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Article

Microwave-Assisted Extraction of Chicken Skin Gelatin: A Process–Structure–Property Study

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Abstract

Gelatin, a collagen-derived biopolymer, has attracted increasing interest as a sustainable material for advanced engineering applications owing to its biodegradability, biocompatibility, and tunable physicochemical properties. However, understanding of how microwave-assisted extraction (MAE) influences the structural evolution and material characteristics of chicken skin-derived gelatin remains limited. This study investigates the effect of MAE on the physicochemical, structural, and morphological properties of chicken skin gelatin in comparison with conventional waterbath extraction. Gelatin samples were produced under different extraction conditions and characterized in terms of yield, moisture content, ash content, pH, viscosity, Fourier Transform Infrared Spectroscopy (FTIR), UV–Visible spectroscopy, Sodium Dodecyl Sulfate–Polyacrylamide Gel Electrophoresis (SDS-PAGE), and Scanning Electron Microscopy (SEM). The highest gelatin yield obtained by MAE reached 21.19% at 200 W for 10 min, compared with 12.39% obtained by conventional waterbath extraction under the optimum condition. Microwave treatment also produced observable differences in molecular structure and surface morphology compared with conventional extraction. FTIR and UV–Vis analyses indicated changes in molecular organization, while SDS-PAGE suggested broader peptide distribution following microwave treatment. SEM observations revealed a denser and more compact microstructure in the microwave-extracted gelatin. These findings suggest that microwave-assisted extraction may provide an energy-efficient approach for producing chicken skin-derived gelatin while influencing its structural and physicochemical characteristics. The study contributes to understanding the process–structure–property relationship of gelatin biopolymers and provides useful information for future development of sustainable biomaterials and advanced material processing technologies.

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1. Introduction

The increasing demand for renewable and biodegradable materials has stimulated interest in converting agro-industrial and food-processing by-products into value-added biopolymers. Gelatin, obtained through the partial hydrolysis of collagen, is particularly attractive because of its biodegradability, film-forming ability, biocompatibility, and ability to form thermoreversible gels. These characteristics have enabled gelatin to be investigated not only for food applications but also as a matrix or precursor for biodegradable films, coatings, hydrogels, composite materials, and tissue-engineering scaffolds. Recent experimental studies have demonstrated that gelatin-based materials can be engineered through crosslinking and composite formation to improve their mechanical, barrier, thermal, and structural properties, indicating that gelatin can serve as a versatile platform for advanced-material development [1], [2].

Chicken skin represents an underutilized collagen-rich poultry by-product that can serve as an alternative raw material for gelatin production. However, the efficiency and characteristics of chicken-skin gelatin depend strongly on the extraction conditions. Conventional wet-rendering extraction, for example, produced gelatin yields of only 0.74–2.03% from chicken skin, although the resulting gelatin exhibited favorable rheological and functional properties compared with commercial bovine gelatin [3]. In another study using an enzymatic pretreatment strategy, chicken-skin gelatin yield reached 31.5%, while gelatin with a viscosity of 4.06 mPa·s and gel strength of 190 Bloom was obtained under a different optimized condition, demonstrating that extraction strategy can substantially alter both recovery and functional quality [4]. These contrasting results indicate that extraction yield alone cannot be used to evaluate gelatin quality because processing conditions can simultaneously influence molecular characteristics and functional properties.

The limitations of prolonged conventional heating have encouraged the development of alternative energy-assisted extraction technologies. Microwave-assisted extraction (MAE) is particularly promising because microwave energy can generate volumetric heating through interaction with polar molecules and ions, potentially accelerating heat transfer, collagen disruption, and solubilization. Recent experimental evidence supports the potential of this approach for poultry gelatin. Esmaeili-Kaliji et al. (2025), for example, obtained 10.48% yield from chicken feet using microwave treatment at 540 W for 20 min, with the microwave-extracted gelatin exhibiting improved viscosity and gel strength compared with the corresponding acidic extraction. Other energy-assisted technologies have shown similar advantages: ohmic heating of chicken skin increased gelatin yield and functional properties while reducing extraction time, whereas ultrasound-assisted treatment of poultry skin–head–feet mixtures improved extraction efficiency, rheological properties, gel strength, and viscosity while reducing the processing burden [5], [6], [7], [8].

Despite these developments, the available literature remains heterogeneous in terms of raw material, pretreatment, heating mechanism, and extraction conditions, making direct comparison of gelatin performance difficult. Recent chicken-feet research, for instance, reported yields of 7.06–7.93%, moisture below 15%, and high Bloom strengths of 203–326 g, whereas microwave extraction of chicken feet produced a yield of 10.48% under optimized conditions [6], [9]. These differences demonstrate that extraction technology can influence gelatin recovery and functionality, but they also indicate that results obtained from chicken feet cannot necessarily be extrapolated to chicken skin. More importantly, many studies emphasize yield, rheology, or functional properties independently, while fewer studies integrate extraction efficiency with molecular and microstructural characterization to explain how processing conditions translate into material properties [10], [11], [12].

This limitation represents an important process–structure–property gap for chicken-skin gelatin. A higher extraction yield does not necessarily indicate a superior engineering material because excessive thermal or chemical treatment may alter peptide-chain distribution, intermolecular interactions, and the resulting morphology. Conversely, preservation of characteristic protein functional groups does not by itself establish suitability for a specific material application. A more integrated assessment is therefore required to determine whether an accelerated extraction process can improve gelatin recovery while maintaining physicochemical and structural characteristics that are relevant to subsequent material processing. This issue is particularly important for gelatin-based packaging and biomaterial systems, where the extracted gelatin generally functions as a precursor or matrix and additional formulation, crosslinking, or reinforcement is often required to obtain application-specific mechanical and barrier properties [1], [13].

Recent material studies illustrate this distinction clearly. Gelatin-based films can provide biodegradable film matrices, but their mechanical and moisture-barrier performance often requires modification through crosslinking or reinforcement. For example, incorporation of bacterial cellulose into gelatin films combined with transglutaminase crosslinking increased tensile strength by 42.79% and reduced water-vapor and oxygen permeability by 19.33% and 40.15%, respectively [13]. Similarly, gelatin-based composite films crosslinked with gallic acid have demonstrated improved mechanical, surface, and water-resistance characteristics compared with unmodified films [1]. These findings indicate that the extraction step should be regarded as an upstream material-processing stage: it establishes the molecular and physicochemical characteristics of the gelatin precursor, whereas final engineering performance depends on subsequent formulation and processing [14].

Accordingly, the present study aims to compare microwave-assisted extraction and conventional waterbath extraction of chicken skin gelatin and to establish the relationship between extraction conditions, structural characteristics, and resulting material properties.

The study quantitatively evaluates extraction yield, moisture content, ash content, pH, and viscosity and subsequently characterizes the extracted gelatin using FTIR spectroscopy, UV–Visible spectroscopy, SDS–PAGE, and SEM. The results are integrated to elucidate how microwave processing affects collagen-to-gelatin conversion, molecular organization, peptide distribution, and surface morphology. The scientific contribution of the study therefore extends beyond demonstrating extraction efficiency: it provides an integrated assessment of the process–structure–property relationship of microwave-extracted chicken skin gelatin and evaluates its potential as an upstream biopolymer precursor for subsequent engineered material formulations. Because application-specific mechanical, barrier, degradation, and biological properties were not directly measured in this study, the discussion of packaging and biomaterial applications is limited to material feasibility and precursor potential, rather than direct claims of application performance.

2. Materials and Methods

2.1. Materials

Fresh chicken skin (*Gallus gallus domesticus*) was obtained from a local poultry slaughterhouse in Padang, Indonesia and used as the collagen source for gelatin extraction. Citric acid (1%, w/v), sodium hydroxide (0.15%, w/v), and distilled water were used during the pretreatment and extraction processes. All chemicals were of analytical grade and used without further purification.

2.2 Instruments

The experimental work was performed using a thermostatic water bath (B-One, Indonesia), a domestic microwave oven operating at a frequency of 2.45 GHz with a fixed microwave output power of 200 W, a dehydrator (Lohame, China), a drying oven (Jisico, Republic of Korea), a muffle furnace (Jisico, Republic of Korea), an Ostwald capillary viscometer, a pH meter (Mediatech, China), a Fourier Transform Infrared (FTIR) spectrometer (Specord 210, Germany) equipped with an Attenuated Total Reflectance (ATR) accessory, a UV–Visible spectrophotometer (Thermo Scientific, USA), an SDS–PAGE electrophoresis system, laboratory filter paper with a diameter of 11 cm, an analytical balance, a magnetic stirrer, and standard laboratory glassware.

For morphological characterization, a Thermo Scientific Phenom Desktop Scanning Electron Microscope (SEM) was used. The SEM observation was performed at an accelerating voltage of 15 kV and a magnification of 1000×.

2.3 Experimental design

The study employed a comparative exploratory experimental design to evaluate the effects of conventional waterbath extraction and microwave-assisted extraction (MAE) on the characteristics of chicken skin gelatin. Conventional extraction was performed at a fixed temperature of 65 °C using extraction times of 10, 20, and 30 min, whereas MAE was conducted at a fixed microwave output power of 200 W for 1, 3, 5, 7, and 10 min.

To minimize variation arising from pretreatment, the NaOH concentration, citric acid concentration, water-to-sample ratio, and pretreatment procedure were maintained consistently for both extraction methods. The water-to-sample ratio was maintained at 10:1 (v/w), while NaOH and citric acid concentrations were maintained at 0.15% (w/v) and 1% (w/v), respectively.

The study was designed primarily to identify comparative processing trends rather than to establish statistical significance among treatment means. Therefore, the results are interpreted as exploratory comparative observations.

2.4 Sample preparation

Sample preparation was adapted from a previously reported chicken-skin gelatin preparation procedure with slight modifications [15]. Fresh chicken skin was manually cleaned to remove adhering fat, connective tissue, and residual meat before being washed thoroughly with running tap water. The cleaned skin was cut into approximately 2–3 cm pieces and washed once again to remove remaining impurities. The prepared samples were subsequently air-dried at room temperature for 2–3 days till visibly dry before pretreatment. The overall experimental procedure, including sample preparation, gelatin extraction,

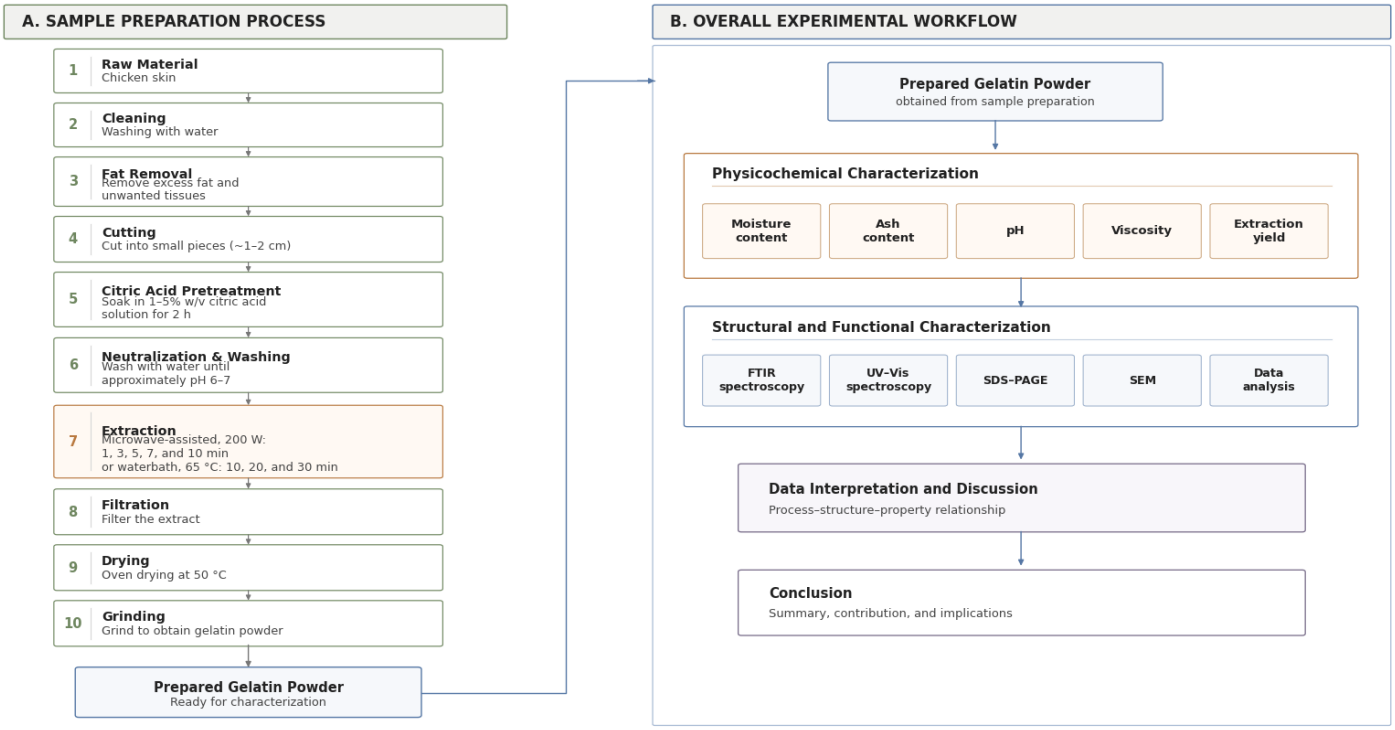

Figure 1. Flowchart illustrates the sample preparation process and the overall experimental workflow employed in this study.

physicochemical characterization, and structural analysis, is summarized in the flowchart shown in Figure 1.

2.5 Collagen pretreatment

Approximately 15 g of dried chicken skin was immersed in 200 mL of 0.15% (w/v) sodium hydroxide solution and continuously stirred using a magnetic stirrer for 40 min. The alkaline solution was replaced after each 40-min treatment, and the treatment was repeated three times to facilitate the removal of non-collagenous proteins and other impurities.

After alkaline treatment, the samples were thoroughly washed with distilled water and subsequently immersed in 200 mL of 1% (w/v) citric acid solution under the same treatment conditions. The citric acid solution was replaced every 40 min, and the treatment was repeated three times. Following acid treatment, the samples were washed with distilled water until the washing solution reached pH 4–6.

2.6 Gelatin extraction

Gelatin extraction was carried out using a water-to-sample ratio of 10:1 (v/w). For the conventional extraction, pretreated chicken skin was heated in a thermostatic water bath at 65°C for 10, 20, or 30 min. For microwave-assisted extraction, the samples were irradiated in a domestic microwave oven operating at 2.45 GHz with a fixed microwave power of 200 W for 1, 3, 5, 7, or 10 min.

After extraction, the gelatin solution was filtered through laboratory filter paper (11 cm diameter) to remove insoluble residues. The filtrate was subsequently dried in a laboratory drying oven (Jisico, Republic of Korea) until a constant weight was obtained. The dried gelatin sheets were collected and stored in airtight containers before further characterization. The extraction procedure was adapted from Kim et al. (2020) with modifications according to the present experimental conditions [16].

2.7 Sample coding

Samples were coded based on extraction method and processing conditions to facilitate systematic analysis, as summarized in Table 1.

Table 1. Sample codes for chicken skin gelatin extracted by waterbath and microwave methods.

No	Sample Description	Code
1	Waterbath 65°C for 10 minutes	GW65-10
2	Waterbath 65°C for 20 minutes	GW65-20
3	Waterbath 65°C for 30 minutes	GW65-30
4	Microwave 200 W for 1 minute	GM200-1
5	Microwave 200 W for 3 minutes	GM200-3
6	Microwave 200 W for 5 minutes	GM200-5
7	Microwave 200 W for 7 minutes	GM200-7
8	Microwave 200 W for 10 minutes	GM200-10

2.8 Physicochemical characterization

The extracted gelatin was characterized in terms of extraction yield, moisture content, ash content, pH, and viscosity. The quality characteristics of the extracted gelatin were evaluated with reference to SNI 01-3735-1995, which specifies the quality requirements for gelatin, including moisture and ash content (Badan Standardisasi Nasional, 1995)

2.8.1 Extraction yield

Extraction yield was determined gravimetrically by comparing the dry weight of gelatin obtained after extraction with the initial dry weight of chicken skin used in the extraction process. The yield was calculated according to Equation (1).

$$Y\ (\%) = \frac{W_g}{W_0} \times 100 \quad (1)$$

where is the Y is the extraction yield (%), W_g is the weight of dried gelatin obtained after extraction (g), and W_o is the initial dry weight of chicken skin used for extraction (g). The extraction-yield determination was performed using a gravimetric procedure specific to the present extraction experiment and was not treated as a conformity test under SNI 01-3735-1995.

2.8.2 Moisture content

Moisture content was determined by the oven-drying method. Approximately 2 g of gelatin was dried at 105 °C until a constant weight was obtained. The moisture content was expressed as a percentage of the initial sample weight. The analytical procedure was based on the oven method specified in SNI 01-2891-1992, Section 5.1 (Oven Method) [18].

2.8.3 Ash content

Ash content was determined by incinerating the dried gelatin samples in a muffle furnace at 600 °C until a constant weight was obtained. The result was expressed as a percentage of the initial sample weight.

The ash content was evaluated with reference to SNI 01-3735-1995, which specifies a maximum ash content of 3.25% for gelatin. Because the experimental ashing temperature used in the present study (600 °C) differs from the maximum temperature specified in the general total-ash procedure of SNI 01-2891-1992, the present procedure is reported explicitly as the experimental method used rather than being described as a strict replication of the SNI 01-2891-1992 procedure.

2.8.4 pH and viscosity measurement

The pH of the gelatin solution was measured at room temperature using a calibrated pH meter. For viscosity measurement, gelatin was prepared at a concentration of 6.67% (w/v) by dissolving 0.667 g of gelatin in 10 mL of distilled water. The gelatin solution was heated at approximately 55–60 °C to facilitate complete dissolution. The viscosity was measured at 60 °C using an Ostwald capillary viscometer. The gelatin concentration and measurement temperature were selected with reference to the Gelatin Manufacturers Institute of America (GMIA), Standard Methods for the Testing of Edible Gelatin (revised July 2013), which specifies viscosity determination using a 6.67% gelatin solution at 60 °C[19]. Because the present study employed an Ostwald capillary viscometer rather than the standard 100-mL pipette specified in the GMIA procedure, the GMIA method was used as a methodological reference rather than as an exact replication of the standard procedure.

2.9 Structural characterization

The structural characteristics of the extracted gelatin were evaluated using Fourier Transform Infrared (FTIR) spectroscopy, UV–Visible spectroscopy, sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS–PAGE), and scanning electron microscopy (SEM). These complementary techniques were used to evaluate the characteristic functional groups, electronic absorption behavior, molecular-weight distribution, and surface morphology of the extracted gelatin, respectively.

FTIR spectroscopy was performed using a Specord 210 FTIR spectrometer equipped with an Attenuated Total Reflectance (ATR) accessory. The spectra were recorded over the range of 4000–600 cm^{-1}. The gelatin samples were analyzed by direct measurement without KBr pellet preparation. The dried sample was placed directly onto the ATR crystal and pressed using the pressure arm to obtain adequate contact between the sample and the crystal surface before spectral acquisition. The resulting spectra were evaluated with particular attention to the characteristic Amide A, Amide I, Amide II, and Amide III regions.

UV–Visible spectroscopy was performed using a Thermo Scientific UV–Visible spectrophotometer. Gelatin solutions were analyzed over the wavelength range of 200–800 nm using a quartz UV-grade cuvette. The resulting absorption spectra were compared among the different extraction conditions to evaluate changes in the electronic absorption characteristics of the extracted gelatin.

SDS–PAGE was used to qualitatively evaluate the molecular-weight distribution of the proteins present in the extracted gelatin. Electrophoresis was performed using a 4% stacking gel and a 7% separating gel, with a prestained molecular-weight marker covering 11–245 kDa. Samples were initially electrophoresed at 80 V to facilitate protein stacking. After the protein bands reached the interface between the stacking and separating gels, the voltage was increased to 110 V until electrophoresis was completed. The gel was stained using Coomassie Brilliant Blue R-250 and subsequently destained until a clear background was obtained before documentation of the protein banding patterns.

SEM analysis was performed using a Thermo Scientific Phenom Desktop SEM to examine the surface morphology of the optimum microwave-extracted gelatin (GM200-10). The sample was mounted on an aluminum stub and examined at an accelerating voltage of 15 kV and a magnification of 1000×. The resulting micrograph was evaluated for surface features such as compact regions, pores, cracks, and other morphological characteristics. The coating condition was not specified in the available experimental record and is therefore not stated here.

2.10. Statistical limitation

This study employed a comparative exploratory experimental design to evaluate differences between conventional waterbath extraction and microwave-assisted extraction. The independent variables consisted of extraction method and extraction time, whereas the dependent variables included gelatin yield, moisture content, ash content, pH, viscosity, molecular characteristics, and surface morphology.

Because the objective of this work was to establish preliminary comparative observations and identify processing trends, replicated experimental runs were not performed. Consequently, statistical significance testing and uncertainty estimation were beyond the scope of the present investigation. The findings should therefore be interpreted as comparative observations intended to support future studies involving replicated experiments and statistical validation.

3. Results and Discussion

3.1. Physicochemical properties

3.1.1. Yield

Yield is a primary indicator of extraction efficiency because it reflects the extent to which collagen is converted into soluble gelatin during the extraction process. As shown in Figure 2, microwave-assisted extraction (MAE) consistently produced higher gelatin yields than conventional waterbath extraction under the investigated conditions. The highest yield (21.19%) was obtained at 200 W for 10 min, whereas the optimum yield achieved by the waterbath method was 12.39% after 10 min of extraction.

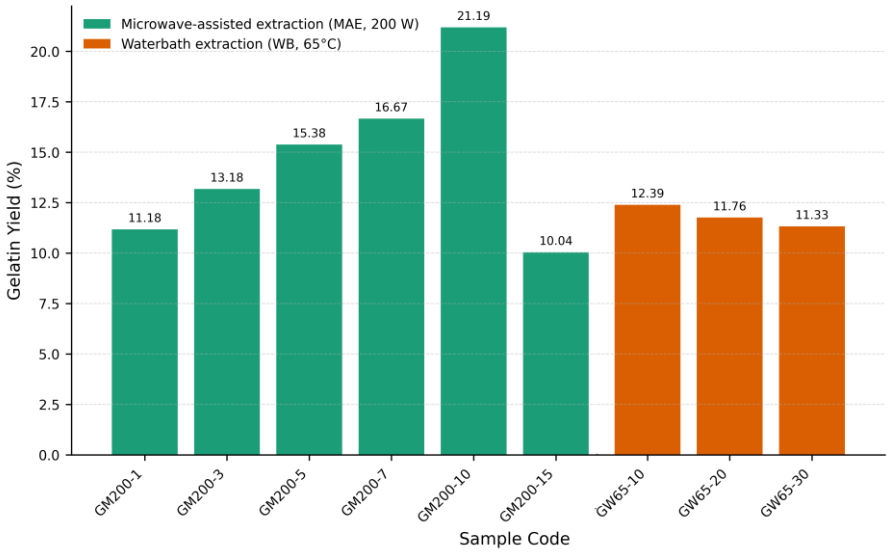

Figure 2. Gelatin yield by extraction method.

Increasing microwave extraction time from 1 to 10 min increased the yield from 11.18% to 21.19%, corresponding to an approximately 89.5% increase. In contrast, extending waterbath extraction from 10 to 30 min decreased the yield from 12.39% to 11.33%. The opposite trends indicate that extraction time interacted differently with the two heating mechanisms. Under microwave irradiation, increasing treatment time within the investigated range appears to have promoted progressive collagen disruption and gelatin solubilization. In contrast, prolonged conventional heating beyond 10 min did not improve gelatin recovery and may have promoted further degradation or changes in soluble collagen-derived components that reduced the amount of material recovered as dried gelatin.

The higher yield obtained by MAE can be attributed to the volumetric heating characteristic of microwave irradiation. Unlike conventional waterbath heating, in which heat is transferred primarily from the external heating medium toward the sample, microwave energy is absorbed directly by polar species in the extraction system. This mechanism can accelerate heating and facilitate collagen swelling, disruption of intermolecular interactions, and diffusion of soluble gelatin molecules into the extraction medium. The rapid response observed in the present study is therefore consistent with the broader evidence that microwave treatment can shorten the extraction time required to obtain collagen-derived gelatin.

A quantitative comparison with previous studies demonstrates that the yield obtained in the present study is competitive, although the comparison must account for differences in raw material and processing conditions. Microwave extraction of duck skin at 200 W for 10 min produced a gelatin extraction yield of 28.51%, whereas microwave extraction of rabbit skin at 500 W produced a maximum yield of 6.44% after 60 min [16], [20]. The substantially different yields despite the use of microwave energy demonstrate that microwave power alone does not determine extraction efficiency; raw-material composition, pretreatment, solid-to-liquid ratio, extraction temperature, and processing time also strongly influence collagen conversion.

More directly relevant poultry evidence also illustrates this variability. Microwave extraction of chicken feet under an optimized condition of 540 W for 20 min produced a yield of 10.48%, while acid extraction under the corresponding optimized condition produced 11.44% [6]. Conventional extraction of broiler chicken feet has also produced yields of approximately 7.93%, with moisture below 15% and viscosity of 3.85 cP at 6.67% gelatin concentration (Usman et al., 2024). A more directly relevant comparison can be made with chicken gelatin obtained from mechanically deboned residues, for which an optimized hot-water extraction at 80 °C for 6 h produced a yield of 13.85% [21]. The present yield of 21.19% is therefore higher than these reported chicken-feet values, but the comparison should not be interpreted as proof that chicken skin is universally superior as a gelatin source because anatomical tissue composition and pretreatment conditions differ substantially among poultry by-products.

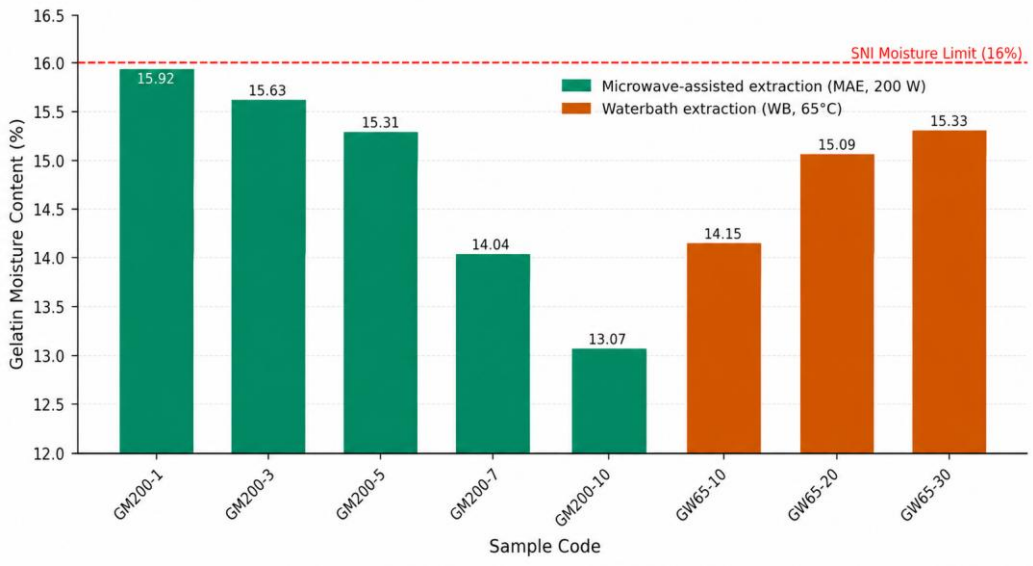

Figure 3. Gelatin moisture content by extraction method.

The most important result from the present comparison is therefore not simply the absolute yield but the combination of yield and processing time. The optimum MAE condition achieved 21.19% recovery within only 10 min, whereas extending waterbath treatment to 30 min did not improve recovery. This finding supports MAE as an efficient upstream extraction strategy for chicken skin gelatin and provides the basis for examining whether the increased extraction efficiency is accompanied by acceptable physicochemical and structural characteristics.

3.1.2. Moisture content

Moisture content is an important quality parameter because it influences the storage stability, shelf life, and handling characteristics of gelatin. As shown in Figure 3, the moisture content of gelatin obtained under different extraction conditions ranged from 13.07% to 15.92%, corresponding to a relatively narrow variation of approximately 2.85 percentage points. This limited variation indicates that the extraction method had only a minor influence on the final moisture content of the dried gelatin.

The slightly broader variation observed among the MAE samples may be associated with rapid volumetric heating and localized evaporation during microwave treatment. Such effects can modify the distribution of water within the gelatin-containing matrix and consequently influence the subsequent drying behavior. Nevertheless, the differences remained relatively small, indicating that the higher extraction efficiency achieved by MAE was not accompanied by a substantial increase in residual moisture.

The moisture values of 13.07–15.92% were below the 16% maximum specified by SNI 01-3735-1995, which is currently listed by the Indonesian National Standardization Agency (BSN) as a valid standard for gelatin (Badan Standardisasi Nasional, 1995). The result therefore indicates that the extracted gelatin satisfied the applicable moisture specification despite the differences in extraction method.

The moisture content should, however, be distinguished from the water-barrier performance of a gelatin-based material. Moisture content describes the residual water in the extracted gelatin powder, whereas packaging performance depends on properties such as water-vapor permeability, water solubility, and moisture sorption after film formation. Therefore, the present values demonstrate acceptable moisture characteristics of the gelatin precursor but do not by themselves establish its suitability as a moisture barrier.

This distinction is important for engineering applications because gelatin-based films generally require further modification to overcome their hydrophilic character. Recent experimental work showed that gelatin films reinforced with 4% bacterial cellulose and crosslinked with transglutaminase exhibited a 42.79% increase in tensile strength, together with reductions of 19.33% in water-vapor permeability and 40.15% in oxygen permeability, demonstrating that subsequent formulation can substantially modify the performance of gelatin-based materials [13].

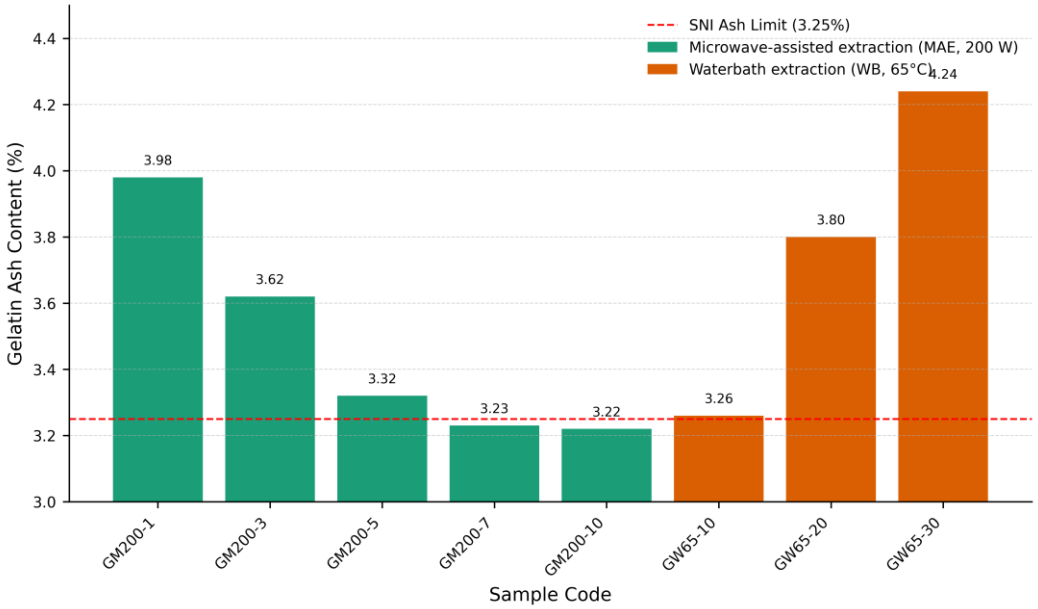

Figure 4. Gelatin ash content by extraction method.

3.1.3. Ash content

Ash content represents the residual inorganic constituents remaining after extraction and is widely used as an indicator of gelatin purity. Lower ash content generally reflects more effective removal of mineral residues and other inorganic impurities during processing. As shown in Figure 4, microwave-assisted extraction (MAE) produced a gradual decrease in ash content with increasing extraction time, reaching the lowest value of 3.22% after 10 min of microwave treatment.

The ash content obtained in the present study can also be compared with gelatin produced from mechanically deboned chicken meat by-products. An ash content of approximately 5.0% was reported under optimized enzymatic conditioning and gelatin extraction at 73–78 °C for 100–150 min [22]. In comparison, the present microwave extraction produced a lower ash content of 3.22% after only 10 min of treatment. Although the difference suggests more effective removal of inorganic residues under the present conditions, the comparison should be interpreted cautiously because the raw materials and pretreatment strategies were substantially different.

The reduction in ash content with increasing microwave treatment time may be related to improved mass transfer during extraction. Rapid volumetric heating can promote movement of soluble components between the collagen matrix and extraction medium, potentially facilitating the removal of residual inorganic species during the subsequent washing and filtration stages. However, this interpretation should be treated cautiously because ash content is affected not only by the heating mechanism but also by the efficiency of alkaline and acid pretreatment, washing, and the intrinsic mineral content of the raw material.

The residual inorganic fraction may partly originate from salts associated with the NaOH and citric-acid pretreatment stages. Consequently, the final ash value represents the combined effects of raw-material composition, pretreatment, extraction, and post-extraction washing. The lower ash value obtained after 10 min of MAE therefore suggests improved purification under the applied condition, but it does not establish that microwave energy alone was responsible for the difference.

The minimum ash content of 3.22% is very close to the maximum value of 3.25% specified in SNI 01-3735-1995 [17].This result is important because it identifies ash removal as a process parameter that still has limited optimization margin. A small improvement in washing or downstream purification could potentially reduce the ash content further below the specification and provide a more robust starting material for subsequent formulation.

The result is particularly relevant to engineered materials because residual inorganic species can influence the reproducibility of gelatin formulations and interactions with added polymers, nanoparticles, or crosslinking agents. Therefore, although the 3.22% ash content is close to the applicable quality limit, further purification would be desirable before the gelatin is used in applications requiring controlled composition.

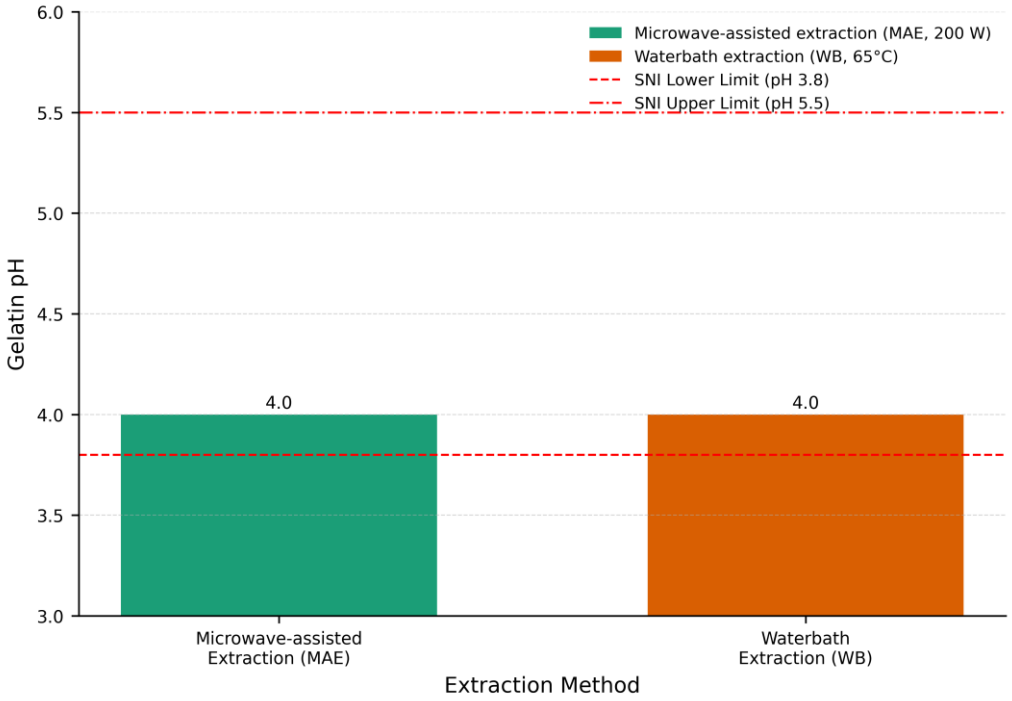

Figure 5. pH values of gelatin by extraction method.

3.1.4. pH

The pH of gelatin is an important quality parameter because it influences chemical stability, protein solubility, and intermolecular interactions that contribute to the functional properties of gelatin. As shown in Figure 5, both microwave-assisted extraction (MAE) and conventional waterbath extraction produced gelatin with similar pH values of approximately 4.0, indicating that the extraction method had only a minor influence on the acidity of the final product under the applied experimental conditions.

The relatively consistent pH values obtained for all treatments are primarily attributed to the use of citric acid during the pretreatment stage. Citric acid provides a controlled acidic environment for collagen swelling and partial hydrolysis, and the subsequent washing process was conducted until the sample reached the predetermined pH range prior to extraction. Consequently, both extraction methods started from comparable chemical conditions, resulting in only slight variations in the final pH of the extracted gelatin.

The pH values obtained in the present study are consistent with the characteristics of acid-processed (Type A) gelatin, although variations in pH have been widely reported depending on collagen source, pretreatment conditions, and extraction procedures. For instance, a pH of approximately 5.2 was reported for chicken skin gelatin, whereas a lower pH of approximately 3.0 was obtained for duck skin gelatin [16].

From a molecular perspective, pH influences the protonation state of amino acid residues and therefore affects electrostatic interactions among gelatin molecules, which are important for molecular organization and gel formation [23]. Although the present study did not evaluate gel strength or other functional properties directly, the relatively stable pH observed across all treatments suggests that microwave-assisted extraction did not substantially alter the chemical environment established during pretreatment. This finding indicates that the differences observed in yield and structural characteristics are more likely associated with the extraction mechanism itself rather than changes in the acidity of the gelatin system.

3.1.5. Viscosity

Viscosity is an important functional property of gelatin because it reflects the flow behavior of gelatin solutions and is closely associated with the molecular weight, chain length, and intermolecular interactions of gelatin molecules. As shown in Figure 6, microwave-assisted extraction (MAE) produced higher viscosity values than conventional waterbath extraction under the investigated conditions. The highest viscosity (3.25 cP) was obtained for the MAE sample extracted at 200 W for 10 min, whereas waterbath-extracted

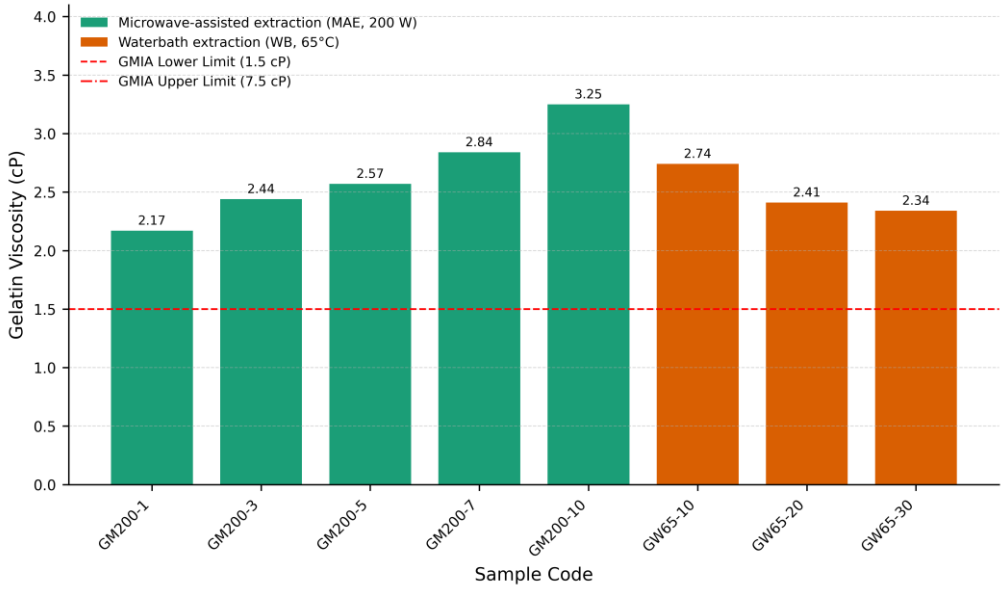

Figure 6. Gelatin viscosity by extraction method.

samples exhibited viscosities ranging from 2.34 to 2.74 cP. All measured viscosity values were within the range commonly reported for commercial gelatin (approximately 1.5–7.5 cP), indicating that the extracted gelatin exhibited acceptable solution-flow characteristics for further processing.

Increasing microwave extraction time from 1 to 10 min increased viscosity from 1.63 to 3.25 cP, corresponding to an approximately two-fold increase. This trend is consistent with the yield results, where increasing microwave extraction time also increased gelatin recovery. The simultaneous increase suggests that longer microwave treatment promoted greater solubilization of collagen-derived gelatin molecules into the extraction medium.

The relationship should nevertheless be interpreted carefully. Viscosity is affected by both molecular characteristics and concentration, and molecular-weight distribution was not quantitatively determined in the present study. Therefore, the increase from 1.63 to 3.25 cP cannot be attributed solely to an increase in molecular weight. Instead, it may reflect a combination of greater gelatin recovery, changes in peptide-chain distribution, and increased intermolecular entanglement in the test solution.

The measured viscosity is within the range reported for commercial gelatin. More specifically, recent work on chicken-feet gelatin reported viscosities of 4.43 cP for native chicken feet gelatin and 3.85 cP for broiler chicken feet gelatin, both within the reported commercial range of approximately 2–7 cP [9]. The present value of 3.25 cP is therefore somewhat lower than those two chicken-feet values but remains within the same practical range, indicating that the extracted chicken-skin gelatin has comparable solution-flow characteristics.

The viscosity obtained in the present study was also comparable to that reported for chicken gelatin extracted from mechanically deboned residues. A viscosity of approximately 2.5–2.7 mPa·s was reported for chicken gelatin obtained at 80 °C for 6 h, whereas the optimum gelatin in the present study exhibited a viscosity of 3.25 cP at a substantially shorter extraction time [21]. This difference suggests that the microwave process was able to produce gelatin with comparable solution-flow characteristics despite the markedly shorter extraction duration. A viscosity of approximately 2.5 mPa·s was reported for gelatin obtained from mechanically deboned chicken meat, whereas the present chicken-skin gelatin exhibited a viscosity of 3.25 cP under the optimum microwave condition, indicating broadly comparable solution-flow behavior [22].

The viscosity result also has processing relevance. A gelatin solution must have sufficient flowability for casting, coating, mixing, or other solution-based processing, but excessive viscosity can hinder mass transfer and film formation. The value of 3.25 cP obtained at the optimum MAE condition therefore provides a useful baseline for subsequent formulation studies. It should not, however, be interpreted as evidence of specific film or scaffold

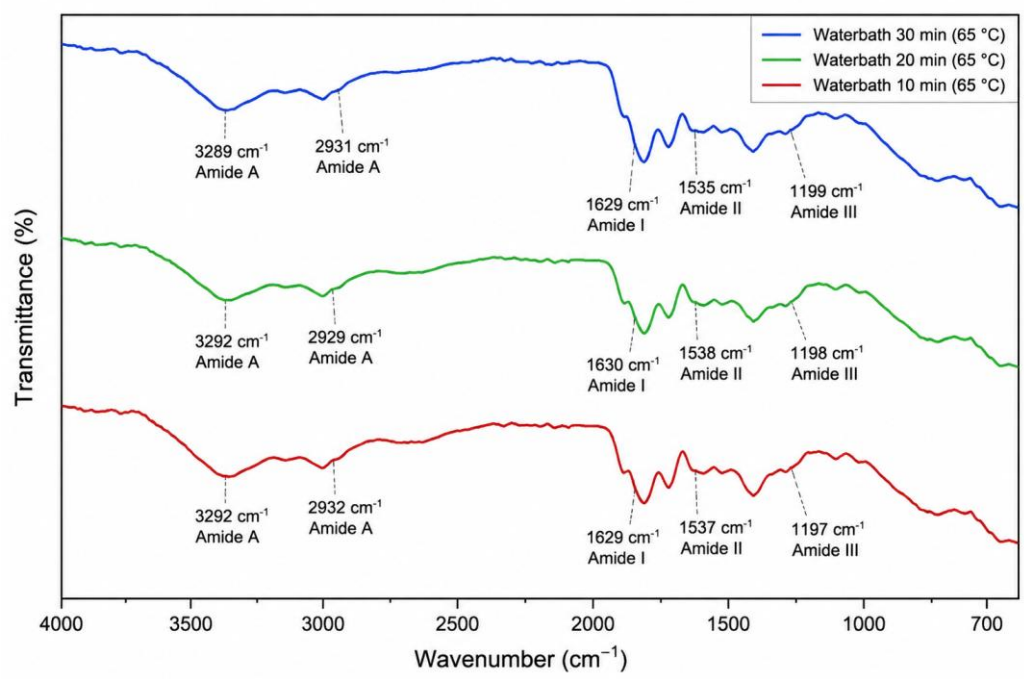

Figure 7. FTIR spectra of chicken skin gelatin extracted by conventional waterbath extraction (65°C) for 10, 20, and 30 minutes.

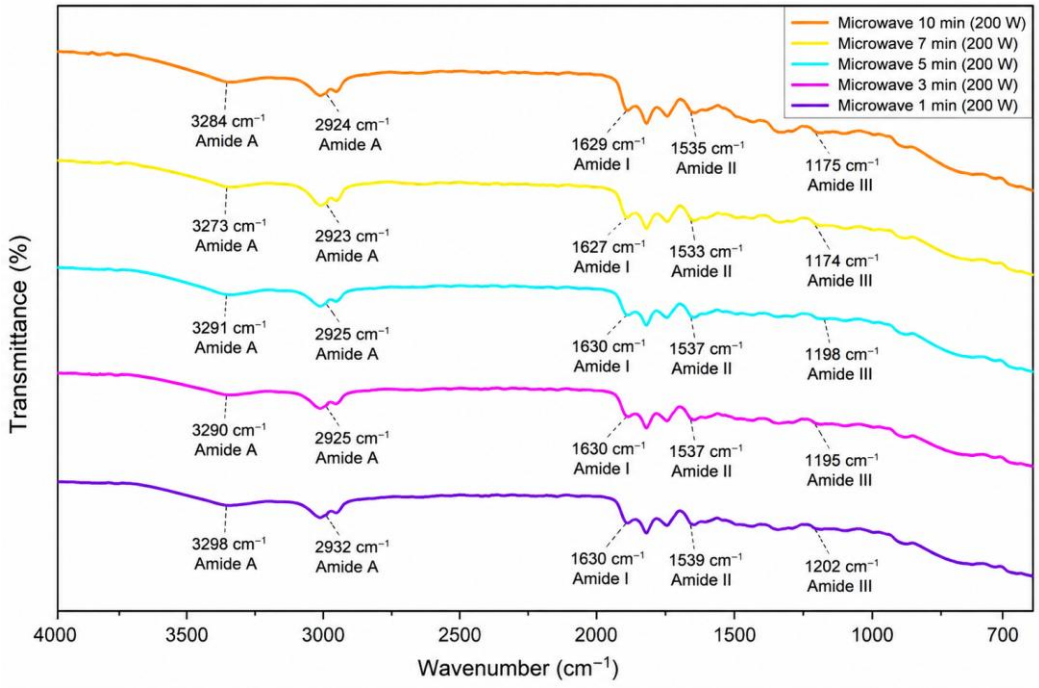

Figure 8. FTIR spectra of chicken skin gelatin extracted by microwave extraction at 200 W for 1, 3, 5, 7, and 10 minutes.

performance because those properties depend on formulation concentration, crosslinking, drying conditions, and post-processing.

3.2. Structural and morphological characterization

3.2.1. FTIR analysis

Fourier Transform Infrared (FTIR) spectroscopy was employed to evaluate whether microwave-assisted extraction altered the chemical structure of the extracted gelatin. As

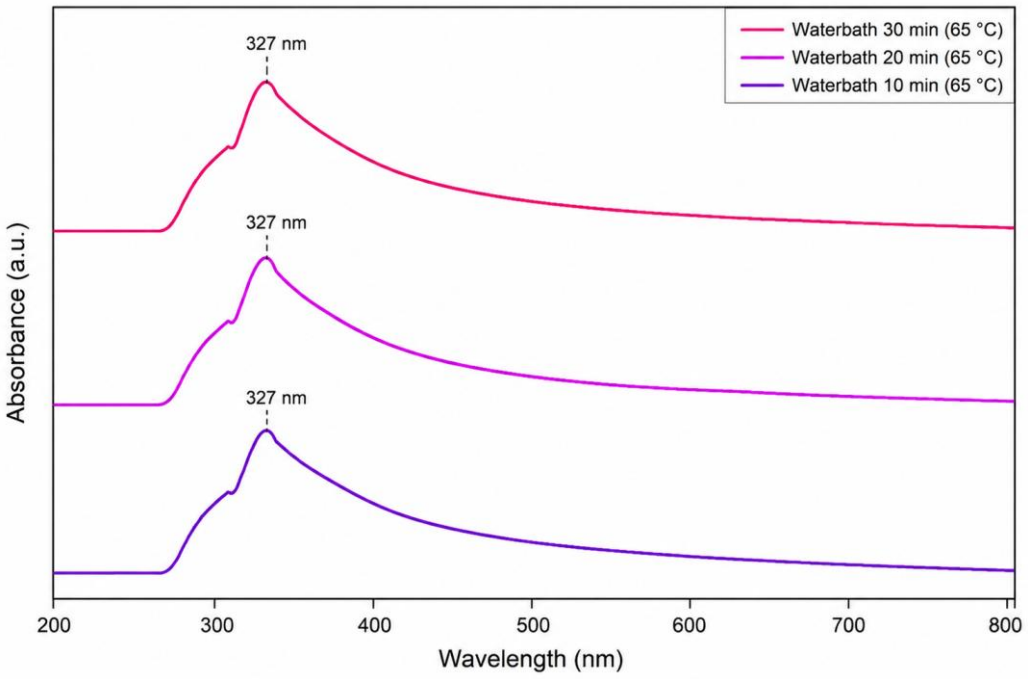

Figure 9. UV-Vis spectra of chicken skin gelatin extracted by waterbath extraction at 65°C for 10, 20, and 30 minutes.

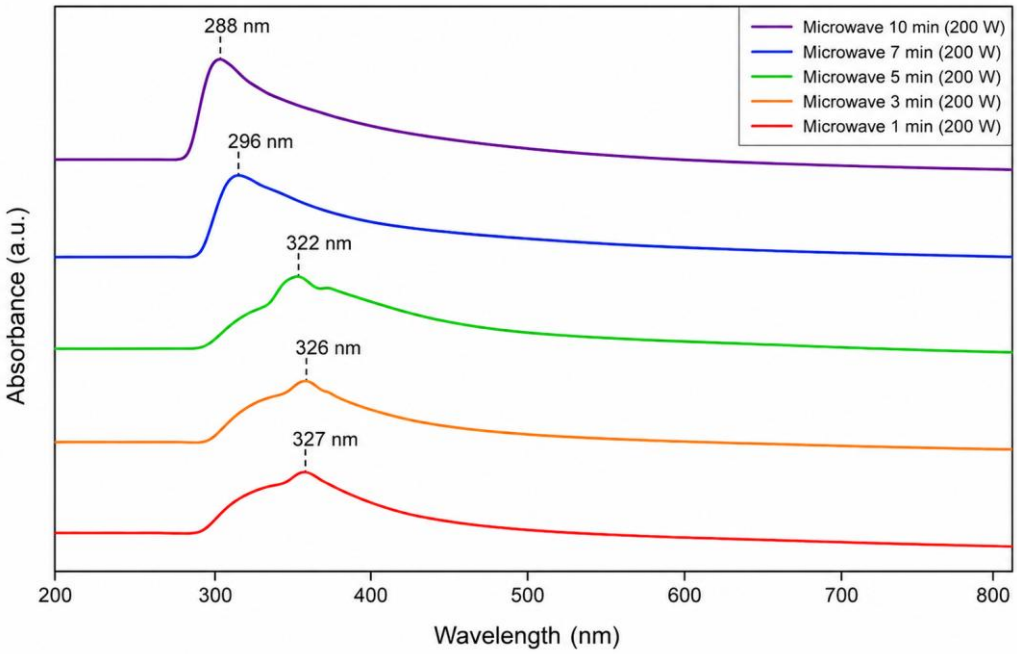

Figure 10. UV-Vis absorption spectra of chicken skin gelatin extracted by microwave extraction at 200 W for 1, 3, 5, 7, and 10 minutes.

shown in Figure 7 and Figure 8, both waterbath- and microwave-extracted gelatin exhibited the characteristic absorption bands of gelatin in the Amide A, Amide I, Amide II, and Amide III regions, confirming the presence of the principal protein functional groups associated with collagen-derived gelatin.

The preservation of these characteristic amide regions indicates that the fundamental chemical functionality of gelatin was retained under both extraction conditions. The absence of an obvious new dominant absorption region also suggests that microwave irradia-

tion under the applied conditions did not result in extensive formation of chemically different functional groups.

Nevertheless, differences in peak intensity and position between extraction treatments indicate that preservation of chemical functionality did not necessarily mean preservation of molecular organization. Microwave heating can promote rapid disruption of collagen's ordered structure and subsequent reorganization of the resulting gelatin chains. Changes in hydrogen bonding, chain conformation, and intermolecular interactions can therefore occur without generating entirely new chemical functional groups.

This interpretation is consistent with the physicochemical results. The optimum MAE condition simultaneously produced the highest yield and viscosity while retaining the characteristic gelatin amide bands. Thus, the improved extraction performance appears to result primarily from enhanced collagen disassembly and solubilization rather than extensive chemical modification of the protein backbone.

The FTIR results should also be interpreted together with the SDS–PAGE and SEM observations. FTIR provides evidence regarding functional groups, whereas SDS–PAGE provides information about peptide-chain distribution and SEM reveals the resulting dried morphology. Their combined interpretation therefore provides stronger evidence for structural modification than FTIR alone.

3.2.2. UV–Vis analysis

Ultraviolet–Visible (UV–Vis) spectroscopy was performed to evaluate the effect of the extraction method on the electronic absorption characteristics of the extracted gelatin. As shown in Figure 9 and Figure 10, the UV–Vis spectra of both waterbath- and microwave-extracted gelatin exhibited absorption peaks within the range of 288–327 nm. These absorption bands are commonly associated with chromophoric groups originating from aromatic amino acid residues and carbonyl-containing structures present in gelatin.

The microwave-extracted samples showed slight shifts toward lower wavelengths relative to the waterbath samples. Because the overall spectral profiles remained comparable, these shifts are more appropriately interpreted as changes in the local electronic environment and molecular conformation rather than evidence of extensive formation of new chemical species.

Such changes can occur when collagen chains are unfolded and reorganized during gelatin extraction. Microwave irradiation may accelerate the disruption of ordered collagen interactions and alter the exposure or local environment of chromophoric residues. Consequently, the UV–Vis results provide complementary evidence to the FTIR analysis: the principal chemical functionalities remain characteristic of gelatin, while the local molecular organization is modified by the extraction process.

The UV–Vis findings should nevertheless be interpreted as qualitative structural evidence. The present measurements do not permit direct determination of molecular weight, degree of hydrolysis, or specific conformational populations. These properties would require additional quantitative analyses such as molecular-weight determination, circular dichroism, or thermal analysis.

3.2.3. SDS-PAGE analysis

Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE) was employed to evaluate the molecular weight distribution of proteins present in the extracted chicken skin gelatin. As shown in Figure 11, the electrophoretic profile exhibited a broad distribution of protein fragments with diffuse bands extending across a wide molecular weight range, indicating that the extracted gelatin consisted of heterogeneous peptide populations.

The predominance of smeared bands, rather than distinct protein bands, suggests that collagen was converted into peptide fragments with varying molecular weights during the extraction process. Such heterogeneous molecular distributions are characteristic of gelatin produced through partial collagen hydrolysis, where the native triple-helical collagen structure is disrupted into a mixture of polypeptide chains and smaller peptide fragments. The absence of prominent high-molecular-weight bands further indicates that extensive preservation of intact collagen molecules was unlikely under the applied extraction conditions.

The SDS–PAGE result complements the FTIR analysis. FTIR demonstrated that the characteristic protein functional groups were retained, whereas SDS–PAGE indicates that the organization of those protein chains was substantially altered. In other words, the extraction process appears to modify the molecular architecture of collagen without eliminating its fundamental protein chemistry.

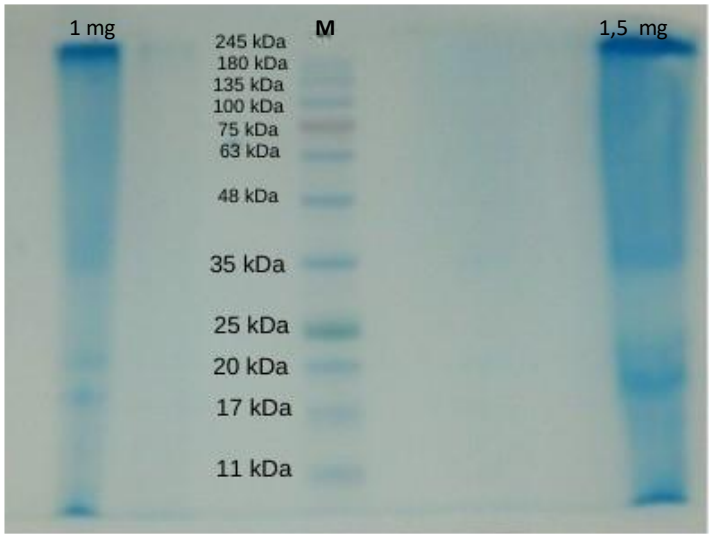

Figure 11. SDS–PAGE profile of chicken skin gelatin extracted under the optimum microwave condition (GM200-10). Lane M: molecular-weight marker; lanes 1 and 2: gelatin samples.

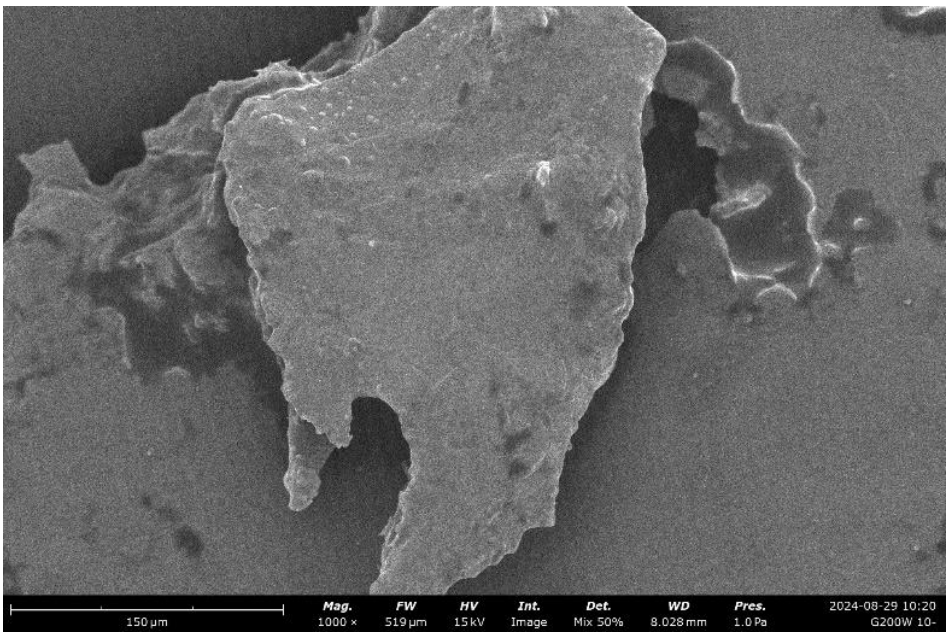

Figure 12. SEM micrograph of chicken skin gelatin extracted under the optimum microwave condition (GM200-10) at 1000× magnification and 15 kV accelerating voltage.

The heterogeneous molecular distribution may also contribute to the viscosity behavior observed in Section 3.1.5. However, because the present study did not quantitatively determine molecular-weight averages or molecular-weight distribution, a direct quantitative relationship between individual electrophoretic bands and viscosity cannot be established. The SDS–PAGE result should therefore be regarded as qualitative structural evidence supporting the process–property interpretation.

3.2.4. SEM analysis

Scanning Electron Microscopy (SEM) was employed to examine the surface morphology of the optimum microwave-extracted chicken skin gelatin (GM200-10). As shown in Figure 12, the gelatin exhibited a dense, compact, and irregular surface morphology with regions of heterogeneous texture. In addition, several pores and microcracks were observed throughout the dried gelatin matrix, indicating that the material possessed a non-uniform microstructure.

The observed morphology is consistent with the heterogeneous molecular distribution identified by the SDS-PAGE analysis. During microwave-assisted extraction, collagen is converted into gelatin molecules with varying chain lengths, which subsequently reorganize

during solvent removal and drying. As water evaporates, these molecules interact through hydrogen bonding and other intermolecular forces, producing a compact yet heterogeneous matrix. The irregular surface texture therefore likely reflects differences in molecular organization that developed during the extraction and drying processes rather than a completely uniform polymer network.

The presence of pores and microcracks may be associated with localized moisture evaporation and shrinkage during drying, phenomena that have also been reported for gelatin and other protein-based biopolymers [24]. Such microstructural features are commonly observed in dehydrated gelatin matrices and reflect the combined influence of molecular organization, solvent removal, and drying conditions. When considered together with the FTIR, UV–Vis, and SDS-PAGE results, the SEM observations provide complementary evidence that microwave-assisted extraction primarily affected the structural organization of gelatin without fundamentally altering its characteristic chemical functional groups [25].

The SEM morphology also provides an important connection between molecular and macroscopic observations. The heterogeneous peptide distribution indicated by SDS–PAGE is consistent with the non-uniform surface morphology observed by SEM, while the retained amide functionalities identified by FTIR indicate that these structural changes occurred without complete loss of the characteristic gelatin chemistry.

3.3. Process–structure–property relationship

The results demonstrate a consistent relationship between extraction conditions, molecular organization, and physicochemical properties of chicken skin gelatin. Under the investigated conditions, MAE increased the gelatin yield from 11.18% to 21.19% as extraction time increased from 1 to 10 min, while viscosity increased from 1.63 to 3.25 cP. At the same time, moisture content remained within 13.07–15.92%, ash content decreased to 3.22%, and pH remained approximately 4.0. These results indicate that the increase in microwave treatment time enhanced gelatin recovery and solution viscosity without causing a substantial deterioration in the basic physicochemical characteristics measured in this study.

The structural analyses provide a mechanistic basis for this behavior. FTIR confirmed retention of the characteristic amide functionalities, indicating that the principal chemical identity of gelatin was preserved. UV–Vis spectroscopy nevertheless indicated subtle changes in the electronic environment of the protein chromophores, suggesting molecular reorganization. SDS–PAGE further demonstrated heterogeneous peptide-chain distributions, while SEM showed a dense and irregular dried morphology. These observations collectively indicate that microwave treatment primarily promoted collagen disassembly and molecular reorganization rather than extensive chemical transformation.

The process–structure relationship can therefore be described across several scales. At the process scale, rapid volumetric microwave heating accelerates energy transfer and collagen disruption. At the molecular scale, this results in gelatin chains with heterogeneous molecular sizes and altered conformational organization. At the macroscopic scale, these molecular changes are reflected in the increased viscosity and the morphology of the dried gelatin matrix. This interpretation is consistent with previous microwave extraction studies in which processing conditions affected both gelatin recovery and structural characteristics [16], [20].

Importantly, the present results demonstrate why yield alone is insufficient for evaluating an extraction process. The optimum condition was not selected merely because it produced the highest recovery; it also produced the highest viscosity while maintaining characteristic gelatin functional groups and acceptable moisture and ash characteristics. Thus, the 21.19% yield at 200 W for 10 min represents a processing condition in which extraction efficiency and the measured material descriptors were simultaneously favorable.

Nevertheless, the process–structure–property relationship established here remains preliminary. Molecular weight was evaluated qualitatively by SDS–PAGE rather than quantitatively, and application-specific properties such as Bloom gel strength, tensile strength, elongation, water-vapor permeability, swelling, degradation, and biocompatibility were not measured. Therefore, the present results establish a material precursor profile, rather than demonstrating final engineering performance.

3.4. Engineering application feasibility

The physicochemical and structural characteristics obtained in this study indicate that the extracted gelatin can be considered a potential precursor for gelatin-based engineered materials, particularly biodegradable films, coatings, hydrogels, and composite biomateri-

als. The optimum MAE condition produced a yield of 21.19%, moisture content within 13.07–15.92%, minimum ash content of 3.22%, pH of approximately 4.0, and viscosity of 3.25 cP. The retention of characteristic amide functionalities and the observed molecular and microstructural characteristics further establish a useful baseline for subsequent material formulation.

For biodegradable packaging, gelatin is attractive because it can form continuous films and coatings. However, gelatin is intrinsically hydrophilic, and therefore its moisture-barrier and mechanical performance generally require improvement through crosslinking, blending, or reinforcement. Recent experimental work demonstrated that incorporation of bacterial cellulose and transglutaminase crosslinking increased the tensile strength of gelatin films by 42.79% and reduced water-vapor and oxygen permeability by 19.33% and 40.15%, respectively [13]. This comparison indicates that the gelatin obtained in the present study could reasonably be considered a film-forming precursor, but the present yield, moisture, pH, and viscosity data cannot be directly interpreted as film tensile or barrier performance.

For biomaterial applications, gelatin can similarly function as a matrix or precursor in porous scaffolds and hydrogels. Gelatin/$Na_2Ti_3O_7$ nanocomposite scaffolds, for example, have been reported with 67–85% porosity, and an optimized gelatin:titanate composition exhibited a compressive modulus of 30.57 kPa [2]. These values illustrate the type of application-specific engineering properties that can be achieved after gelatin is incorporated into a designed material system. They should not, however, be compared directly with the present gelatin because the reported compressive modulus belongs to a fabricated gelatin/titanate scaffold rather than the extracted gelatin itself.

The present study therefore provides an upstream material-processing contribution rather than a complete application-performance assessment. The 3.25 cP viscosity indicates that the optimum gelatin solution has a flow behavior comparable to values reported for poultry gelatin, while the characteristic FTIR bands confirm preservation of the protein functionalities required for subsequent chemical or physical modification. Recent chicken-feet gelatin studies reported viscosities of 3.85–4.43 cP at 6.67% gelatin concentration, together with moisture below 15%, demonstrating that the present chicken-skin gelatin has broadly comparable solution characteristics despite differences in raw material and extraction method [9].

From a processing-efficiency standpoint, the principal advantage of the present MAE approach is the combination of relatively high recovery and short processing time. The yield of 21.19% after 10 min at 200 W compares favorably with the 10.48% yield obtained after 20 min at 540 W for microwave-extracted chicken-feet gelatin [6]. The comparison suggests that the present chicken-skin system achieved substantial gelatin recovery at a lower microwave power and shorter treatment time, although direct energy-efficiency comparisons cannot be made because the two studies used different raw materials, pretreatment proce-

Table 2. Quantitative comparison of selected gelatin extraction studies and the present study.

Raw material	Extraction method	Processing condition	Yield (%)	Moisture (%)	pH	Viscosity (cP)	Selected property	Ref.
Duck skin	Microwave	200 W, 10 min	28.51	—	—	—	Higher microwave yield	[16]
Rabbit skin	Microwave	500 W, 60 min	6.44	—	—	—	Microwave extraction; structural characterization	[20]
Chicken feet	Microwave	540 W, 20 min	10.48	—	—	—	Gel strength 253 g	[6]
Broiler chicken feet	Conventional	—	7.93	<15	5.31	3.85	High Bloom strength; 203 g	[9]
Chicken skin	Microwave	200 W, 10 min	21.19	13.07–15.92	≈4.0	3.25	Ash 3.22%; characteristic gelatin amide bands	Present study

Note**:** Values are reported as published in the respective studies. Direct comparison should be interpreted cautiously because raw materials, pretreatment procedures, solid-to-liquid ratios, extraction systems, drying conditions, and calculation methods differed among studies. Data for the present study are derived from the experimental results reported in this manuscript.

dures, sample masses, and microwave systems.

A comparison with selected literature is summarized in Table 2. The table is intentionally restricted to parameters that are directly comparable across studies and does not treat film or scaffold properties as properties of the extracted gelatin itself.

The comparison demonstrates that the present study occupies a useful position between extraction-efficiency studies and material-characterization studies. The yield of 21.19% is lower than the 28.51% reported for duck skin but substantially higher than the 6.44% and 10.48% reported for rabbit skin and chicken feet, respectively. More importantly, the present study combines yield evaluation with moisture, ash, pH, viscosity, FTIR, UV–Vis, SDS–PAGE, and SEM characterization, allowing the extraction process to be discussed in terms of both recovery and resulting material structure.

Accordingly, the extracted gelatin should be regarded as a potential engineering-material precursor, rather than as a finished engineering material. Before application in packaging, coatings, hydrogels, or biomaterial scaffolds, further processing and characterization are required, including film tensile strength, elongation at break, water-vapor permeability, oxygen permeability, swelling, degradation, gel strength, thermal stability, and, for biomedical applications, cytocompatibility and biological response. This limitation does not diminish the process contribution of the present work; rather, it defines the next stage of development from optimized extraction toward application-specific material engineering.

4. Conclusions

This study investigated the effect of microwave-assisted extraction (MAE) on the physicochemical, molecular, and morphological characteristics of gelatin extracted from chicken skin and compared its performance with conventional waterbath extraction. Chicken skin was extracted by microwave irradiation at 200 W for 1–10 min and by waterbath heating at 65 °C for 10–30 min, followed by evaluation of yield, moisture content, ash content, pH, viscosity, FTIR, UV–Vis, SDS–PAGE, and SEM characteristics. Under the investigated conditions, MAE achieved the highest gelatin yield of 21.19% after 10 min, compared with 12.39% for waterbath extraction, while the viscosity increased to 3.25 cP; the extracted gelatin exhibited 13.07–15.92% moisture, a minimum ash content of 3.22%, and a pH of approximately 4.0. FTIR confirmed preservation of the characteristic gelatin amide groups, whereas UV–Vis and SDS–PAGE indicated changes in molecular organization and heterogeneous peptide distribution, and SEM revealed a dense, irregular microstructure with pores and microcracks. Collectively, these findings demonstrate that microwave-assisted extraction can substantially improve gelatin recovery within a short processing time while maintaining the principal chemical characteristics and acceptable physicochemical properties of the extracted gelatin, establishing a clear process–structure–property relationship. The resulting gelatin therefore represents a promising precursor for further development of gelatin-based films, coatings, hydrogels, and composite biomaterials, although application-specific properties such as gel strength, mechanical performance, barrier properties, thermal stability, and biocompatibility require further investigation before specific engineering applications can be established.

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Declaration of Generative AI and AI-Assisted Technologies in the Writing Process

The authors used ChatGPT (OpenAI) to assist in improving the English language, refining the organization and clarity of the manuscript, and supporting the revision of selected sections in response to editorial and reviewer comments. All AI-assisted content was critically reviewed, revised, and validated by the authors against the original experimental data and relevant scientific literature. The authors take full responsibility for the scientific accuracy, originality, and integrity of the manuscript, in accordance with the guidelines of the Committee on Publication Ethics (COPE).

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