentration causes a large swelling pressure in the absence of excess mobile salt. At the AlCl_3 concentration of 1.0×10^{-2} M, the difference of ion concentration between the interior and exterior of the films is maximal, leading to a large imbalance in osmotic pressure, which causes the maximum degree of swelling of the films. The decrease in the degree of swelling when the AlCl_3 concentration exceeded 1.0×10^{-2} M occurred because of there was now an excess mobile ion concentration in the external solution. The concentration of amino groups of chitosan in the film available to bind Al^{3+} is depleted, no additional Al^{3+} becomes bound to the network strands. the excess mobile ions can penetrate into the film and screen the bound charges, and the degree of swelling decreases.

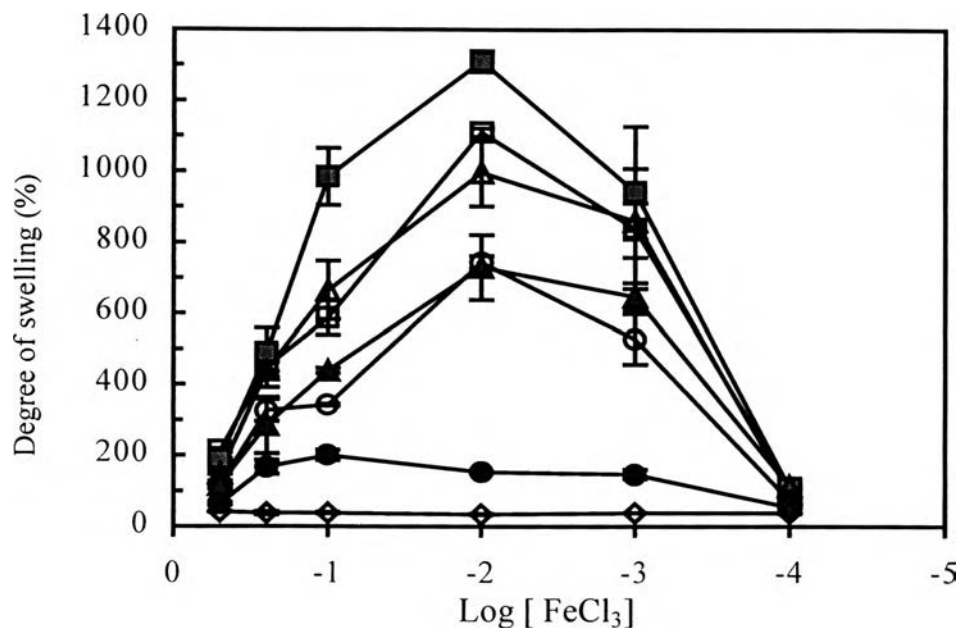


Figure 4.12 The degree of swelling of pure and blend films of chitosan and silk fibroin with crosslinking in various concentrations of FeCl_3 solution.

Chitosan : silk fibroin □ chitosan: ■ 80/20: △ 60/40: ▲ 50/50:
 ○ 40/60: ● 20/80: ◇ silk fibroin

The degree of swelling of pure and blend films with crosslinking in various concentrations of FeCl_3 solution is shown in Figure 12. The effect of FeCl_3 concentration on the degree of swelling of pure and the blend films with crosslinking was similar to that of the films immersed in AlCl_3 solutions. The FeCl_3 concentration that caused the maximum degree of swelling is 1.0×10^{-2} M. The blend film with 80 % chitosan content had the highest degree of swelling. The difference in the degree of swelling of the films immersed in different concentrations of FeCl_3 solution was also caused by Donnan equilibrium effect, as in the case of AlCl_3 solution. The degree of swelling of pure silk fibroin films was not affected by the FeCl_3 concentrations.

4.4 Mechanical Properties

4.4.1 Tensile Strength

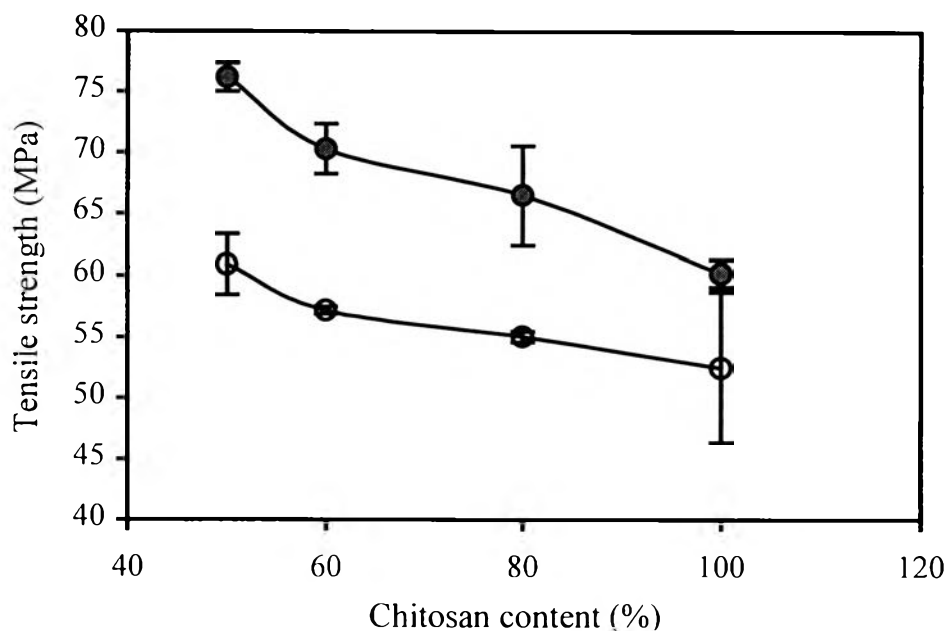


Figure 4.13 The tensile strength in the dry state of pure and blend films.

○ films without crosslinking: ● films with crosslinking

The tensile strength of the films as a function of chitosan content is shown in Figure 4.13. The tensile strength of pure silk fibroin and the blend films with less than 50 % chitosan content could not be monitored by Lloyd tensile tester because of their brittleness. Beyond these compositions, the tensile strength of the blend films containing from 50 % chitosan content could be measured. These results indicated that chitosan imparted the flexibility to the films by lowering the brittleness of silk fibroin when an appropriate amount of chitosan is added to the blend films. Chitosan has hygroscopic property that makes it absorb moisture easily. This absorbed

moisture has a plasticized effect to the blend films resulting in an increase in flexibility. As chitosan content increased, the tensile strength slightly decreased in both with and without crosslinking. It is known that silk fibroin film in the dry state is very brittle. Freddi, *et al.* (1995) found that the addition of cellulose to silk fibroin could enhance the mechanical properties due to the hygroscopicity of cellulose. In Figure 4.13, the crosslinked blend films shows a slightly higher tensile strength than the non-crosslinked blend films in the dry state because the crosslinks play a key role as a bridge that links chitosan chain together. The network structure made the films stronger, and hence they exhibited a slightly higher tensile strength in the dry state. However, the increase in tensile strength also depended on the amount of crosslinking agent added. In this research, the amount of glutaraldehyde added was low resulting in a small increase in tensile strength.

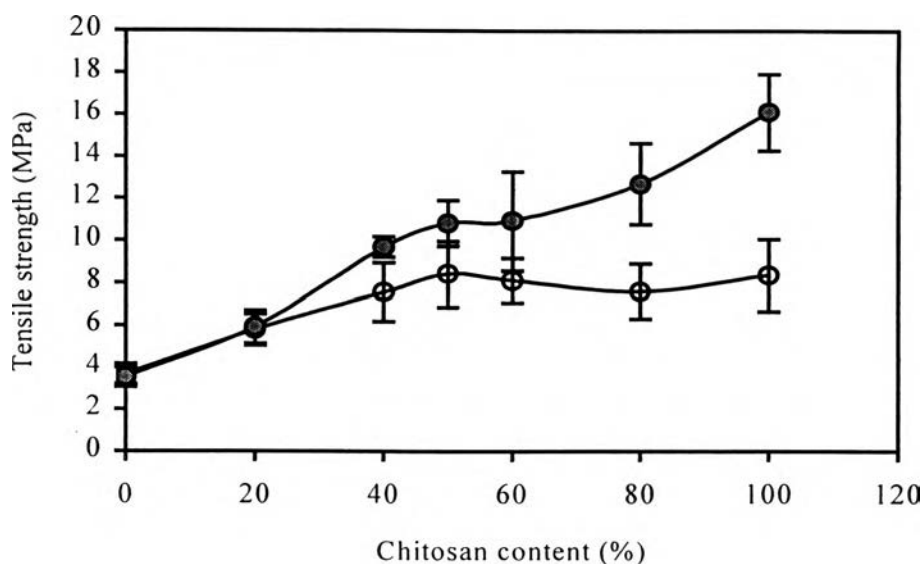


Figure 4.14 The tensile strength in the wet state of pure and blend films.
 ○ films without crosslinking; ● films with crosslinking

Because many end-uses of films involve the contact with water, the strength of films in the wet state is also important. The tensile strength in the wet state of pure and the blend films is shown in Figure 4.14. The tensile strength in the wet state remarkably decreased in comparison with that of the dry state because, in the wet state, the hydrogen bonding between polymer chains within the films is diminished by the water molecules. Hosogawa *et al.* (1991) found that the tensile strength in the wet state of chitosan and homogenized cellulose blend films was lower than that of the dry state. Figure 4.14 indicates that the tensile strength of the blend films without crosslinking tended to increase with up to 50 % chitosan content. For the blend films with chitosan content higher than 50 %, the tensile strength became rather constant as shown in Figure 4.14. It can be explained that, in the wet state, silk fibroin film is soft and very weak. The presence of chitosan in the blend films could improve the strength of the blend films because of the rigidity of chitosan chain. The crosslinked blend films exhibited a much

higher tensile strength in the wet state with an increasing content of chitosan. Since the amount of crosslinking agent added was related to chitosan content, the tensile strength of the crosslinked blend films increased in accordance with chitosan content. It can be indicated that, in the dry state, chitosan could improve the strength of pure silk fibroin film by imparting the flexibility to the blend films. Moreover, the tensile strength of the blend films in the wet state was also improved by the presence of chitosan in the blend films. The rigid chains of chitosan and the existence of crosslinks can enhance the strength of the blend films.

4.4.2 Elongation at Break

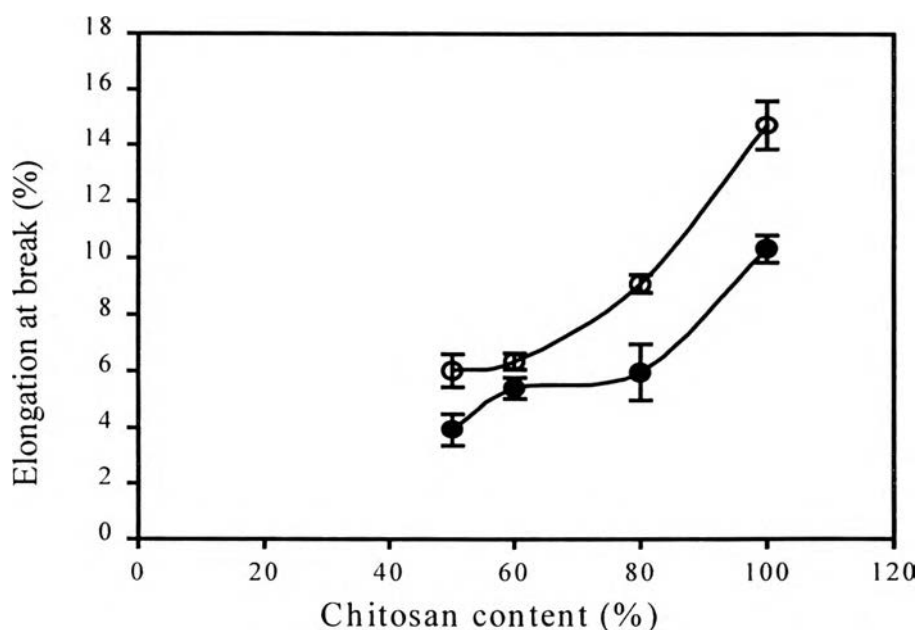


Figure 4.15 Elongation at break of pure and blend films in the dry state. ○ films without crosslinking; ● films with crosslinking

Elongation at break is another mechanical property that is important for determining the application of polymeric films. Figure 4.15 shows the elongation at break of pure and the blend films with and without crosslinking in the dry state. The elongation at break of pure and the blend films depended on the chitosan content. This figure does not show the values of the blend

films with less than 50 % chitosan content because of the brittleness of the films, similar to the case of tensile strength in the dry state. However, when chitosan content was increased, the elongation at break increased. It is found that the presence of at least 50% chitosan content in the blend films can improve the flexibility of the blend films. The reason is that chitosan has a rigid chains which can withstand the tensile force as compared to silk fibroin that is deficient in tensile property. Moreover, this improvement may be derived from hydrogen bonding interaction between chitosan and silk fibroin within the blend films (Chen *et al.*, 1997). Hydrogen bonds act as bridges between polymer chains. When the tensile force is applied, these hydrogen bonds help to relieve the stress concentration between polymer chains slowly leading to higher elongation at break. It was considered that the crosslinked blend films had a lower elongation at break since chitosan chains were held together by crosslinking agent. In addition, the presence of crosslinks limited the extensibility of chitosan chains resulting in decrease of elongation at break.

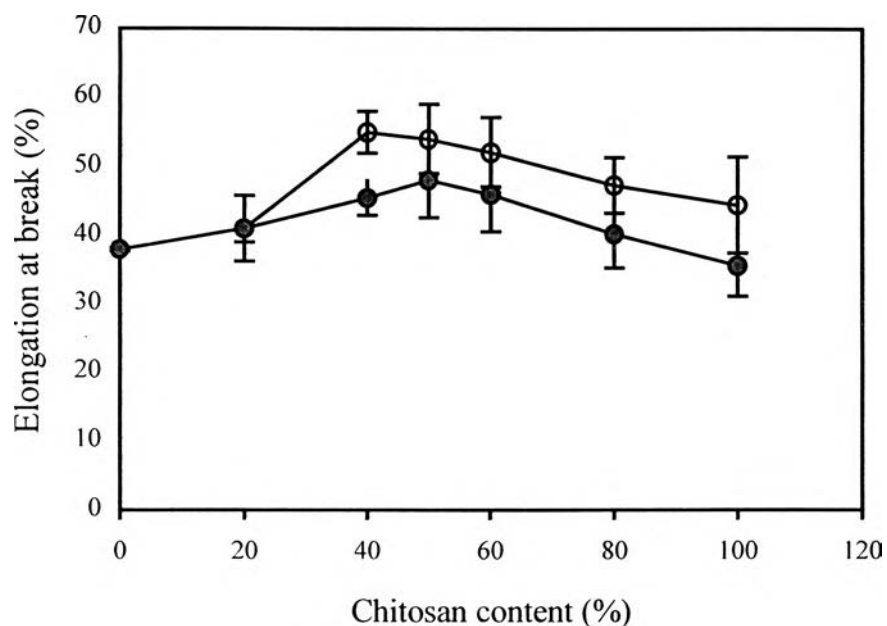


Figure 4.16 Elongation at break of pure and blend films at wet state.

○ films without crosslinking: ● films with crosslinking

The elongation at break in the wet state of pure and the blend films was determined. Figure 4.16 exhibited the elongation at break of pure and the blend films as a function of chitosan content. In the wet state, the elongation at break considerably increased as compared to that of the films in the dry state. At chitosan content between 40-60 %, the blend films had the highest values of elongation at break. When chitosan content is outside this range, the elongation at break of the films dropped. It can be interpreted that in the dry state, the hydrogen bonding between polymer chains is present as confirmed by the FTIR spectra of the films. However, when the films are immersed in water, the water molecules would penetrate inside the films and disrupt the hydrogen bonding between polymer chains. These water molecules have a plasticized effect to the films by allowing the polymer chains segments to move easier (Freddi *et al.*, 1995). When tensile force is

applied, the stress relaxation of polymer chains can occur faster than in the dry state where the chain mobility is restricted. Liang *et al.* (1992), suggested that the ductility of silk fibroin/sodium alginate blend films could be enhanced by the affect of absorbed and free water, which facilitates the chain mobility. Therefore, the blend films in the wet state had higher elongation break than that in the dry state. Similar to the films in the dry state, the crosslinked blend films showed lower elongation at break in the wet state than the blend films without crosslinking. This results from the presence of crosslinks within the films. These crosslinks limit the stretching of the films. Remunan-Lopez and Bodmeier (1997) concluded that the higher the degree of crosslinking, the smaller the water uptake and the elongation of the wet films. Consequently, the crosslinked blend films had lower elongation at break than the blend films without crosslinking.

From these results, in the dry state, the flexibility of silk fibroin film could be enhanced by blending with chitosan. While in the wet state, the elongation at break increased considerably due to the loss of interaction between polymer chains. In addition, the water molecules acted as a plasticizer to facilitate the chain mobility of polymers leading to higher elongation at break.

4.5 Oxygen Permeability

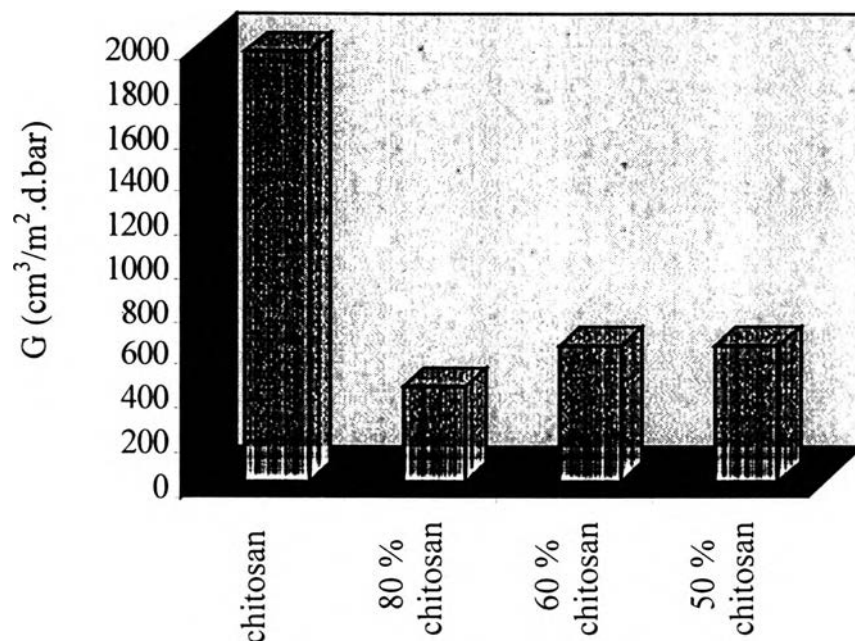


Figure 4.17 The oxygen permeability (G) of pure and the blend films.

Oxygen permeability of pure and the blend films is shown in Figure 4.17. Pure chitosan film showed high oxygen permeability as compared to the blend films. The addition of silk fibroin to the blend films led to the significant decreasing of the oxygen permeability. It may be due to the change in morphology of the blend films. By blending silk fibroin with chitosan, conformation of silk fibroin chains changed from random coil to β -sheet structure, which was the more compact and regular structure (Yashimizu and Asakara, 1990). The more compact structure restricted the oxygen to permeate through the films resulting in the lower oxygen permeability. The oxygen permeability of pure silk fibroin film and the blend films with up to 40 % chitosan content could not be determined due to the brittleness of the films. Hosokawa *et al.* (1992) found that oxygen

permeability of chitosan films with 80 μm in thickness was lower than that of polyethylene but comparable to that of nylon and PET films. In these results, the addition of silk fibroin reduced the oxygen permeability to the blend films.