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Storage stability of soy protein isolate films incorporated with mango kernel extract at different temperature

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ABSTRACT

This research investigated the storage stability of antioxidant films made from waste and by-products which are soy protein isolate (SPI) and mango kernel extract (MKE) stored at room temperature (25 °C), refrigeration temperature (4 °C) and frozen temperature (−18 °C) for 90 days. The thickness of the films was maintained from 0.050 to 0.058 mm until the 90th day. The colour properties of SPI films incorporated with MKE (SPI + MKE) were generally not significantly affected by time and temperature except for the *b* value. All the films turned darker over the storage time. There was no dominant factor between temperature and time for the mechanical properties; all the films showed an increase in tensile strength and Young's modulus, and a decrease in elongation. The antioxidant activity of the films was determined by the total phenolic content and radical scavenging activity of DPPH and ABTS. SPI + MKE film at 25 °C showed the highest antioxidant activity as compared to films stored at 4 °C and −18 °C in all the analyses, with the result being significant in DPPH and ABTS analyses. The film stored at 25 °C showed 26 to 50% higher ($p > 0.05$) TPC than films stored at 4 °C and −18 °C, respectively and had the highest antioxidant activity (54%) in ABTS analysis ($p < 0.05$). SPI + MKE film stored at 25 °C also showed only 1% depreciation of radical scavenging activity (RSA) throughout the storage time. The highest decrease (4%) in antioxidant activity was recorded for SPI + MKE film stored at −18 °C, although it was considered very low. This shows that the antioxidant activity of the films is stable for 90 days of storage.

1. Introduction

Manufacturing of conventional food packaging materials may involve the addition of some chemicals such as phthalate esters, alkylphenols, 2,2-bis(4-hydroxyphenyl)propane, (bisphenol A or BPA) and di(2-ethylhexyl) adipate (Fasano, Bono-Blay, Cirillo, Montuori, & Lacorte, 2012). These chemicals may leach from the packaging into food products and due to their toxicity, the long-term exposure to human will raise safety concerns (Weng & Zheng, 2015). Thus, research on the new packaging materials and systems considered as safe are gaining interest today. These include the development of active and smart packaging based on biodegradable and green materials.

Active packaging involves the incorporation of active compounds, such as antioxidant and antimicrobial agents that can interact with the head-space of packaging by absorption or release of the compounds (Bastante, Cardoso, Serrano, & de la Ossa, 2017). Antioxidant film is one of the most popular types of active packaging. Antioxidants incorporated into the polymer matrix do not interfere with the sensorial properties of food. Antioxidant films also provide a controlled and gradual release of the antioxidant, thus providing longer protection to the packaged product (Riquelme, Herrera, & Matiacevich, 2017).

Antioxidant films can be developed by casting method and, generally, the content is made up of biopolymer, antioxidant and plasticizer. Polysaccharides, proteins and lipids are mostly used as the source of biopolymer (Liu, Meng, Liu, Kan, & Jin, 2017). Protein films are superior in terms of the gas barrier and mechanical properties at low moisture content as compared to polysaccharides and lipids, therefore they continue to be the preferred material for film production (Alves, Gonçalves, & Rocha, 2017; Azeredo & Waldron, 2016; Ou, Kwok, & Kang, 2004).

Soy protein isolate (SPI) is made by defatted soybean which is the by-product of soybean oil industry (Maryam Adilah, Jamilah, & Nur Hanani, 2018). It contains a minimum of 90% protein and it is normally used in agriculture, adhesives and biotechnology. SPI is biodegradable, renewable and has a good film-forming ability (Wang, Kang, Zhang, Zhang, & Li, 2017).

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The green consumerism has prompted the usage of natural antioxidants over synthetic ones. Some of the research has used natural antioxidants such as clove essential oil (Ortiz, Salgado, Dufresne, & Mauri, 2018), licorice residue extract (Han, Yu, & Wang, 2018), pine-needle extract (Yu et al., 2018) and also mango kernel extract which was used in the previous research (Maryam Adilah et al., 2018). Mango kernel is the waste product of the mango industry that is currently not yet being fully utilized. Although mango kernel is the waste product, it contains high active compounds such as phenolic acids, flavonoids, gallotannins and ellagitannins (Torres-León, Rojas, Serna-Cock, Belmares-Cerda, & Aguilar, 2017; Torres-León et al., 2016) that contribute to its high antioxidant activity. According to the Japanese regulation, the usage of mango kernel in food is permissible (Saito, Kohno, Yoshizaki, & Niwano, 2008), thus, it is safe for consumption. The usage of mango kernel extract (MKE) as a source of natural antioxidants will provide a new alternative to fully utilize this waste product.

Inevitably, packaging material should have the ability to protect the packaged product throughout its lifetime. The storage stability of the film either in terms of physical or functional properties should be maintained at a reasonable level to ensure the safety of the packaged product. However, aging process is inevitable to biopolymer due to physical changes (recrystallization process and migration of compounds such as water and glycerol) and chemical changes (oxidation of the protein sulfhydryl groups) (Anker, Stading, & Hermansson, 2001; Osés, Fernández-Pan, Mendoza, & Maté, 2009). The aging process also differs according to the temperature and time.

Therefore, the objective of this study was to determine the stability of antioxidant films made from SPI and MKE stored at room, refrigeration and freezing temperatures (25 °C, 4 °C and −18 °C, respectively) for 90 days.

2. Materials and methods

2.1. Chemicals

Soy protein isolate (SPI) with 92% protein content was purchased from MP Biomedicals (Solon, Ohio, USA). ABTS and Folin Ciocalteu reagent were supplied by Merck and Co. (Darmstadt, Germany). Potassium persulphate, sodium carbonate and gallic acid were purchased from R&M Chemicals (Selangor, Malaysia) whereas anhydrous glycerol (99.5% purity) was supplied by Systerm (Karlsruhe, Germany). DPPH was purchased from Tokyo Chemical Industry (Tokyo, Japan). Absolute ethanol (99.9% purity) and soy lecithin were purchased from John Kollin Corporation (Midlothian, UK) and Modernist Pantry (Eliot, Maine USA), respectively.

2.2. Extraction of mango kernel

Mango seeds from Chokanan variety (maturity stage 5; fully ripened) were collected from mango juice producer in Serdang, Selangor. The mango kernel was extracted according to Maryam Adilah et al. (2018). The mango kernels were separated from the seeds and cleaned. The kernels were diced and dried at 50 °C for 24 h (Abdalla, Darwish, Ayad, & El-Hamahmy, 2007; Augustin & Ling, 1987). The dried kernel was ground using a blender (Tefal, Rumilly, France). Ethanol was added in 5:1 (v/w) ratio. The mixture was left in dark for 24 h with regular shaking. The residue was removed by using Whatmann no. 4 filter paper and the supernatant was evaporated at 40 °C. The MKE was used for the film preparation.

2.3. Film preparation

The film was prepared according to Maryam Adilah et al. (2018) and Tongnuanchan, Benjakul, and Prodpran (2013). Distilled water was heated to 70 °C and 3.5% SPI was added and mixed for 30 min. Glycerol as a plasticizer (30% w/w based on SPI content), MKE (1, 3 and 5% w/w based on SPI content) and soy lecithin as an emulsifier (25% w/w based on MKE content) were added and stirred at 50 °C for other 30 min. The mixture was then homogenized at 5000 rpm for 3 min using a homogenizer (Heidolph Instruments GmbH & Co., Schwabach, Germany). Then, 14 ml of the film solution was spread on polystyrene petri dish plate ($14 \times 14\ cm^2$) and dried at 25 °C and 50% relative humidity (RH) for 24 h. The film solution without the addition of MKE was used to prepare the control film sample.

2.4. Storage of films

The films were stored at three different temperature which are the ambient temperature (25 °C), refrigerating temperature (4 °C) and freezing temperature (−18 °C) at 40–60% RH. These temperatures were chosen as most of the packaged food in the industry are stored at these conditions. The films were stored for 90 days and analyses were conducted for every 10 days interval.

2.5. Film thickness

The film thickness was determined with a digital micrometer (Mitutoyo Absolute, Tester Sangyo Co. Ltd., Japan). The thickness was measured in ten randomly selected locations on each film and then an average value was calculated.

2.6. Colour

The colour of the films was measured using a MiniScan XE Plus Hunter colourimeter (Hunter Associates Laboratory, Inc., Reston, Virginia). The *L*, *a* and *b* values were determined to indicate white/black, red/green and yellow/blue, respectively. The machine was calibrated using a standard white tile.

2.7. Mechanical properties

The mechanical properties were measured according to the method by Maryam Adilah et al. (2018) as adapted from Ili Balqis, Nor Khaizura, Russly, and Nur Hanani (2017). The mechanical properties were expressed in terms of tensile strength, elongation at break and Young's modulus and were determined using INSTRON 4302 Series IX Machine (Instron Co., Canton, Massachusetts, USA). The films were cut into rectangular strips ($1.5 \times 9\ cm^2$) and were conditioned at 23 ± 2 °C and 50 ± 5% RH for 2 days. The film strip was stretched between the grips with 50 mm initial separation and 50 mm/min cross head speed. The tensile load used was 5 kN.

2.8. Total phenolic content (TPC)

The total phenolic content was determined according to previous research by Maryam Adilah et al. (2018) using the method as adapted from Ruiz-Navajas, Viuda-Martos, Sendra, Perez-Alvarez, and Fernández-López (2013). Twenty-five mg of film sample was immersed in 3 ml ethanol to get the extract. After that, 0.3 ml of the extract was added to 2.5 ml Folin Ciocalteu reagent (10% v/v) followed by 2 ml 7.5% (w/v) sodium carbonate solution. Next, it was kept at 50 °C for 5 min. The absorption was measured at 760 nm using Genesys 10 UV–Vis spectrophotometer (Thermo Fisher Scientific, Madison, Wisconsin, USA). Gallic acid solutions (0–1000 ppm) were used to obtain the standard curve. The result was expressed as microgram gallic acid equivalent per gram film (μg GAE/g film).

2.9. DPPH radical scavenging assay

DPPH free radical scavenging assay was carried out according to previous researches (Maryam Adilah et al., 2018; Siripatrawan & Harte, 2010). Twenty-five mg of film sample was immersed in 3 ml ethanol to