Effect of protein concentrations on the properties of fish myofibrillar protein based film compared with PVC film

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Abstract The effect of protein concentrations on the properties of fish myofibrillar protein film (FMP) were investigated and compared with commercial wrap film (polyvinyl chloride; PVC). FMP (2 %, w/v) showed the highest mechanical properties [tensile strength: 4.38 MPa and elongation at break: 133.05 %], and water vapor permeability [$2.81 \times 10^{-10} \text{ g m}^{-1} \text{ s}^{-1} \text{ Pa}^{-1}$]. FMP contained high molecular weight cross-links, resulting in complex film network, as indicated by lower film solubility (19–22 %) and protein solubility (0.6–1.3 %). FMP showed excellent barrier properties to UV light at the wavelength of 200–280 nm. FMP had the thickness [0.007–0.032 mm], color attributes and transparency similar to PVC film [thickness: 0.010 mm]. Therefore, protein concentration majority influenced the properties of develop FMP. The protein content of 1 % (w/v) had potential to be developed the biodegradable film with comparable properties to the commercial wrap film.

Keywords Biodegradable film · Fish myofibrillar protein · Polyvinyl chloride · Water vapor permeability

Introduction

In recent years, there has been an increasing concern of the environmental problem caused by the excessive use of synthetic packaging materials. This is because of their made from non-biodegradable polymeric material, which providing the major source of environmental problem. As a result, considerable effort has been made to develop biodegradable based from biopolymers to produce environmental friendly packaging. Natural biopolymers have been gained increasing attention for manufacturing of biodegradable films, owing to their biocompatibility and non-toxic material that might replace synthetic one. The main biopolymers that used to produce biodegradable films are derived from polysaccharides, proteins and lipids. Among these materials, proteins are the most attractive used because of its ability to form films with satisfactory mechanical and gas barrier properties (Kaewprachu and Rawdkuen 2014) and relative abundance. As one kind of protein, myofibrillar proteins of fish muscle have been used as film-forming material (Shiku et al. 2003). These proteins are insoluble in water, but can be made soluble by adjusting the pH of solution (Iwata et al. 2000). Basically, protein-based films are prepared from solution comprised of the following three main components; protein, plasticizer and solvent. The film formation and the final properties of films are affected by various factors such as the source and concentration of protein, pH of film-forming solution (FFS), plasticizers, the preparation condition and substances incorporated into FFS (Kaewprachu and Rawdkuen 2014; Sobral et al. 2005). According to Wittaya (2012) who reported that the concentration of FFS can influence the self-adhesion of high polymers and the rate of polymer matrix forming in film solution. Cuq et al. (1995) who recommended the optimum concentration of fish myofibrillar protein from Atlantic sardines for prepared film was 2.0 g protein/100 g FFS, while Iwata et al. (2000)

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suggested that blue marlin film was successfully prepared at 3 % protein content. Though the effect of protein concentration on film properties of FMP has been reported, the information concerning the different protein concentrations on the FMP films' properties is scarce and few information in terms of comparison study between commercial wrap film and FMP film have been reported. Therefore, the objective of this investigation was to study the effect of protein concentrations on the properties of fish myofibrillar protein film and compared with polyvinyl chloride film.

Materials and methods

Samples

Fresh tilapia (*Oreochromis niloticus*) (400–500 g/fish) was purchased from a local market in Chiang Rai, Thailand. PVC wrap films of 10 μm thickness (Quick Pack Pacific, Thailand) were used in this study.

Glycerol and other analytical grade reagents were obtained from Merck (Darmstadt, Germany). Electrophoresis reagents were obtained from Bio-Rad Laboratories (Hercules, CA, USA).

Preparation of washed mince

Washed mince was prepared according to the method described in Oujifard et al. (2013). The mince was homogenized with five volumes of 50 mM NaCl at a speed of 11,000 rpm for 2 min. It was then centrifuged at 10,000 g at 4 °C for 10 min. The washing process was repeated twice. Washed mince was freeze-dried, packed under vacuum condition and stored at $-20\text{ }^{\circ}\text{C}$ until utilization.

Preparation of fish myofibrillar protein films (FMP)

FMP film was prepared as described by Oujifard et al. (2013) with a slight modification. Washed mince was mixed with the distilled water to obtain the final protein concentrations of 0.5, 1.0, 1.5 and 2.0 % (w/v). The mixture was homogenized at 11,000 rpm for 1 min, followed by addition glycerol at 25 % (w/w, protein content) and the mixture was stirred gently for 30 min at room temperature. The pH of mixture was adjusted to 3 using 1 N HCl to solubilize the protein by electrostatic repulsion, and finally, it was centrifuged at 3000 g for 10 min at room temperature to remove air bubbles and aggregated proteins. The supernatant was used for film casting. De-aerate FFS ($4 \pm 0.01\text{ g}$) was casted onto a rimmed silicone resin plate (50×50 mm) and then evaporated at room temperature for 24 h before dried with a ventilated oven environmental chamber at $25 \pm 0.5\text{ }^{\circ}\text{C}$ and $50 \pm 5\%$ relative

humidity (RH) for another 24 h. The obtained dry films were manually peeled.

Mechanical properties

Prior to testing the mechanical properties, the films were conditioned for 48 h at $50 \pm 5\%$ RH at $25\text{ }^{\circ}\text{C}$. The tensile strength (TS) [pulling force per film cross-sectional area required to break the film] and elongation at break (EAB) [degree to which film can stretch before breaking] were determined by using a Universal Testing Machine (Lloyd Instrument, Hampshire, UK). Five samples (2×5 cm) with an initial grip length of 3 cm were used for testing. The cross-head speed was set at 30 mm/min with 1 kN load cell use.

Film thickness

The film thickness was measured with a hand-held micrometer (Bial Pipe Gauge, Peacock Co., Tokyo, Japan). Nine random locations around each of the 10 film samples were used for thickness determination.

Film solubility and protein solubility

The film solubility was determined according to the method described in Gennadios et al. (1998). The conditioned films were weighed and placed in a 50 ml centrifuge tube containing 10 ml of distilled water. The mixture was shaken at a speed of 250 rpm with a shaker (Heidolph Inkubator 10,000, Schwabach, Germany) for 24 h. The un-dissolved debris was filtered through a Whatman filter paper No. 1 (Schleicher & Schuell, Maidstone, England). The pellet was dried at $105\text{ }^{\circ}\text{C}$ for 24 h, then weighed. The weight of the solubilized dry matter was calculated by subtracting its difference from the initial weight of the dry matter.

To determine the protein solubility, the protein concentration in the supernatant was determined using the Biuret method (Robinson and Hodgen 1940). Protein solubility was expressed as the percentage of the total protein in the film, which was solubilized with 1 M NaOH for 24 h.

Color, light transmittance and transparency

The color of the film was determined using a Hunter lab color meter (Color QuestXE, Virginia, USA). The color of the film was expressed as L^* (lightness), a^* (redness/greenness) and b^* (yellowness/blueness) values.

The light transmission of the films against ultraviolet (UV) and visible light were measured at select wavelengths between 200 and 800 nm using a UV-Vis spectrophotometer (G105 UV-VIS, Thermo Scientific Inc., USA) according to the method describe in Jongjareonrak et al. (2006).

The transparency value of the film was calculated as follows (Eq. 1) (Han and Floros 1997):

$$\text{Transparency} = -\log T_{600}/x \quad (1)$$

where T_{600} is the fractional transmittance at 600 nm, and x is the film thickness (mm).

Film appearance (visual observation)

The film appearance was determined by using a Fujifilm Finepix S4900 digital camera (Fujifilm Thailand Co. Ltd., Bangkok, Thailand).

Electrophoretic analysis

The protein pattern was analyzed by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) according to the method of Laemmli (1970) using a 10 % running gel and 4 % stacking gel. To solubilize the protein films, the films were mixed with a solubilizing solution (5 % SDS). The mixtures were homogenized at 11,000 rpm for 1 min using IKA homogenizer. The homogenated samples were then stirred continuously for 24 h at room temperature. The supernatant obtained after the centrifugation at 3000 g for 5 min were subjected to SDS-PAGE analysis.

Water vapor permeability (WVP)

The films' WVP was measured by using a modified ASTM method (1989). The films were sealed onto a permeation cup containing silica gel (0 % RH) with silicone vacuum grease and an O-ring to hold the film in place. The cups were then placed in a desiccator saturated with water vapor at 30 °C. The cups were weighed at 1 h intervals over a period of 8 h, and the films' WVP was calculated as follows (Eq. 2) (McHugh et al. 1993):

$$\text{WVP} = wxA^{-1}t^{-1}(\Delta P)^{-1} \quad (2)$$

w is the weight gain of the cup (g); x is the film thickness (m); A is the area of exposed film (m^2); t is the time of gain (s); and (ΔP) is the partial vapor pressure differences between both side of the film (Pa). The WVP was expressed as $\text{g m}^{-1} \text{s}^{-1} \text{Pa}^{-1}$. A total of five samples were determined for each film.

Differential scanning calorimetry (DSC)

Thermal property of the film was carried out using Mettler Toledo Schwerzenbach instrument. The film (2–5 mg) was accurately weighed into aluminium pans, hermetically sealed and scan over the range of 25–300 °C with a heating rate of

5 °C/min in a nitrogen atmosphere (20 ml/min). The empty aluminum pan was used as a reference.

Statistical analysis

Analysis of variance (ANOVA) was performed. The mean comparison was carried out by Duncan's Multiple Range Tests. Significance of difference was defined at $P < 0.05$. The analysis was performed by using an SPSS package (SPSS 16.0 for window, SPSS Inc., Chicago, IL).

Results and discussion

Film thickness

The thickness of FMP film at different protein concentrations in comparison with PVC film are presented in Table 1. The thickness values of FMP film were increased from 0.007 ± 0.0007 mm for the 0.5 % FMP content to 0.032 ± 0.0012 mm for the 2.0 % FMP content, while the thickness of PVC was 0.010 ± 0.0008 mm. The difference in thickness was significant ($P < 0.05$) in all comparison of films made from the different concentrations of FMP. This result indicated that when the protein concentrations increased, film thickness was also increased. The result was consistent with those reported by Chinabhark et al. (2007), Gounga et al. (2007), Kokoszka et al. (2010) and Rocha et al. (2013) who noted a thickness value of surimi film (1–2 % w/v) of 0.018 to 0.031 mm, whey protein isolate film (5, 7 and 9 % w/v) of 0.0166 to 0.0447 mm, soy protein film (6–9 % w/w) of 52.6 to 83.6 μm and argentine anchovy protein isolate film (3.0, 3.5 and 4 % w/w) of 0.113 to 0.176 mm, respectively. Film thickness generally affects the properties of films such as mechanical properties, water vapor permeability, light transmission, and film transparency. Therefore, controlling of this parameter is crucial for the properties of films. According García and Sobral (2005), the formation of films that contained with

Table 1 Thickness, tensile strength and elongation of FMP films as a function of protein concentration in comparison with PVC film

% FMP	Thickness (mm) ($n = 10$)	Tensile strength (MPa)*	Elongation (%)*
0.5	0.007 ± 0.0007^c	3.15 ± 0.10^b	50.38 ± 7.05^d
1.0	0.013 ± 0.0014^c	3.52 ± 0.35^b	116.53 ± 8.40^c
1.5	0.024 ± 0.0033^b	3.81 ± 0.25^b	121.85 ± 7.87^c
2.0	0.032 ± 0.0012^a	4.38 ± 0.28^b	133.05 ± 9.99^b
PVC	0.010 ± 0.0008^d	46.92 ± 2.16^a	268.31 ± 7.27^a

Different superscripts in each column are significantly different ($P < 0.05$)

* Values are mean $\pm$ SD of 5 independent determinations

higher protein concentrations, promoting an increase the solids in the polymer matrix formed after drying the FFS, resulting in increased the thickness of the films.

Mechanical properties

The mechanical properties of FMP film at different protein concentrations in comparison with PVC film were expressed in terms of tensile strength (TS) and elongation at break (EAB) (Table 1). TS and EAB of FMP film increased with increasing protein concentration from 0.5 to 2 % (w/v), but no significantly difference ($P > 0.05$) was observed in TS. Actually, TS would be increased with increasing protein concentration, since an increase in the number of protein chains per surface unit generally leads to an increase in the number of potential intermolecular interactions (Cuq et al. 1996). EAB of FMP film was increased from 50.38–133.05 %, this indicated that the higher protein content in FFS of FMP might result in a higher aggregation intermolecular of protein, compared with the lower amount, resulting in improve flexibility. The result was in accordance with those reported on sarcoplasmic protein film from Blue Marlin (Iwata et al. 2000), fish skin gelatin film (Jongjareonrak et al. 2006) and gelatin film from beef, pork and fish (Nur Hanani et al. 2012), in which the TS and EAB increased with increasing protein concentrations. As compared to PVC film, it showed greater TS over FMP films by about 11 to 15-times. For EAB, FMP films were still less than PVC by around 50 to 80 %. Therefore, the improvement method should be require for develop FMP films' properties to comparable with PVC. However, the films require high or low TS and EAB values depend on the application of the films, for example, film must tolerate normal stress during transport, handling to maintain its integrity or film must high extensibility for food wrap application.

Light transmission and transparency

Light transmission in UV (200–280 nm) and visible ranges (350–800 nm), as well as the transparency of all films, is shown in Table 2. FMP film at various concentrations had

the excellent barrier for light transmission in UV range, while PVC film showed the poorer barrier property. The light transmission in the UV range was from 0.02–84.68 %, while the transmission in the visible range was from 83.28–91.85 %. The highest transmission in all selected wavelengths was found in the PVC film, while the lowest was found in film containing protein content of 2 % (w/v). The result suggested that FMP films could prevent UV transmission, whilst the PVC film analyzed in this study would not. The light transmission in the UV range slightly decreased with increasing protein concentration from 0.5 to 2.0 % (w/v). Hamaguchi et al. (2007) reported that protein-based film exhibited good UV barrier properties, owing to their high content of aromatic amino acids that absorb the UV light. Aromatic amino acids (tyrosine and phenylalanine) are well known to be sensitive chromophores in absorbing light at a wavelength below 300 nm (Li et al. 2004). Therefore, FMP films could retard lipid oxidation induced by UV light in a food system. The film prepared of 2 % (w/v) protein content, which had a greater thickness, showed the lower light transmission in UV ranges. The thicker film would prevent the UV light more effectively than the thinner film. The result was in accordance with Prodpran and Benjakul (2005) who reported that the threadfin bream surimi film (1–2 % w/v) made from the higher protein content exhibited the slightly lower light transmission. Film containing higher protein had the higher thickness, in which light could not be transmitted effectively.

The transparency of FMP film decreased with increasing protein concentration (Table 2). The film was less transparent when the lower protein concentration was used as indicated by the higher transparency value ($P < 0.05$). The film containing 2 % (w/v) protein content had a transparency of 3.44 indicating that the film was more transparent than that film made from surimi film (Chinabhark et al. 2007), blue shark gelatin film (Limpisophon et al. 2009), and PVC film (in this studied).

Color value

Color characteristic of films is an important factor in consumer acceptance of such films in packaging applications. The

Table 2 Light transmission and transparency of FMP films as a function of protein concentration in comparison with PVC film

% FMP	Wavelength (nm)								Transparency*
	200	280	350	400	500	600	700	800	
0.5	0.07	38.46	86.83	87.50	87.64	88.69	88.93	89.14	4.08 ± 0.0008 ^a
1.0	0.04	16.34	85.25	86.71	87.51	88.42	88.98	89.17	3.84 ± 0.0003 ^c
1.5	0.05	7.06	84.11	86.24	87.44	88.02	88.23	89.06	3.56 ± 0.0001 ^d
2.0	0.02	2.98	83.28	85.39	86.97	87.85	88.08	89.44	3.44 ± 0.0005 ^e
PVC	12.06	84.68	88.04	89.40	90.35	91.29	91.61	91.85	3.95 ± 0.0002 ^b

Different superscripts in each column are significantly different ($P < 0.05$)

* Values are mean ± SD of 3 independent determinations

lightness (L^*), redness/greenness (a^*) and yellowness/blueness (b^*) values of FMP film with different protein concentrations in comparison with PVC film are shown in Table 3. Results showed that the color of FMP films was affected by protein concentrations. All films had significantly difference in L^* , a^* and b^* values ($P < 0.05$). The greater b^* value with lower L^* value were noticeable with films having protein concentration of 2 % (w/v), compared with those containing the lower protein concentration (0.5–1.5 %, w/v). Thus, the films with protein concentration of 2 % (w/v) had more yellowness but lower lightness than those FMP film. The result was in accordance with those reported on surimi film (Chinabark et al. 2007) and gelatin film (Nur Hanani et al. 2012), in which L^* value decreased, while b^* value increased with increasing protein concentration. Chinabark et al. (2007) also reported that film prepared at acidic condition might induce the formation of yellowish pigment, especially via Maillard reaction, resulting in greater b^* value but lower L^* value. When compared to PVC film, films having protein concentration of 1 % (w/v) had a^* and b^* values closer to PVC film, while L^* value showed lower than PVC film.

Film and protein solubility

Solubility of FMP film with different protein concentrations in terms of water and protein solubility is shown in Table 4. From visual observations, FMP films maintained their integrity after 24 h immersion in water, suggesting that the protein network is quite stable. Film solubility and protein solubility of the FMP films showed in range of 19–22 % and 0.6–1.3 %, respectively, while PVC film showed insoluble in water. Solubility (water and protein) of FMP film decreased as the protein concentration increased from 0.5 to 2.0 % (w/v). However, the solubility of FMP films in terms of water was not influenced by protein concentration ($P > 0.05$). This results suggested that FMP contains high molecular weight proteins (Fig. 3) are generally insoluble or only slightly soluble in water and thus have potential for forming water-resistant films (Cuq et al. 1998). The lower film and protein solubility were

Table 3 The color values of FMP films as a function of protein concentration in comparison with PVC film

% FMP	Color values [*]		
	L^*	a^*	b^*
0.5	91.82 ± 0.04 ^b	−1.10 ± 0.01 ^a	0.48 ± 0.02 ^{ab}
1.0	91.79 ± 0.02 ^b	−1.17 ± 0.01 ^{cd}	0.44 ± 0.02 ^{bc}
1.5	90.98 ± 0.02 ^c	−1.14 ± 0.01 ^b	0.41 ± 0.01 ^c
2.0	90.53 ± 0.02 ^d	−1.15 ± 0.01 ^{bc}	0.52 ± 0.01 ^a
PVC	92.55 ± 0.14 ^a	−1.18 ± 0.03 ^d	0.41 ± 0.07 ^c

Different superscripts in each column are significantly different ($P < 0.05$)

^{*} Values are mean ± SD of 5 independent determinations

Table 4 Film solubility and protein solubility of FMP films as a function of protein concentration in comparison with PVC film

% FMP	Film solubility (%) [*]	Protein solubility (%)
0.5	21.82 ± 1.10 ^a	1.27 ± 0.25 ^a
1.0	20.30 ± 4.39 ^a	0.97 ± 0.08 ^b
1.5	19.68 ± 2.15 ^a	0.85 ± 0.10 ^{bc}
2.0	18.98 ± 1.27 ^a	0.60 ± 0.09 ^c
PVC	ND ^{**}	ND

Different superscripts in each column are significantly different ($P < 0.05$)

^{*} Values are mean ± SD of 3 independent determinations

^{**} ND, not detected

observed in FMP at the concentration of 2 % (w/v). This result suggested that protein concentration in FFS increased, the polypeptide in FMP more aggregation, leading to more cross-linking and larger molecular weight. This was associated with lower solubility. According to Shiku et al. (2003) who reported that the main associative forces involved in FMP film may be intermolecular covalent bonds with hydrophobic and hydrogen interactions, these induced strong protein-protein interactions in FMP film, resulting in the lowered solubility of film. Water solubility is an important property of the films. Films with high solubility have potential for develop packaging materials intended for easy solubility. However, potential application may require water insolubility to enhance product integrity and water resistance.

Film appearance

The appearance of FMP film with various protein concentrations were similar to PVC (Fig. 1). All of films were flexible, homogeneous and their surfaces appeared smooth, without visible pores or cracks. When the film sheet covered the white background, the color of the background remained clearly observable. These results were similar to the light transmission and transparency of the films. Based on the film appearance, the applications of protein-based films were interesting, especially in food products. When the consumers see the product packed inside the package, it is easy to decide for buying that product.

Water vapor permeability

The WVP of FMP film at different protein concentrations in comparison with PVC film are shown in Fig. 2. The differences in WVP were observed among films prepared from different protein concentrations. The WVP of FMP film showed in range of $0.80\text{--}2.81 \times 10^{-10} \text{ g m}^{-1} \text{ s}^{-1} \text{ Pa}^{-1}$, while WVP of PVC film was $0.25 \times 10^{-10} \text{ g m}^{-1} \text{ s}^{-1} \text{ Pa}^{-1}$. WVP of FMP film increased with increasing protein content in the FFS

Fig. 1 Appearance of FMP films as a function of protein concentration in comparison with commercial wrap film (PVC). Numbers represent protein concentration (% w/v) a: 0.5; b: 1.0; c: 1.5; d: 2.0 % (w/v) and e: PVC film

($P < 0.05$). The result was in agreement with Zavareze et al. (2014) who reported that film produced with a high concentration of muscle myofibrillar protein (Whitemouth croaker) (5 %) had higher WVP values compared to lower protein concentrations (3 and 4 %). Furthermore, FMP film with greater protein content had the higher thickness, resulting in higher WVP of film. Thus, WVP of FMP film could be related to film thickness (Table 1). The same behavior were observed by Kokoszka et al. (2010) and Chang and Nickerson (2015). The film with higher protein content was most likely hygroscopic, compared with that containing the lower protein content. However, as compared to commercial wrap film, FMP film exhibited poor moisture barrier properties by around 3 to 11-times. In addition, WVP of FMP film could be improved to close to commercial wrap film by blend with other hydrocolloids or lipids. Tanaka et al. (2001) found that fish water soluble proteins/lipid blend films showed lower WVP than for the fish water soluble proteins one.

Fig. 2 Water vapor permeability of FMP films as a function of protein concentration in comparison with PVC film. The results represent the means of each film in 3 replications. Bars represent the standard deviation ($n = 3$). Different letters indicate the significantly different ($P < 0.05$)

Protein pattern

Protein patterns of film FFS and FMP film are shown in Fig. 3. Myosin heavy chain (MHC), actin, tropomyosin and troponin were observed in the FMP as the dominant protein. Protein with a MW of 200, 42, 33–35 and 18–30 kDa were likely MHC, actin, tropomyosin and troponin, respectively (Gennadios 2002). Protein patterns of FFS and FMP film with different protein concentration was not observed difference. This result indicated that protein aggregation was bond by weak bonding such as hydrogen bond. Therefore, hydrogen bond between proteins was disrupted when the film was diluted in SDS solution. Chinabhark et al. (2007) and Le et al. (2015) also observed that no differences in protein patterns of bigeye snapper surimi films and gelatin films, respectively, with different protein contents. Protein patterns between the FFS and films analyzed under reducing and non-reducing conditions were no marked differences. However, protein pattern of the film showed the lower band intensity of MHC, compared to FFS. This might be associated with the degradation during film casting and drying. The result was in agreement with Cuq et al. (1995) who found the degradation of MHC in sardine myofibrillar proteins film, especially in the acidic condition, due to the cathepsins. Prodpran and Benjukul (2005) and Chinabhark et al. (2007) who also found the degradation of MHC in threadfin bream surimi film and bigeye snapper surimi film, respectively.

Differential scanning calorimetry

DSC thermogram of FMP film with different protein concentrations and PVC film are shown in Fig. 4. The denaturation temperature (T_p) was observed to be maximum for

Fig. 3 Protein pattern of film-forming solution (a) and films (b) from FMP as a function of protein concentration under reducing and non-reducing condition. M: Protein markers; MHC: Myosin heavy chain. The numbers designate protein concentrations (% w/v)

FMP film with protein content of 0.5, 1.0, 1.5 and 2.0 % (w/v) were 72.34, 78.66, 78.83 and 80.33 °C, respectively. This endothermic peak has been associated with the disruption of the protein interaction formed during film preparation

Fig. 4 DSC thermograms of FMP films as a function of protein concentration in comparison with PVC film. The numbers designate protein concentrations (% w/v)

(Jongjareonrak et al. 2006). Furthermore, FMP film showed the second endothermic peak around 219.46 to 235.27 °C, this was mostly associated with the degradation of the larger size or associate with protein fraction. T_{dm} was related to the heat stability of protein network structure. The increase in thermal stability of films was possibly associated with increased the interaction of proteins such as hydrogen bonds, ionic-interaction, hydrophobic-hydrophobic interactions, which stabilized the network structure of film (Barreto et al. 2003). Therefore, film containing higher protein content required the higher enthalpy for destroying the inter-chain interactions. The high enthalpy observed in the film containing a greater concentration of protein was coincidental with the high T_{dm} . Thus, thermal properties of FMP film were markedly affected by the level of protein used. In PVC film, it can be seen a relatively small glass transition temperature peak (T_g) at around 69.56 °C and they also showed endothermic peak at around 289.31 °C, this peak might associated with the degradation of PVC film.

Conclusion

The properties of the protein based films from FMP were affected by protein content. FMP film exhibited homogeneous and smooth surface, similar to PVC film. The thickness, tensile strength, elongation at break, water vapor permeability and b^* value of FMP film increased, while the solubility (water and protein) and L^* values were decreased with increasing protein concentration. However, as compared to the PVC, FMP film had relatively poor mechanical and barrier properties. Therefore, the FMP film should be improved the properties comparable to commercial wrap film through optimization of other parameters (such as amount and type of plasticizers or cross-linking agents). FMP at 1 % (w/v) had the potential to develop the biodegradable film with comparable properties to PVC film.

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Compliance with ethical standards

Competing interests There are non-financial competing interests (political, personal, religious, ideological, academic, intellectual, commercial or any other) in relation to this manuscript to declare.

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