

[HTSL#134]

Comparison Studies on the Physicochemical and Functional Properties of Gelatin Extracted from Red Tilapia (*Oreochromis mossambicus*) Fish Skin and Bovine Skin using Chemical Pre-treatment

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ABSTRACT

Gelatin is widely used in the food industry as a thickening agent, emulsifier, wetting agent, and stabilizer. However, traditional sources like mammalian gelatin face sociocultural and health-related challenges, while poultry gelatin is affected by avian flu issues. To overcome these limitations, recent studies have focused on alternative gelatin sources, prompting this investigation. This study aimed to extract gelatin from red skin tilapia and bovine sources. Prior to hydrolysis, both gelatins underwent pre-treatment using 0.2 M sodium hydroxide (NaOH) and 0.05 M acetic acid (CH₃COOH) at 60 °C for 3 hours. Fourier-transform infrared (FTIR) analysis confirmed that the extracted gelatins displayed comparable peaks to commercial gelatin. In terms of gel strength, the extracted fish gelatin (EFG) exhibited higher strength than commercial fish gelatin (CFG). However, for bovine gelatin, the commercial variant (CBG) demonstrated superior gel strength compared to the extracted bovine gelatin (EBG). The study also assessed foaming capacity, with EFG displaying a higher capacity than EBG. This investigation highlighted the potential of EFG and EBG for commercial gelatin applications due to their promising physicochemical and functional properties. By exploring alternative gelatin sources, this research can offer new possibilities for the food industry.

Keywords: Gelatin, Pre-treatment, Red tilapia, Bovine, Gel strength

1 Introduction

The global gelatin demand is expected to expand by 230 million metric tons over the next five years, with a compounded annual growth rate of 5.6%. Global demand for gelatin was 620.6 kilotons in 2019 and is expected to grow at a volume-based compound annual growth rate of 5.9% from 2020 to 2027 according to Global Gelatin Market Size 2020. Gelatin from mammalian surely has more and large amounts of collagen and has been a pursuit for a long time ago. But it has a certain limitation for Muslim consumer as the halal status of mammalian and poultry-based gelatin and also has shown occurrence issues related to foot and mouth disease (FMD) in cattle. It is reported that fish gelatin (especially warm-water fish) has similar properties to porcine gelatin and can thus be used in food products as a replacement for mammalian gelatin [1]. Today, gelatin is mostly derived from bovine and porcine sources, although it may also be derived from fish and fowl. Researchers were indeed mainly



interested in the manufacturing of mammalian gelatin and the relationship between the chemical compositions and structure-function relationships of gelatins derived from mammals and warm water fish species. Therefore, in this current study, crude collagen from red tilapia skin and bovine skin could replace current mammalian as in pigs and poultry-based gelatin. Red tilapia skin and bovine skin was investigated and studied in this project to see whether they could provide stronger gelling, antioxidant, and functional properties.

2 Materials and Methods

Chemicals: Hydrochloric acid (HCl), sodium hydroxide (NaOH), acetic acid, and sulfuric acid (H₂SO₄) were obtained from MERCK.

Chemical pre-treatment: The red tilapia skin and bovine skin were pre-treated with concentration of 0.20 M NaOH about 2 hours twice under stirring condition at 27°C. The skins were then rinsed until pH 7. Then continue with pre-treated with concentration of 0.05 M CH₃COOH for another 1 hours. Pre-treated red tilapia skin and bovine skin were being rinsed with tap water until pH 7.

Water extraction: Pre-treated red tilapia skin and bovine skin were then undergo hot water extraction (60°C) in beaker fill with distilled water for 3 hours. Then the gelatin solution was filtered and dried before further analysis.

Analysis: FTIR analysis was conducted using FTIR spectrometer. Gel strength was performed using a texture analyser. Foaming capacity (FC) was assessed according to Tinrat & Sila-asna [2] where 5 ml sample solution was homogenized and centrifuged for 1 minute. The percentage of increased protein scattered throughout blending was measured as the capacity for foaming following the equation 1.

$$\text{Foam capacity} = \frac{\text{Volume of foam}}{\text{Volume of total solution}} \quad (1)$$

3 Results and Discussion

3.1 FTIR analysis

FTIR spectroscopy is advantageous for identifying the intermediate structure, confirmation of rearrangements, structural dynamics, and the stability of gelatin (Al-Saidi et al., 2012). Figure 1 shows the FTIR analysis of bovine skin gelatin (BS), red tilapia skin gelatin (FG), commercial fish gelatin (CFG) and commercial bovine gelatin (CBG).

According the FTIR spectrum, extracted gelatin from bovine and red tilapia skin exhibited a comparable pattern to commercial fish and bovine gelatin. Both extracted gelatin exhibited important protein functional group at Amide A, Amide B, and Amide I, II, and III. The EBG shows peak at 3273.59 cm⁻¹, 2918.35 cm⁻¹, 1631.27 cm⁻¹, 1530 cm⁻¹, 1077.27 cm⁻¹ whereas EFG showed peaks at 3198.34 cm⁻¹, 2922.82 cm⁻¹, 1631.27 cm⁻¹, 1530 cm⁻¹, and 1237.30 cm⁻¹ for Amide A, B, I, II, III respectively. Amide I shown that the gelatin derivative with a characteristic

coiled conformation that contributes to the stability of the triple helical structure. This shows that the C=O stretching vibration may be diminished as a result of a loose hydrogen bond formed by N-H bonding during the soaking time in acid solution. The absorption of amide-I revealed the C=O stretching vibration of the amide group and the C-N stretching vibration, whereas the absorption of amide-II revealed the N-H bending and C-N stretching vibrations. The amide III peaks corresponded to the combination of C-N stretching vibrations and N-H deformations caused by amide links, also with absorptions caused by CH₂ wagging vibrations out from glycine backbone with proline side chains.

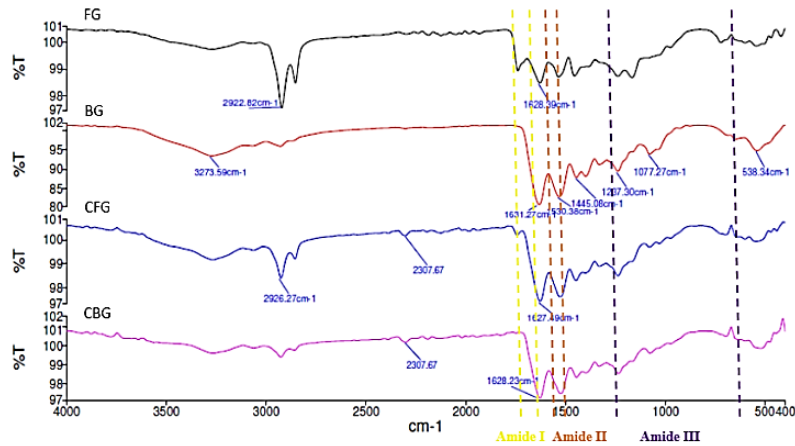


Figure 1: FTIR spectra of extracted red tilapia skin gelatin (FG), bovine skin gelatin (BG), commercial fish gelatin (CFG) and commercial bovine gelatin (CBG) along with represented amide I, amide II and amide III from wave numbers of 400-4000 cm^{-1}

3.2 Gel strength

The most essential functional attribute of fish gelatin is its gel strength, which directly affects the packaged foods quality. The gel strength of gelatin obtained from red tilapia skin, bovine skin, fish commercial and bovine commercial is shown in Figure 2.

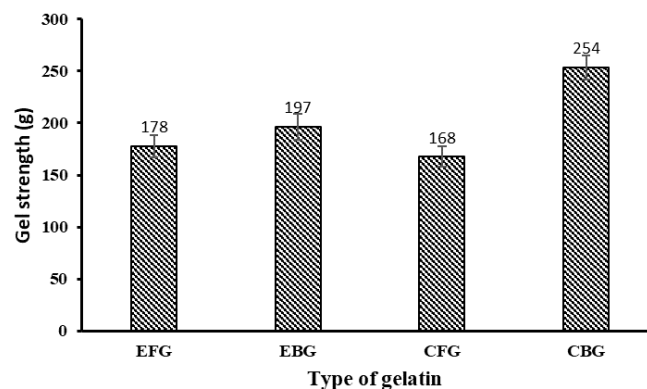


Figure 2: Gel strength (g) of extracted fish gelatin (EFG), extracted bovine gelatin (EBG), commercial fish gelatin (CFG) and commercial bovine gelatin (CBG).

Gelatin produced from CBG has the greatest gel strength, followed by EBG, EFG, and CFG which had gel strengths of 254 g, 197 g, 178 g, and 168 g respectively. Red tilapia skin gelatin has a much higher gel strength than both CFG. High gel strength of gelatin enables it to be

used in a variety of food products, including confections, to promote chewiness, texture, and foam stability [2]. Acid extraction methods produce gelatines with higher gel strengths and viscosities, whereas they have higher pressures treatment [4]. Gelatin has a high capacity for hydrogen bonding with molecules of water, which results in the formation of a resilient three-dimensional gel. Derkach and coworkers [5] claimed that fish gelatin, in contrast to gelatin obtained from mammals, has a lower content of proline and hydroxyproline, amino acids responsible for the stabilization of collagen-like triple helices, and also of lower molecular weight. For this reason, fish gelatin gels are less durable and have lower gelation and melting temperatures than mammalian gelatin.

3.3 Foaming capacity

The foaming capacity of gelatin recovered from EFG and EBG was investigated at three different gelation concentrations: 1%, 2%, and 3% as shown in Figure 3. The foaming capacity of red tilapia skin gelatin was 1.25%, 1.35%, and 1.4% at 1%, 2%, and 3% gelatin concentrations, respectively. On the other hand, the foaming capacity of bovine skin gelatin was 1.01%, 1.26%, and 1.47% at 1%, 2%, and 3% gelatin concentrations, respectively.

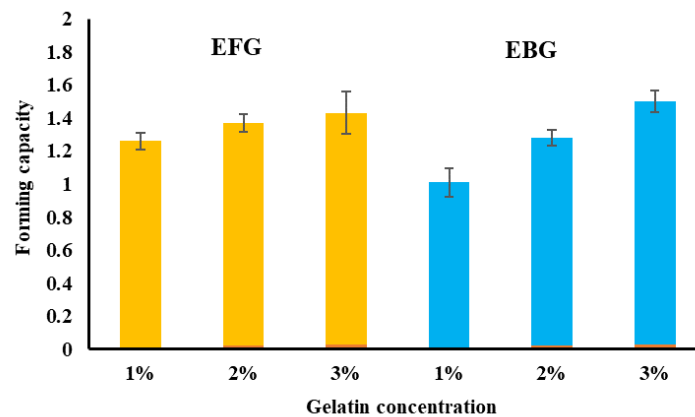


Figure 3: Foaming stability (%) of EFG and EBG at different 1%, 2% and 3% gelatin concentration.

According to the findings, increasing the gelatin content leads in an increase in the foaming capacity of both EFG and EBG. Foam generation is reliant on the transport, penetration, and structural alteration of protein molecules at the air-water interface. Proteins must be able to rapidly disperse into the air-water interface, unfold, and reorganise themselves at the interface, which can be achieved by excellent foaming capacity. EFG has a slightly higher foaming capacity than EBG might be due to its higher hydrophobic amino acid content. The foaming ability may be enhanced by the insertion of hydrophobic residues that generate a huge hydrophobic sphere on the polypeptide's surface[6].

Author Contributions

Nurfarahin Mohd Rasidi: Faiqah Fazial: Investigation, Data curation, Validation, Writing-Original Draft. Farah Faiqah Fazial: Visualization, Methodology, Supervision, Conceptualization, Funding acquisition, Writing-review & editing. Azfar Al Ariff Ahmad: Con Conceptualization, and writing review and editing.

Funding

This research received no external funding.

Acknowledgments

Authors would like to thank the Universiti Malaysia Perlis for the lab facilities and En Wan Farizan from Beseri, Perlis who supply bovine skin for this research.

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