CHAPTER III
EXPERIMENTAL

3.1 Materials

Poly(lactic acid) (PLA) (Ingeo 4043D) was bought from NatureWorks. Natural rubber (NR, STR 5L) was purchased from Natural Art and Technology Co., Ltd. The rice straw (RS) was obtained from a rice processing facility located in Nakhon Ratchasima Province, Thailand. Dimethylsulphoxide (DMSO) (AR grade), hexane (AR grade), methyl alcohol (AR grade), and chloroform (HPLC grade) were supplied by RCI Labscan Limited.

3.2 Preparation and characterization of PLA blended with masticated NR at different mastication times

3.2.1 Preparation of PLA/NR blends

PLA was dried in an oven at 80 °C overnight to eliminate moisture. Before blending with PLA, NR was masticated at various times (10, 20, and 30 min) with a roll speed of 10 rpm in two roll mill (Yong Fong Machinery, YFY-R-6). PLA/NR blends were mixed in an internal mixer (Haake Rheomix, 3000P) at a temperature of 170 °C with a rotor speed of 60 rpm for 10 min. The ratio of PLA to NR was fixed at 60/40 wt.%. The polymer blends were cut down into smaller pieces using a plastic recycling machine (Tranekaer, DK-5953), and after that, blend films were produced using compression molding (Labtech, LP20-B). The thickness of the film was 0.5 mm.

3.2.2 Melt flow index

In accordance with the ASTM D1238 standard, a melt flow indexer (Kayeness, D40004HV) was used to measure the melt flow index (MFI) of neat PLA and PLA/NR blend. A weight of 2.16 kg was applied at a melt temperature of 190 °C. The average MFI value was determined after cutting and weighing at least 6 extrudates from each

formulation at regular intervals. Equation (3.1) is used to determine the melt flow index.

$$\mathrm{MFI} = \frac{600 \times m}{t} \tag{3.1}$$

where MFI is the melt flow index (g/10 min), m is mass (g), and t is cut time (s) (Djellali, Sadoun, Haddaoui, and Bergeret, 2015).

3.2.3 Tensile properties

Tensile properties of all PLA/NR blend films were evaluated following ASTM D882 (thickness less than 1.0 mm (0.04 in.)) using a universal testing machine (Instron, 5565) with a load cell of 5 kN, a crosshead speed of 12.5 mm/min, and a gauge length of 50 mm. The specimen dimension was 15 mm width and 120 mm length. The average data for at least 10 specimens was reported.

3.3 Preparation and characterization of PLA/NR/RS biocomposite films

3.3.1 Rice straw fiber preparation

Rice straw was dried in an oven at 60 °C overnight and ground using a mechanical crusher (Hight speed multi-function mill) at a rotor speed of 1,500 rpm to obtain 44-250 μm particles. After that, ground rice straw was sieved to get <53 μm particles.

3.3.2 Preparation of PLA/NR/RS biocomposites

PLA/NR/RS biocomposites were prepared in various compositions, as shown in Table 3.1. The PLA/NR/NR blend that showed the best properties was used as a composite matrix. The amount of rice straw in the polymer composite was varied in the range of 3, 5, and 10 wt.% based on the total weight of the PLA/NR blend. All compositions of polymer composites were mixed using an internal mixer (Haake Rheomix, 3000P) at a temperature of 170 °C with a rotor speed of 60 rpm for 10 min. The polymer composite was subsequently crushed into small pieces in a plastic recycling machine (Tranekaer, DK-5953).

Table 3.1 Formulations of PLA/NR/RS biocomposites.

Sample code	PLA (%wt.)	NR (%wt.)	Rice straw (%wt.)
Neat PLA	100.0	-	-
PLA/NR (60/40)	60.0	40.0	-
PLA/NR/3%RS	58.2	38.8	3.0
PLA/NR/5%RS	57.0	38.0	5.0
PLA/NR/10%RS	54.0	36.0	10.0

3.3.3 Biocomposite films preparation

Cast film extrusion (Betol, BC 32) was used to produce neat PLA, PLA/NR blend, and PLA/NR/RS biocomposites films. The barrel (zones 1, 2, 3, 4) temperature was 45, 165, 180, 190 ℃, respectively, and the slit die zone was 190 ℃. The screw speed and chill roll speed were 40 and 6 rpm, respectively.

3.3.4 Characterization of PLA/NR/RS biocomposite films

In this study, the melt flow index (MFI), tensile properties, thermal properties, and morphological properties of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films were observed. The MFI was determined using the same testing method as described in Section 3.2.2. The tensile properties were evaluated using the same testing method as described in Section 3.2.3. The thermal properties were assessed using a differential scanning calorimeter (DSC), while the morphological properties were examined using a field emission scanning electron microscope (FE-SEM).

3.3.4.1 Thermal properties

A differential scanning calorimeter (METTLER TOLEDO, DSC 3+) was used to determine the thermal characteristics of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films. An aluminum pan was used to hold the sample, which weighed between 5 and 10 mg and was sealed with an aluminum cover. To remove the prior thermal history, during the first heating process, the sample increased in temperature from -100 ℃ to 200 ℃ at a rate of 5 ℃/min. Subsequently, it was maintained at a temperature of 200 ℃ for a duration of 2 minutes in a nitrogen atmosphere. The sample was subsequently cooled to a temperature of -100 ℃ at a

rate of 5 °C/min (cooling) and then heated to a temperature of 200 °C at a rate of 5 °C/min for the second heating process. The crystallinity degree (Xc) of the PLA phase in polymer composites was determined by utilizing the enthalpy value of a crystalline PLA, as defined by equation (3.2).

$$X_c(\%) = \frac{(\Delta H_m - \Delta H_c)}{\Delta H_m^{\infty}} \; x \; 100 \qquad (3.2)$$

where ΔH_m is a measured endothermic enthalpy of melting and ΔH_c is the cold crystallization exothermic enthalpy during the heating scans. The theoretical melting enthalpy of 100% crystalline PLA (93.7 J/g) will be taken to be ΔH_m^{∞} (Tang, Zhang, Liu, and Zhu, 2012).

3.3.4.2 Morphological properties

The morphological properties of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films were investigated using a field emission scanning electron microscope (FE-SEM) (Zeiss AURIGA FE-SEM). To identify different phases, the rubber phase of the tensile fractured surfaces was stained with osmium tetroxide for 24 hours and coated with gold. FE-SEM images of the samples were collected using a 3 keV acceleration voltage.

3.4 Biodegradability of neat PLA, PLA/NR blend and the PLA/NR/RS biocomposite films

3.4.1 Soil burial biodegradability

At least 5 samples of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films (30x30 mm) were buried in a box containing soil having a pH of 7 and maintained at 30% moisture by weight. The samples were buried at a depth of 120-150 mm. After being buried for 30, 60, and 90 days, each sample was retrieved from the soil. The obtained specimens were washed with DI water and dried in a vacuum oven at 40 °C to a constant weight. The degradation of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films in soil was assessed using various methods. These included measuring the weight loss percentage, analyzing the chemical components using X-ray analysis and EDX, determining the molecular weight using gel permeation chromatography (GPC), examining the crystalline structure using wide-

angle X-ray scattering (XRD), evaluating the tensile properties, analyzing the thermal properties using differential scanning calorimetry (DSC), and studying the morphology using field-emission scanning electron microscopy (FE-SEM). The testing techniques for the tensile, thermal, and morphological properties were conducted according to the methods specified in Sections 3.2.3, 3.4.4.1, and 3.4.4.2, respectively.

3.4.1.1 Weight loss percentage

The weight loss percentage for the specimen test was determined using equation (3.3).

$$\text{Weight loss } (\%) = \left(\frac{W_i - W_t}{W_i}\right) \times 100 \quad (3.3)$$

where w_i is the samples' initial dry weight and w_t is the dry weight of the samples after being buried in soil for a certain period (Boonmee, Kositanont, and Leejarkpai, 2016).

3.4.1.2 Molecular weight characterization

To extract the PLA phase from the PLA/NR blend and PLA/NR/RS biocomposite films, which were collected both before and after the soil burial test, the film samples were dissolved in dimethyl sulfoxide (DMSO). The solution was transferred into the separatory funnel, and hexene was added. The mixture was shaken to facilitate the separation of the components into two different layers. The DMSO layer was drained out and subsequently precipitated using methanol. After the extracted PLA was obtained, it was dried in a hot air oven at a temperature of 40 °C for a duration of 24 hours. Gel permeation chromatography (GPC, Waters Alliance e2695) was employed to examine the molecular weight and polydispersity index (PDI) of both neat PLA and the PLA extracted from each composite film. Chloroform was used as a solvent and a flow rate of 0.5 ml/min. The volume that was injected was 40 µl, and the concentration of the sample was 0.7% w/v. A calibration curve was generated using polystyrene standards from Waters Corporation, USA.

3.4.1.3 Morphological properties

Morphological properties of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite films were investigated using a FE-SEM (Zeiss AURIGA FE-SEM). To identify different phases, the rubber phase was stained with osmium tetroxide for

a duration of 24 hours. Before performing FE-SEM analysis, a carbon coating on the surfaces of the samples was applied. FE-SEM images of the samples were collected using a 3 keV acceleration voltage.

3.4.1.4 Chemical component characterization

The chemical composition of neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite film samples was analyzed using field emission scanning electron microscopy (FE-SEM, JEOL JSM-7800) with energy dispersive X-ray analysis (EDX) at an acceleration voltage of 3 kV. The analysis was conducted both before and after the samples were buried in soil. The samples were prepared using a similar technique to that utilized for FE-SEM analysis, with the only distinction being the use of a gold coating on the sample surfaces.

3.4.1.5 X-ray diffraction analysis

Neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite film samples were exposed to X-ray diffraction (XRD) investigations. The analyses were conducted using a Bruker D2 PHASER X-ray diffractometer equipped with a CuKα (λ = 1.54 Å) source. The objective was to identify the crystalline phases present in the materials. The samples were scanned in the angular intervals ranging from 5 - 50 degrees.

3.5 An application of biocomposite films as Seedling bags

The neat PLA, PLA/NR blend, and PLA/NR/RS biocomposite film were cut, and two pieces were connected by heat sealing to produce seedling bags sized 10x15 cm. Each seedling bag was punctured to create 12 holes, each about 0.5 cm in diameter, as shown in Figure 3.1. Subsequently, the chili seedlings were transferred into bags filled with soil and placed in a plant pot filled with soil. Watering is carried out every day, while fertilizer takes place once a month. To evaluate the efficacy of plant development in biocomposite bags, various parameters, including the plant's dry weight, stem diameter, height, and total weight of chili fruits per plant, were assessed after a three-month period. The measurements were taken from three chili plants, and the results were averaged. For dry weight evaluation, the plants were washed with

deionized water and then dried in a vacuum oven at 100 °C until a constant weight was achieved. Subsequently, the dry plants were weighed.

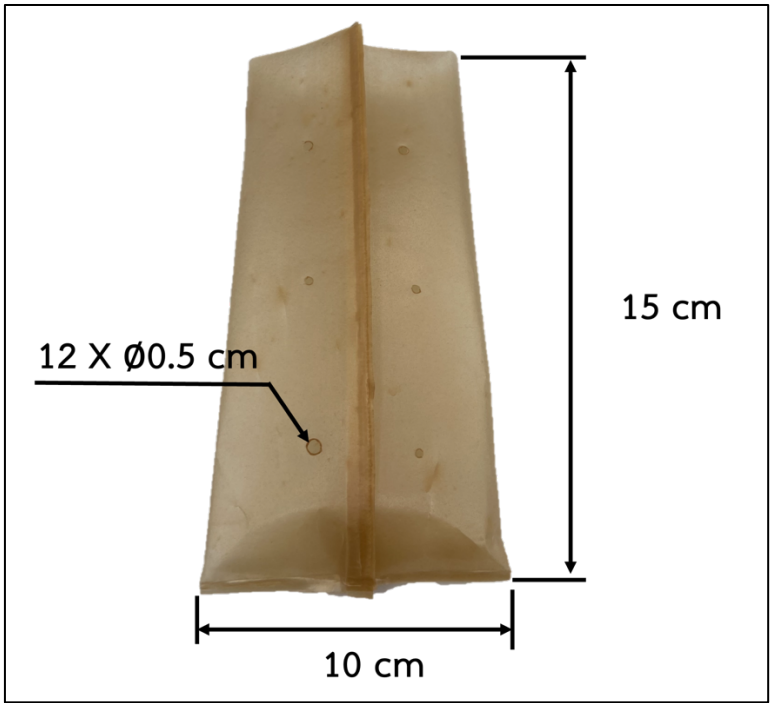

Figure 3.1 Dimensions of seedling bags.

The biodegradability of the seedling bags was assessed after a three-month period of growing chili plants. The evaluation was conducted using various methods, including measuring the percentage of weight loss, characterizing the molecular weight using GPC, analyzing the tensile properties, conducting wide-angle x-ray scattering (XRD), examining the thermal properties using DSC, and assessing the morphological properties using FE-SEM. The testing was performed using the procedures stated above.

3.6 An application of biocomposite films as mulch film

To produce a mulch film, two 4 strips of cast film measuring around 10 cm in width and 100 cm in length were combined using chloroform, resulting in a single film measuring 40x100 cm. Subsequently, the films were punctured to form a pair of holes at midpoints sized 5 cm, 50 cm apart (see Figure 3.2). After that, the film was used as mulch for chili plants. Five replications were conducted for neat PLA, PLA/NR blend, and PLA/NR biocomposite films. To examine the effectiveness of different types of

films used for mulch in plant development, several parameters were measured after a three-month period. This data included the plant's dry weight, stem diameter, height, and total weight of chili fruits per plant. Measurements were obtained from three chili plants and were averaged. The biodegradability of the mulch films was assessed using techniques that include molecular weight characterization (GPC), wide-angle x-ray scattering (XRD), thermal analysis (DSC), and examination of morphological features (FE-SEM). The testing was carried out employing the methods stated above.

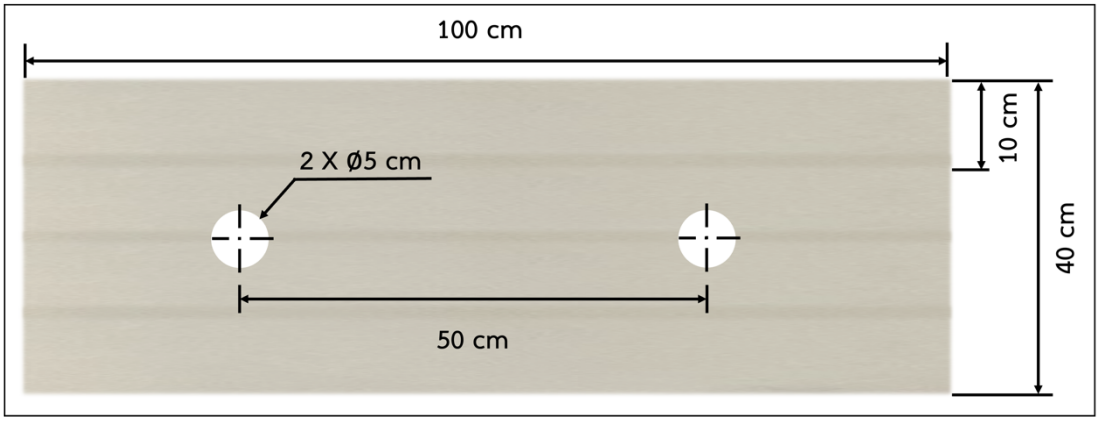

Figure 3.2 Dimensions of mulch films.