Food Hydrocolloids 55 (2016) 136–143

Contents lists available at ScienceDirect

Food Hydrocolloids

journal homepage: www.elsevier.com/locate/foodhyd

Physical properties and application of a red pepper seed meal protein composite film containing oregano oil

Ji-Hyeon Lee, Hyun-Ju Yang, Ka-Yeon Lee, Kyung Bin Song*

Department of Food Science and Technology, Chungnam National University, Daejeon 305-764, Republic of Korea

ARTICLE INFO

Article history:
Received 30 June 2015
Received in revised form
28 October 2015
Accepted 10 November 2015
Available online 14 November 2015

Keywords:
Composite film
Oregano oil
Physical property
Red pepper seed meal

ABSTRACT

Red pepper seed meal protein (RMP) was extracted from the by-product of red pepper seed oil production. RMP composite films were prepared, and their physical properties were evaluated. The optimal concentration for the RMP composite film was suggested to be 4% RMP/1% gelatin, and the tensile strength (TS) and elongation at break (E) values of the RMP composite film were 20.14 MPa and 11.45%, respectively. In addition, RMP composite films containing oregano oil (OR) were prepared by incorporating different amounts (0, 0.1, 0.3, 0.5, 0.7, and 1%) of OR. OR affected the physical properties of the RMP composite film, and the composite film containing 0.5% OR had optimal physical properties: a TS of 8.71 MPa, a Young's modulus of 142.83 MPa, and an E of 75.32%. The antimicrobial and antioxidant activities of the RMP composite films with OR increased as the OR content increased. Wrapping fatty tuna meat with the RMP composite film containing 0.5% OR reduced the inoculated *Listeria monocytogenes* and *Salmonella* Typhimurium populations by 0.73 and 0.89 log CFU/mL, respectively, compared with the control sample after storage at 4 °C for 12 days. Moreover, the RMP composite film containing 0.5% OR decreased the degree of lipid oxidation in the fatty tuna meat during storage. Consequently, a RMP composite film containing OR can be useful in prolonging the shelf life of fatty tuna meat.

1. Introduction

Environmental concerns over non-biodegradable plastic packaging materials have increased research interest in biodegradable film sources (Rhim, Hong, & Ha, 2009). Biodegradable films can be prepared from proteins, polysaccharides, and lipids (Lee & Song, 2015); in particular, protein-based films have a good film-forming ability (Cao, Fu, & He, 2007). However, they have some limitations, such as poor thermal stability, poor physical properties, and high cost (Song, Song, Jo, & Song, 2013). Therefore, studies on inexpensive protein materials as well as suitable plasticizers and composite materials for the preparation of protein films are needed.

Red pepper seed meal (RM) is produced from red pepper oil extraction. Firatligil-Durmus and Evranuz (2010) reported that red pepper seeds are good source of protein (26%), but until now, have been mostly used for animal feed only. Therefore, food-processing by-products, such as RM, can be used as a film base material for protein films (Jo, Song, Lee, & Song, 2014; Shin, Jang, & Song, 2011; Song, Jo, et al., 2013; Song, Song, et al., 2013). In our preliminary experiment, plain red pepper seed meal protein (RMP) film was too weak and brittle, and a composite film was suggested. In general, the purpose of a composite film is to enhance the physical properties of protein films, and gelatin and chitosan are usually used as a composite material (Lee, Song, Jo, & Song, 2014; Shin et al., 2011; Song, Lee, Al Mijan, & Song, 2014). In particular, gelatin is incorporated into the protein films due to its excellent film-forming property and biodegradability (Cao et al., 2007).

Essential oils can be incorporated into the films because they have phenolic compounds with antimicrobial and antioxidant activities (Burt, 2004). An essential oil, oregano oil (OR), is extracted from *Origanum vulgare* L. and has primary active constituents, including carvacrol, thymol, c-therpinene, and p-cymene (Benavides, Villalobos-Carvajal, & Reyes, 2012).

Previously, protein-based films have been prepared using the proteins obtained from plant seed oil extraction processes (Lee et al., 2014; Song, Jo, et al., 2013; Song, Song, et al., 2013). However, red pepper seed meal protein (RMP) composite films have never been studied before. Therefore, this study aims to prepare RMP composite films and to apply the RMP composite film containing

* Corresponding author.
E-mail address: kbsong@cnu.ac.kr (K.B. Song).

http://dx.doi.org/10.1016/j.foodhyd.2015.11.013
0268-005X/

OR to the packaging of fatty tuna meat.

2. Materials and methods

2.1. Materials

RM was obtained from a local market (Boeun, Korea). Ammonium sulfate, gelatin, and plasticizers (glycerol, sorbitol, fructose, and sucrose) were purchased from Sigma–Aldrich Chemical Co. (St. Louis, MO, USA). OR was obtained from The Certification Academy for Holistic Aromatherapy (Seoul, Korea).

2.2. Extraction of RMP

RMP was extracted according to the method of Firatligil-Durmus and Evranuz (2010), with modifications. RM was ground in a blender and mixed with 10 volumes of 0.1 N NaOH (v/w) using a stirrer at 70 °C for 2 h. The supernatant was obtained by centrifugation at 10,000× g at 20 °C for 20 min and then precipitated using 70% ammonium sulfate. After centrifugation, the precipitated protein was desalinated and lyophilized.

2.3. Sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE)

SDS-PAGE was performed according to the method of Laemmli (1970). The samples were loaded onto 12% polyacrylamide separating and 5% stacking gels. The electrophoresis was conducted at 20 mA per gel, and the gel was stained using Coomassie Brilliant Blue R-250 and then destained.

2.4. Preparation of the RMP film

RMP was mixed with distilled water, and then, various plasticizers (glycerol, sorbitol, fructose, and sucrose) were added. Based on a preliminary experiment (data not shown), 5 g of RMP and 40% (w/w, RMP basis, 2 g) glycerol was chosen for the preparation of the RMP films. To prepare the RMP/gelatin composite films, the RMP (3, 4, and 5 g) solution and gelatin (2, 1, and 0 g) solution were mixed with 2 g glycerol. In addition, OR (0.1, 0.3, 0.5, 0.7, and 1.0 g) and the film-forming solution with 0.25 g of emulsifier (Tween #80) were added for the RMP/gelatin composite films containing OR. Subsequently, the suspension (80 mL) was cast onto a Teflon-coated plate and dried at 25 °C and 50% relative humidity for 18 h.

2.5. Measurement of mechanical properties

The RMP film's tensile strength (TS), elongation at break (E), and Young's modulus (YM) were determined using an Instron (M250–2.5 CT, The Testometric Company Ltd., Lancashire, UK). Prior to the measurement, the films were stored in a chamber controlled at 25 °C and 50% relative humidity for 2 days, and then, the TS, E, and YM of the films were evaluated. Ten replicates of each film were performed.

2.6. Water vapor permeability (WVP)

Water vapor permeability (WVP) was measured according to the method of ASTM Method E96–95 (1995).

2.7. Scanning electron microscopy (SEM)

SEM images were measured using a focused ion beam scanning electron microscope (LYRA3 XMU, Tescan, Warrendale, PA, USA) at an accelerating voltage of 30 kV. Prior to visualization, the film samples were coated with a layer of platinum.

2.8. Thermo-gravimetric analysis (TGA)

The thermo–gravimetric curves were obtained using a Mettler Toledo DSC 1 (Mettler Toledo, Columbus, OH, USA) under nitrogen flow of 50 mL/min. The samples were heated from 25 to 600 °C at the rate of 10 °C/min.

2.9. Particle size measurement

The volume-weighted mean diameter ($D_{4,3}$) of the film-forming solution was determined using a particle size analyzer (Mastersizer S, Malvern Instrument, Worcestershire, UK) at room temperature.

2.10. Antimicrobial activity of the RMP/gelatin composite film

Listeria monocytogenes (ATCC 19111), *Staphylococcus aureus* (KCTC 1621), *Salmonella* Typhimurium (ATCC 14028), and *Escherichia coli* O157:H7 (NCTC 12079) were cultured in Tryptic Soy Broth at 37 °C for 24 h. For the disc agar diffusion test, the inoculums of each microorganism (0.1 mL) were placed in selective medium. *L. monocytogenes*, *S. aureus, S.* Typhimurium, and *E. coli* O157:H7 were plated onto Oxford medium base, Mannitol salt agar, XLD, and MacConkey agar, respectively. The discs (10 mm, diameter) were cut from the RMP films containing 0.1, 0.3, 0.5, 0.7, and 1% OR and placed on the inoculated medium. Subsequently, the plates were incubated at 37 °C for 24 h, and the inhibition zones were determined. The results were obtained as the mean of three replicates.

2.11. Antioxidant activity of the RMP/gelatin composite film

The antioxidant activity of the RMP films was evaluated using DPPH and ABTS as radical scavenging methods. To prepare the film extract solution, the RMP composite film sample (25 mg) was dispersed in distilled water (5 mL) and shaken at 20 °C for 30 min. The DPPH radical scavenging activity was determined using the method as described by Shojaee-Aliabadi et al. (2013). The ABTS radical scavenging activity was measured following the procedures established by Bitencourt, Fávaro-Trindade, Sobral, and Carvalho (2014), with modification. The results were expressed as a percentage of the DPPH and ABTS radical scavenging activity, and every test was performed in triplicate.

2.12. Application of the RMP/gelatin composite film containing OR to fatty tuna meat

L. monocytogenes (ATCC 19111) and *S.* Typhimurium (ATCC 14028) were cultured in TSB at 37 °C for 24 h until the populations reached approximately 10^6 CFU/mL. Fatty tuna meat (10 g) was sanitized under UV light for 10 min, and pathogenic bacteria were inoculated onto each side of the tuna meat and desiccated for 30 min. The fatty tuna meat samples were classified into three groups: control samples in polyethylene terephthalate boxes, samples wrapped with the RMP film without OR, and samples wrapped with the RMP film containing OR. Samples of the three groups were stored at 4 °C and microbial enumeration was evaluated during a storage period of 12 days.

2.13. Microbial analysis of fatty tuna meat

Fatty tuna meat (10 g) was dispersed in 0.1% peptone water for 3 min using a stomacher (MIX 2, AES Laboratoire, Combourg, France). The mixture was serially diluted on Oxford medium base and XLD, and the plates were incubated at 37 °C for 24 h. The