



Preparation and functional properties of fish gelatin–chitosan blend edible films

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ABSTRACT

With the goal of improving the physico-chemical performance of fish gelatin-based films, composite films were prepared with increasing concentrations of chitosan (Ch) (100G:0Ch, 80G:20Ch, 70G:30Ch, 60G:40Ch and 0G:100Ch, gelatin:Ch), and some of their main physical and functional properties were characterised. The results indicated that the addition of Ch caused significant increase ($p < 0.05$) in the tensile strength (TS) and elastic modulus, leading to stronger films as compared with gelatin film, but significantly ($p < 0.05$) decreased the elongation at break. Ch drastically reduced the water vapour permeability (WVP) and solubility of gelatin films, as this decline for the blend film with a 60:40 ratio has been of about 50% ($p < 0.05$). The light barrier measurements present low values of transparency at 600 nm of the gelatin–chitosan films, indicating that films are very transparent while they have excellent barrier properties against UV light. The structural properties investigated by FTIR and DSC showed a clear interaction between fish gelatin and Ch, forming a new material with enhanced mechanical properties.

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1. Introduction

The environmental impact of non-biodegradable plastic material wastes is of increasing global concern. There is an urgent need to develop renewable and environmentally friendly bio-based polymeric materials (Khwaldia, Arab-Tehrany, & Desobry, 2010). Edible bio-based films have been investigated for their abilities to avoid moisture loss or water absorption by the food matrix, oxygen penetration to the food material, aromas loss and solute transports (Dutta, Tripathi, Mehrotra, & Dutta, 2009). Gelatin one of the most studied biopolymers for its film-forming capacity and applicability as an outer covering to protect food against drying, light, and oxygen (Gómez-Guillén et al., 2009a).

Fish gelatin has gained importance in recent years as a result of the outbreak of bovine spongiform encephalopathy, and the banning of collagen from pig skin and bone in some regions for religious reasons. Furthermore, fish skin, which is a major byproduct of the fish-processing industry, causing waste and pollution, could provide a valuable source of gelatin (Badii & Howell, 2006). With the appropriate film forming properties and good barriers against oxygen and aromas at low and intermediate relative humidity, gelatin is suitable for production of biodegradable packaging materials, as reviewed by Gómez-Guillén et al. (2009b). However, gelatin has relatively poor water barrier and mechanical properties, which is the main drawbacks of gelatin films for application as packaging material (Chiou et al., 2008).

One of the effective strategies, allowing maintaining the biodegradability, is to elaborate biocomposites by association of fish gelatin with other biopolymers exhibiting film forming properties and derived from renewable resources.

Chitosan is a natural polymer obtained by deacetylation of chitin, and when compared with other polysaccharides, chitosan has several advantages such as biocompatibility, biodegradability and no toxicity, while also presenting functional properties as bacteriostatic and fungistatic (Dutta et al., 2009; Shahidi, Arachchi, & Jeon, 1999). Chitosan-based materials may be used as edible films or coatings due to their unique property of increased viscosity upon hydration. Furthermore, chitosan films are tough, long-lasting, flexible, and very difficult to tear (Jeon, Kamil, & Shahidi, 2002). Hence, due to these interesting properties such as excellent film-forming capacity and good barrier and mechanical properties, it's a suitable material for designing packaging structures (Aider, 2010; Martins, Cerqueira, & Vicente, 2012). Because of its properties such as bioactivity, high mechanical strength and moisture transfer generally lower than pure gelatin films, a combination of fish gelatin with chitosan as blend could produce biocomposites with novel properties (Kolodziejaska & Piotrowska, 2007; Sionkowska, Wisniewski, Skopinska, Kennedy, & Wess, 2004).

Although some studies dealing with gelatin and chitosan composites have been performed, there is no previous report on the effect of different proportions of these biopolymers in the form of composites on the physico-chemical properties of fish gelatin-based films. Moreover, most of these studies have been focused on bovine hide gelatin and research about composite films prepared from fish gelatin are limited (Pereda, Ponce, Marcovich,

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Ruseckaite, & Martucci, 2011; Rivero, García, & Pinotti, 2009). Fish gelatin (especially cold-water fish gelatins) film, exhibits lower water vapour permeability than bovine or porcine gelatin, and this attributed to increased hydrophobicity due to lower proline and hydroxyproline contents, as the hydroxyl group of hydroxyproline is normally available to form hydrogen bonds with water molecules (Avena-Bustillos et al., 2006). Therefore, fish gelatin is a good alternate to consider when dealing with composite edible films aimed at reducing water vapour transmission. Upon considering the best characteristics of fish gelatin and chitosan separately, we speculated that their combination would lead to better films than those formed by each individual material alone.

Hence, the objectives of the present work were to examine the effect of different ratios of fish gelatin/chitosan on the functional characteristics of blend films such as water vapour permeability (WVP), film solubility, transparency and mechanical properties. The characterisation of biocomposites by thermal analysis (DSC) and infrared spectroscopy (FTIR) was performed.

2. Materials and methods

2.1. Materials

Gelatin from cold water fish skin and chitosan (medium molecular weight, 75–85% deacetylated) were purchased from Sigma–Aldrich (St. Louis, MO, USA). Glycerol (analytical grade) and acetic acid were purchased from Merck Chemicals Co. (Darmstadt, Germany).

2.2. Preparation of the films

The gelatin films were prepared according to the casting technique described by Gómez-Estaca, Gómez-Guillén, Fernández-Martín, and Montero (2011) with slight modifications. Five groups of composite films with different ratios of gelatin to chitosan (100G:0Ch, 80G:20Ch, 70G:30Ch, 60G:40Ch and 0G:100Ch) were prepared. Single gelatin film forming solution (4%, w/v) was prepared by dissolving 4 g gelatin in 100 ml distilled water for 30 min and then heated at 45 °C for 30 min under continuous stirring. Ch solution was prepared with 1.5% (w/v) Ch in 1% (v/v) acetic acid, stirred overnight at room temperature, and filtered through a micron wire mesh to remove impurities.

The gelatin–Ch film forming solutions were prepared by mixing a 6%, 4% and 2% fish-skin gelatin solution with a 1.5% solution of Ch, in a proportion of 1:1 (v/v), to obtain a film-forming solution with different proportions, respectively (solutions 80G:20Ch, 70G:30Ch and 60G:40Ch). All mixtures were warmed and stirred at 45 °C for 30 min to obtain a good blend. Glycerol (0.3 g/g gelatin or Ch or gelatin plus Ch) was added as a plasticiser and solutions were again warmed and stirred at 45 °C for 15 min. The filmogenic solutions were degassed under vacuum for 15 min to remove air bubbles. Then, aliquots of 50 g of film-forming solutions were poured in polystyrene petri dishes (14 cm of diameter) and dried at ambient temperature (23–25 °C) for 72 h to yield a uniform thickness of 90 µm ± 12 in all cases except the 0G:100Ch formulations, which was 80 µm ± 7 thick. Thickness of films was determined using a micrometer (Mitutoyo Manufacturing Co. Ltd., Tokyo, Japan) to the nearest 0.001 mm at 9 random positions around the film, and average values were used in calculations. Films were conditioned at 25 °C and a relative humidity (RH) of 50 ± 4% RH for 48 h for further analysis.

2.3. Characterisation of the films

2.3.1. Mechanical properties

A universal testing machine (SMT-20, Santam, Tehran, Iran) equipped with a 60 N load-cell was used to measure tensile

strength (TS), elongation at break (EAB%) and elastic modulus (EM) according to the ASTM standard method D 882-09 (2009). Films strips of 110 × 20 mm were conditioned at 23 ± 2 °C and 53 ± 2% relative humidity for 48 h in an environmental chamber before testing. Initial grip separation and mechanical crosshead speed were set at 50 mm and 5 mm/min, respectively. Each type of film was tested by at least five replicates.

2.3.2. Water vapour permeability

The water vapour permeability (WVP) of films was measured gravimetrically according to the ASTM E96-05 method (2005) and adapted to hydrophilic edible films by McHugh, Avena-Bustillos, and Krochta (1993). Circular test cups were made of glass with 49 mm internal diameter and 1.1 cm height. Films without pinholes or any defects were sealed to the cup mouth containing 6 ml distilled water (100% RH; 2.337 × 10³ Pa vapour pressure at 20 °C), placed in a desiccators at 20 °C and 0% RH (0 Pa water vapour pressure) containing silica gel. The water transferred through the film and adsorbed by the desiccant was determined from the weight loss of the permeation cell. The films were allowed to equilibrate for 2 h before the cells were initially weighed. The cells were weighted at intervals of 2 h during 10 h with an analytical balance (±0.0001 g). The slope of weight loss versus time was obtained by a linear regression ($r^2 \geq 0.99$). The measured WVP of the films was determined as follows:

$$WVP = \frac{WVTR \times L}{\Delta P}$$

where WVTR is the water vapour transmission rate (g mm/kPa h m²) through a film, calculated from the slope of the straight line divided by the exposed film area (m²), L is the mean film thickness (mm), and ΔP is the partial water vapour pressure difference (kPa) across the two sides of the film. Three replicates of each film were tested.

2.3.3. Water solubility

Water solubility of the films was carried out according to the method of Gontard, Guilbert, and Cuq (1992). Three pieces (1 × 4 cm²) of film were weighed (±0.0001 g) and subsequently dried in an air-circulating oven at 105 °C for 24 h. After this time films were recovered and re-weighed (±0.0001 g) to determine their initial dry weight (W_i). Afterwards, the samples were immersed in 30 ml of distilled water and the system gently shaken (100 rpm) for 24 h at room temperature (22–25 °C). The samples were then passed through a filter paper (Whatman 1). Then the filter paper together with unsolubilised fraction was dried in a forced-air oven (105 °C, 24 h) and weighed (W_f). The film solubility (FS%) was calculated using the following equation:

$$FS\% = \frac{W_i - W_f}{W_i} \times 100$$

W_i = initial dry film weight; W_f = final dry film weight.

2.3.4. Light transmission and transparency

The barrier properties of films against ultraviolet (UV) and visible light were measured at selected wavelength between 200 and 800 nm using the method described by Fang, Tung, Britt, Yada, and Dalgleish (2002). Film portions of 10 × 40 mm were placed into the test cell of a UV-1650 spectrophotometer (Model PC, Shimadzu, Kyoto, Japan), using an empty test cell as the reference. The measurement was done in triplicate and the average of three spectra was calculated. The transparency of the films was calculated by the following equation:

$$\text{Transparency}(A/\text{mm}) = -\log T/x$$

where A = absorbance at each wavelength, T = transmittance (%) at each wavelength, x = film thickness (mm). According to the equation, high transparency indicates opaque.

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