Food Hydrocolloids 74 (2018) 176–186

Contents lists available at ScienceDirect

Food Hydrocolloids

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

Composite bioactive films based on smooth-hound viscera proteins and gelatin: Physicochemical characterization and antioxidant properties

Ola Abdelhedi [a], Rim Nasri [a], Mourad Jridi [a, *], Hela Kchaou [a], Benbettaïeb Nasreddine [b], Thomas Karbowiak [b], Frédéric Debeaufort [c], Moncef Nasri [a]

[a] Laboratory of Enzyme Engineering and Microbiology, National School of Engineering of Sfax (ENIS), University of Sfax, P.O. Box 1173, 3038, Sfax, Tunisia
[b] UMR A 102-02 PAM-PCAV Physico-Chemistry of Food and Wine Laboratory, 1 esplanade Erasme, Univ. Bourgogne Franche-Comté/Agrosup Dijon, F-21000, Dijon, France
[c] Dpt BioEngineering, IUT Dijon-Auxerre, 7 blvd Docteur Petitjean, 21078, Dijon cedex, France

ARTICLE INFO

Article history:
Received 3 May 2017
Received in revised form
5 August 2017
Accepted 7 August 2017
Available online 9 August 2017

Keywords:
Composite films
Mechanical properties
Water vapor permeability
Wettability
Microstructure
Antioxidant activity

ABSTRACT

The bioconversion of fish by-products through the development of biodegradable films offers the possibility to their valorization and, on the other side, to decrease the use of synthetic packaging, responsible for several ecological problems due to their non-biodegradability. Thus, in the present study, blend films (Bl-F) and bilayer films (Bi-F) based on commercial bovine gelatin (CBG) and smooth-hound viscera proteins, incorporated or not with sulfated polysaccharides (SP) or smooth-hound peptides (SHP) with molecular weight below 1 kDa, were successfully made using the casting method. Results of the scanning electron microscopy micrographs showed that the two biopolymers were compatible. Moreover, the mechanical properties analyses revealed that films based on the smooth-hound viscera proteins possessed higher tensile strength (TS) but lower elongation at break (EAB), compared to the gelatin film, where Bl-F were mechanically stronger and less deformable than Bi-F. Based on the differential scanning calorimetry analyses, Bl-F have the highest glass transition temperature (T_g) (63–67 °C), confirming their glassy character at room conditions. In addition, Bl-F were found more effective on preventing moisture transfer, in a 30–84% relative humidity differential, than Bi-F, though the SHP addition slightly reduced the water vapor barrier efficiency. The analysis of surface properties demonstrated that all films possessed hydrophobic surface. Interestingly, SHP and SP addition greatly enhanced the antioxidant property of the films allowing them to be successfully used for food industries as bioactive packaging materials.

1. Introduction

Biodegradable packagings are recently paying attention as an alternative for synthetic-derived polymers due to their edible and environment-friendly criteria. They have attracted much interest all over the world in different sectors such as food, pharmacology, agriculture and environment. Biodegradable films can be made from biopolymers, including proteins, polysaccharides, and lipids or their combination (Arfat, Benjakul, Prodpran, & Osako, 2014, 2017; Guerrero, Garrido, Leceta, & de la Caba, 2013; Hajji et al., 2016; Manivasagan & Oh, 2016). Compared to polysaccharides and lipids, edible films based on proteins possess valuable characteristics for the production of food packaging due to their good film-forming ability, mechanical properties, transparency and excellent natural barrier capacity against oxygen and carbon dioxide diffusion (Arfat et al., 2014; Lacroix & Vu, 2014). In addition, food proteins have high nutritional value for human feeding. Thus, proteins extracted from agro-food waste industry could be envisaged for active packaging applications.

Fish proteins including fish gelatin and myofibrillar and sarcoplasmic proteins have been widely used as film-forming materials for foods bio-packaging (Athaillah & Park, 2016; Blanco-Pascual, Fernández-Martín, & Montero, 2014; Jridi et al., 2013). Gelatin

* Corresponding author. Laboratoire de Génie Enzymatique et de Microbiologie, Université de Sfax, Ecole Nationale d'Ingénieurs de Sfax, B.P. 1173-3038, Sfax, Tunisia.
E-mail address: jridimourad@gmail.com (M. Jridi).

http://dx.doi.org/10.1016/j.foodhyd.2017.08.006
0268-005X/

films have been extensively studied as an outer covering to protect foods against drying, while they have poor mechanical, thermal and water vapor barrier properties (Gómez-Guillén et al., 2009). Thus, blending two or more polymers is one of the simplest methods to improve physicochemical film properties, such as gelatin/chitosan (Benbettaïeb, Kurek, Bornaz, & Debeaufort, 2014), chitosan/gelatin/poly-vinyl alcohol (Bento, Pereira, Chaves, & Stefani, 2015), and gelatin/fish protein isolate (Arfat et al., 2014) can be a promising way for the development of a final network with high functional properties. In addition, multi-layer films have gained relevance in many industrial applications, especially in the fresh food packaging industry, due to their ability on extending the shelf life of food products. In fact, Hosseini, Javidi, and Rezaei (2016) showed that multi-layer films based on poly-lactic acid and fish gelatin reduce oxygen and water vapor permeability of the resulting films. Particularly, proteins from fish byproducts are thermoplastic and contained, both, polar and non-polar amino acids, which are able to form numerous intermolecular linkages. Generally, globular proteins must be denatured by heat, acid or alkali treatments to form more extended structures that are required for film formation (Krochta, 2002). In our previous study, hydrogels from smooth-hound viscera proteins were extracted after heat treatment (Abdelhedi et al., 2016a). These hydrogels may serve as a promising starting biopolymer for film-forming solution.

Current scientific interests are being focused on incorporating bioactive substances, i.e. antioxidants, in the packaging material with the purpose to extend foods' shelf-life through the controlled release of the active component. α-tocopherol, phenolic compounds, and essential oils (Benito-Peña et al., 2016; Etxabide, Uranga, Guerrero, & de la Caba, 2016; Prodpran, Benjakul, & Artharn, 2007) are the most incorporated antioxidants in packaging biomaterials. Wang and Rhim (2015) have incorporated grapefruit seed extract, as an antimicrobial agent, in the ternary blend film hydrogel, prepared with agar, alginate and collagen.

In our previous studies, smooth-hound viscera were used as the raw material to extract bioactive sulfated polysaccharides (Abdelhedi, Nasri, Souissi, Nasri, & Jridi, 2016b) and protein hydrolysates (Abdelhedi et al., 2016c). In addition, viscera proteins were used as the protein matrix for hydrogels preparation (Abdelhedi et al., 2016a). Thus, the purpose of the present study is to prepare films (blend and bilayer) based on smooth-hound viscera gels (SHG) and gelatin. The effect of mixing these polymers on the physicochemical, structural, thermal, mechanical and surface wettability properties of films was studied. Moreover, the effect of the incorporation of smooth-hound peptides (SHP) and sulfated polysaccharides (SP), extracted from the same material, on the antioxidant activity of the resulting films was investigated.

2. Materials and methods

2.1. Chemical materials

Commercial bovine gelatin (CBG) was purchased from Sigma Chemical Co. (St. Louis, Mo., U.S.A.). Anhydrous glycerol (Loba Chemie, 98% purity, India) was used as plasticizer. Purafect® (from *Bacillus licheniformis*) was purchased from Genencor International, USA. All other reagents were of analytical grade.

2.2. Smooth-hound viscera collection

Visceral wastes (stomach and intestine) were obtained following the processing of fresh filleted smooth-hound (*Mustelus mustelus*) available in the local fish market of Sfax City, Tunisia. The biological material was brought to the research laboratory in polyethylene bags, in iced conditions, within 30 min. Upon arrival, they were immediately rinsed with tap water to remove contaminants. Then, they were stored in plastic bags at −20 °C until they were used for gel, protein hydrolysate and polysaccharide production.

2.3. Smooth-hound protein gels, sulfated polysaccharide and protein hydrolysate preparation

Alkaline smooth-hound protein gel (SHG) was prepared as described in our previous study (Abdelhedi et al., 2016a). The homogenized viscera mixture was thermally treated at 50 °C for 1 h after adjusting its pH value to 9.0. After centrifugation, the gelling solution was then freeze-dried (Bioblock Scientific Christ ALPHA 1–2, IllKrich-Cedex, France) and the resulting powder was stored at −20 °C. SHG powder was recovered with a yield of 12 g per 100 g of fresh viscera.

Sulfated polysaccharides (SP) were prepared from fish viscera by cetylpyridinium chloride precipitation (CPC) as described in our previous work (Abdelhedi et al., 2016b). On the other side, protein hydrolysate from smooth-hound visceral mass was prepared by enzymatic hydrolysis, using Purafect® (pH 10.0, 50 °C, 580 min) with an enzyme/protein ratio of 6/1 (U/mg of protein) (Abdelhedi et al., 2016c). The obtained protein hydrolysate was fractionated by ultra filtration (UF) membrane technology, in a tangential filtration mode, using a stirred cell system (Amicon, Inc., MA, USA) equipped with a molecular weight cut-off membrane of 1 kDa. Smooth-hound peptides (SHP) with molecular weight (MW) below 1 kDa were collected and freeze-dried. Peptides powder was recovered with a yield of 22.9 g per kg of fresh viscera. Both SP and SHP were stored at −20 °C to be then added in the film forming solutions.

2.4. Preparation of composite films

To prepare film-forming solutions (FFS), SHG and CBG powders were dissolved in distilled water to achieve a final concentration of 6% and 2% (w/v), respectively. These concentrations were selected based on a preliminary study and visual evaluation of films quality. Glycerol was added as plasticizer to the gelatin or SHG solutions at a level of 15% (based on protein) and FFS were mixed at 40 °C for 30 min with gentle stirring. All films were obtained by casting 25 ml of FFS on a rimmed silicone resin plate (12 cm × 12 cm), dried at 25 °C at 50% relative humidity (RH) and then peeled off.

The diagram explaining the methodology used to prepare films is presented in Fig. 1. To prepare blend film (Bl-F), 12.5 ml of the CBG-FFS and 12.5 ml of SHG-FFS were gently stirred for 30 min at 40 °C. After glycerol addition (15% based on protein), the solution was cast on the surface of the plate. For the bilayer film (Bi-F), a two-step casting technique was adopted. A volume of 12.5 ml of gelatin-FFS (2% CBG + 15% glycerol) were cast onto the surface of the plate and dried at a temperature of 25 °C and 50% RH until a firm surface formation. Thereafter, 12.5 ml of SHG-FFS (6% SHG + 15% glycerol) were directly poured on the top of the previously dried first layer and the system was dried again at 25 °C and 50% RH. After total drying of the second layer, all films were peeled off from the casting surfaces. Films resulting from gelatin and SHG film-forming solutions were named CBG-F and SHG-F, respectively, and serve as controls.

For the incorporated films, before FFS casting, SP or SHP (1 mg/ml) were dispersed in the SHG-FFS and mixed at 25 °C until complete solubility. The summary of film compositions and the methodology used for films preparation are presented in Table 1 and Fig. 1, respectively. Prior to characterization, all films were conditioned at 25 °C and 50% RH for 2 weeks.