CHAPTER III
EXPERIMENTAL

3.1 Materials

The HDPE blown film grade, Thai-zex 7000F, was provided by Bangna Plaschem Co.,Ltd. Shells of shrimp (*Penaeus merguiensis*), as a starting material of chitin were kindly supplied by Surapon Foods Public Co., Ltd. and rice starch was kindly supplied by Siam Modified Starch Co.,Ltd. NaOH and HCl for chitin preparation were food reagent grade and used without further purification.

3.2 Equipment

3.2.1 Restch Centrifugal Ball Mill

The chitin was ground into powder by using Restch Centrifugal Ball Mill, type S1 for 2 hours.

3.2.2 Restch Sieving Machine

The chitin powder with the size of 38 to 53 micron was sieved and collected separately by using Restch Sieving Machine, type Vibro.

3.2.3 Bruker FTIR Spectrophotometer

The FTIR spectrum of chitin was recorded with Vector 3.0 Bruker FTIR Spectrophotometer with 16 scans at a resolution of 4 cm^{-1}. A frequency of 4000–400 cm^{-1} was observed using a deuterated triglycinesulfate detector (DTGS) with a specific detectivity of 1×10^9 $cm.Hz^{1/2}.W^{-1}$.

3.2.4 Perkin Elmer Thermogravimetric Analyzer

The degradation temperatures and water contents of chitin and rice starch were measured by Perkin Elmer Thermogravimetric Analyzer TGA7 with Thermal Analysis Controller TAC7/DX. The heating rate was $10^\circ C/min$ from $40^\circ C$ to $700\,^\circ C$ under a N_2 flowing rate of 20 ml/min.

3.2.5   Brabender Plasti-Corder

The chitin-filled and rice starch-filled HDPE blends were prepared in a Brabender Plastic-Corder, PL-2100, Torque Rheometer with a set of removable roller blades and measuring mixer W50 with a volumetric capacity of 55 cm^3 in Heat and Shear stability mode.

3.2.6   Wabash Compression Molder

Sheets of the blends were obtained by compression molding at 170°C in a Wabash V50H Compression Molder. The compression cycle was preheated for 5 minutes, compressed under a force of 10 tons for 3 minutes, and cooled under compression by means of water and air until 40°C was reached.

3.2.7   Instron Universal Testing Machine

Measurements of tensile and flexural properties were performed on an Instron Universal Testing Machine, model 4206.

3.2.8   Zwick Impact Tester

Measurements of Izod impact strength were performed on a Zwick Impact Tester with a 2.7 joules pendulum.

3.2.9   Shore D Durometer

Shore D Durometer was used to observe the Shore D hardness of the blends.

3.2.10   JEOL Scanning Electron Microscope

The microstructures of the blends were observed with a JEOL JSM5200 Scanning Electron Microscope.

3.3   Methodology

3.3.1   Preparation of Chitin

The preparation of chitin is based on the methods of Shimahara and Takigushi, 1988 and Waiprib,1991. The abdominal shells of shrimp were rinsed with water, dried under sunlight and crushed into chips about 0.5 cm^2 in size. A 100 g of dried shell chips was immersed in 1 liter of 1 N hydrochloric acid. The suspension was kept 2 days at room temperature with occasional stirring. The hydrochloric acid was changed everyday. The demineralized shell chips were collected and washed with water until neutral. A 100 g of the demineralized shell chips was immersed in 1 liter of 4% aqueous sodium hydroxide solution. The suspension was kept overnight at room temperature. After changing alkaline solution, the suspension was boiled for 1 hour with stirring. After boiling, the chitin chips were collected, washed with water until neutral and dried at 110°C. Then the chitin chips were ground into powder, sieved and collected separately.

3.3.2   Degree of Deacetylation of Chitin

The method used to determine degree of deacetylation of chitin is based on infrared spectroscopic measurement by Sannan et al., 1978 and Shimahara and Takigushi, 1988. About 3 mg of chitin powder, passed through a 200-mesh sieve, was mechanically blended with 400 mg of potassium bromide powder to prepare a KBr disk. An infrared spectrum was recorded in a range from 4000 cm^{-1} to 1200 cm^{-1}. The absorbances at 2878 cm^{-1} (the C-H band) and 1550 cm^{-1} (the amide II band) were evaluated by the baseline method. The degree of deacetylation (D) was calculated from Equation 3.1.

$$D\ (\%) = 98.03 - 34.68(A_{1550}/A_{2878}) \qquad (3.1)$$

where A_{1550} and A_{2878} are absorbances at 1550 cm^{-1} and 2878 cm^{-1}, respectively.

3.3.3 Density of Chitin and Rice Starch

The densities of fillers, chitin and rice starch, were measured using pycnometer with a volumetric capacity of 25 cm^3. Distilled water with density of 1.00 g/cm^3 and diethyl ether with density of 0.71 g/cm^3 at 25°C were used to be the liquids for determining density of chitin and rice starch, respectively. The density (ρ) was calculated from Equation 3.2.

$$\rho_{filler} \, (g/cm^3) = \frac{wt._{filler}(g)}{25.00 - \dfrac{wt._{liquid}}{\rho_{liquid}}(cm^3)} \tag{3.2}$$

3.3.4 Thermogravimetric Analysis of Chitin and Rice Starch

The degradation temperatures and water contents of fillers, chitin and rice starch, were determined by using Perkin Elmer TGA7. TGA thermogram of sample about 5 mg was recorded from 40 °C to 700°C.

3.3.5 Sample Preparation

The chitin-filled and rice starch-filled HDPE blends were prepared in a Brabender torque rheometer at 30 rpm, 170°C for 10 minutes. The filler contents of the blends were varied at 5, 10, 20, and 30% by weight. The blends were air-cooled and shredded into small pieces before stored in sealed PE bags.

3.3.6 Mechanical Testing

The tensile properties were measured according to ASTM D638 method on an Instron universal testing machine with 100 kN load cell and 50 mm/min crosshead speed. For flexural testing, following ASTM D790,

measurements were done using 5 kN load cell and 12 mm/min crosshead speed. Izod impact strength, according to ASTM D256, was measured on a Zwick impact tester. Hardness testing, according to ASTM D2240, was measured on a Shore D durometer. Five measurements were conducted for each samples and the results were averaged to obtain a mean value.

3.3.7 Microstructure

The fracture surfaces of specimens from impact testing were sputter-coated with gold and examined by JEOL JSM 5200 scanning electron microscope.

3.3.8 Water Absorption

The water absorption of the blends was determined according to ASTM D570. The 76.2 x 25.4 x 3 mm specimens were conditioned by drying in an oven for 24 hr at $50^{\circ}C$. The weights of the specimens were recorded before immersing them into the distilled water maintained a temperature at $23\pm1^{\circ}C$. They were then removed at specified time intervals. Surfaces of the specimens were wiped off with tissue papers and the weights were recorded. The percentage of water absorbed was calculated from Equation 3.3.

$$\text{Water absorption (\%)} = \frac{\text{wet wt. - initial wt.}}{\text{initial wt.}} \times 100 \qquad (3.3)$$

Three specimens of each sample were tested and the results were reported as a mean value.