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Extracting optimization and physical properties of yellowfin tuna (*Thunnus albacares*) skin gelatin compared to mammalian gelatins

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Abstract

This work is to optimize gelatin extraction from dorsal skin of yellowfin tuna (*Thunnus albacares*) using response surface methodology, and to compare physical properties of yellowfin tuna skin gelatin with those of two mammalian skin gelatins (bovine and porcine). Central composite design was adopted in gelatin processing for extracting optimization. Concentration of NaOH (X_1), treatment time (X_2), extraction temperature (X_3) and extraction time (X_4) were chosen for independent variables. Dependent variables were gel strength (Y_1) and gelatin content (Y_2). Optimal conditions were $X_1 = 1.89$ (%), $X_2 = 2.87$ (days), $X_3 = 58.15$ (°C) and $X_4 = 4.72$ (h), and predicted values of multiple response optimal conditions were $Y_1 = 429.1$ (Bloom) and $Y_2 = 89.7$ (%). In order to investigate physical properties of yellowfin tuna skin gelatin, gel strength, gelling and melting points, and dynamic viscoelastic properties were measured. The gel strength of yellowfin tuna skin gelatin (426 Bloom) was higher than bovine and porcine gelatins (216 Bloom and 295 Bloom, respectively), while gelling and melting points were lower. Dynamic viscoelastic properties of yellowfin tuna skin gelatin did not change at 20 °C, but increase at 10 °C as a similar pattern with mammalian gelatins.

Keywords: Physical properties; Optimization; Response surface methodology; Fish gelatin; Yellowfin tuna

1. Introduction

Gelatin is a gelling protein, which has widely been applied in the food and pharmaceutical industries. According to the report of the Gelatin Manufacturers of Europe (Reference), most of commercial gelatin (95%) is made from hide of porcine and bovine and the remaining part (5%) comes from bones of porcine and bovine. By-products of poultry and fish are rarely used as a resource of gelatin. The amount of gelatin used in the worldwide food industry is increasing annually (Montero & Gómez-Guillén, 2000). However, frequent occurrences of bovine spongiform encephalopathy (BSE) and foot/mouth diseases have been problems for human health and thus by-products of mammalians are limited in utility of processing in functional food, cosmetic and pharmaceutical products. Therefore, the study of gelatin from fish by-products, such as skin and bone, has increased for the replacement of mammalian resources (Gudmundsson, 2002). A few fish gelatins are available commercially, but fish gelatin is not commonly utilized because it is inferior to mammalian gelatin in rheological properties, which affect product quality (Choi & Regenstein, 2000).

In order to be applied to food and pharmaceutical industries, fish gelatin must possess the following characteristics. First, a large quantity of by-product and its economical collection are essential to be continuously produced in industry. Second, gelatin from fish by-products must have rheological properties (gel strength, gelling and melting points, etc.) at the level of mammalian gelatin. However, it is not easy for fish by-products to satisfy the above two categories because of their bad physical properties. Fish captured in large quantities, such as anchovy, are not available for resource of gelatin because they have small body and are used whole body. Also, lumpfish (Osborne et al., 1990), tilapia (Jamilah & Harvinder, 2002), conger and squid (Kim & Cho, 1996), cod, hake, megrim and sole (Gómez-Guillén et al., 2002) have been researched to produce gelatin. They have the advantage of mass by-products such as skin, but show limit of industrial utilization because of less desirable physical properties than mammalian gelatin. Gelatin from fish bone (shark cartilage) does not have also better physical properties than mammalian gelatin (Cho et al., 2004).

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In the meantime, tuna (yellowfin, skipjack and bigeye) is one of the worldwide favorite fish that was captured about 3,400,000 MT every year (2001 World Capture Production of FAO Fisheries Department). Specially, the tuna occupies 12% of total amount of fish production in Korea (2003 Production Database of Ministry of Maritime Affairs and Fisheries of Korea). Tuna is usually processed as canned food and sliced raw meat in a factory, and by-products of tuna are affluent and collected at once. For this reason, if physical properties of gelatin from tuna skin resemble mammalian gelatin, tuna skin can possibly be a replacement resource of mammalian gelatin.

In the present study, investigations were divided into two parts. The first part was the optimization of gelatin extraction from the dorsal skin of yellowfin tuna (*Thunnus albacares*) using response surface methodology (RSM, Box & Wilson, 1951). RSM has effectiveness in the optimization and monitoring of food manufacturing processing. The basic principle of RSM is to determinate model equations that describe interrelations between the independent variables and the dependent variables (Edwards & Jutan, 1997). The second part was focused on the physical properties of yellowfin tuna (*T. albacares*) skin gelatin compared with mammalian gelatins extracted from the skins of bovine and porcine. In order to investigate physical properties of yellowfin tuna skin gelatin, gel strength, gelling and melting points and dynamic viscoelastic properties were measured and compared with bovine and porcine skin gelatins.

2. Materials and methods

2.1. Materials

Yellowfin tuna (*T. albacares*) skin was provided by Dooyoung Fisheries Co., Ltd (Busan 602-030, Korea). The yellowfin tuna skin was parted into abdominal and dorsal skins, the dorsal skin of yellowfin tuna was used in this study. Proximate composition of the skin was 56.1% moisture, 6.8% crude lipid, 1.0% crude ash and 33.6% crude protein. Content of collagen, which is a precursor of gelatin, was 13.54%. Two mammalian gelatins extracted from the skin of bovine (G 9382, 225 Bloom) and porcine (G 2500, 300 Bloom), were purchased from Sigma Chemical Co. All regents used in this study were analytical grade.

2.2. Extraction of gelatin from dorsal skin

The yellowfin tuna skin was washed, chopped and frozen at −15 °C until used. The cleaned skin was treated with 8 volumes (v/w) of alkali solution (1–3% NaOH) at 10 °C in shaking incubator at 200 rpm (HB-201SF, Hanbaek Scientific Co., Korea) for 1–5 days to remove the non-collagen protein and subcutaneous tissue after they were swollen. After the alkali treatment, the skin was neutralized with 6N HCl and washed. For hot-water extraction, six volumes (v/w) of distilled water were added and heated at temperature ranging 40–80 °C for 1–9 h. The extracted solution was centrifuged for 30 min at 900x*g* at 30 °C. The upper phase was vacuum-filtered with a filter paper (5A 110 mm, Advantec, Japan), and the filtered solution was vacuum-concentrated to 10 brix at 60 °C and dried at 1.4 m/s for 24 h in a hot-air dryer (WFO-601SD, EYELA, Japan).

2.3. Measurements of physicochemical characteristics

2.3.1. Determination of proximate components

Moisture content (oven-drying procedure), crude protein ($N \times 6.25$), lipid (ether extraction) and ash content were estimated by the AOAC official method (Horwitz, 2000). The analyses were replicated three times.

2.3.2. Determination of gelatin content

Gelatin content was estimated by measuring hydroxyproline content by the method of Sato, Ohashi, Ohtsuki, and Kawabata (1991), using a conversion factor of 11.42. Hydroxyproline content was determined by the method described in ISO (1978), with slight modifications. Dried gelatin (100 mg) was placed into test tube, and 5 ml of 6N HCl were added to test tubes. The sample solutions were hydrolyzed for 12 h at 110 °C using a dry bath. After acid hydrolysis, the sample solutions were neutralized with 6N NaOH, mixed with 2 ml of acetate/citrate buffer and massed up 25 ml with 0.3 M NaCl. Oxidant solution was prepared mixing in the proportion of one volume of 7% (w/v) chloramine T (the sodium salt of *p*-toluene sulfonchloramide) to four volumes of acetate/citrate buffer. Aliquot was transferred into test tube, and then isopropanol (300 μl) and oxidant solution (600 μl) were added and let it sit at room temperature for 4 min. After 4 min, Ehrlich's reagent solution (4 ml) was added to each tube, mixed, and heated for 25 min in water bath at 60 °C. Absorbance of the solutions was measured with a spectrophotometer (UV-140-02, Shimadzu, Japan) at 660 nm. The hydroxyproline content of the sample solutions was calculated from a calibration curve derived from standard using analytical grade hydroxyproline purchased from Sigma Chemical Co.

2.3.3. Determination of gel strength

Gel strength was determined according to the AOAC official method 948.21 (Horwitz, 2000), using rheometry (Compac-100, Sun Scientific Co. Ltd, Japan). Gelatin was dissolved with distilled water (6.67%, w/v) at 60 °C for 30 min until completely dispersed and then kept at 7 °C for 17 h. Gelatin solutions of yellowfin tuna, bovine and porcine for the measurement of gel strength, had the pH values of 6.5, 5.9 and 4.7, respectively. After cool maturation the gel strength, expressed in Bloom value, was measured with the following conditions; plunger, 12.7 mm diameter; penetration depth, 4 mm; penetration speed, 2 cm/min.