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VALORIZATION OF SMOKED TROUT SKINS AS A GELATIN SOURCE: ANALYSIS OF TECHNOLOGICAL AND STRUCTURAL PROPERTIES

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ABSTRACT
Fish processing by-products are increasingly used as alternative sources of functional biopolymers such as gelatin. This study aimed to evaluate the potential of smoked trout skins for gelatin production and to compare their properties with non-smoked trout gelatin. Gelatin was obtained by extraction from smoked trout skins. Technological and structural properties of smoked trout gelatin were analyzed and compared with non-smoked trout gelatin. Gelatin yield (9.89%) and gel strength (45 g) obtained from smoked skins were lower than those from non-smoked trout gelatin. Despite this, the emulsion activity index is higher compared to non-smoked trout gelatin, while the foam capacity and protein content remain at a similar level. The color difference obtained was quite significant, with a ΔE of 18.93 (very pronounced). Spectroscopic analysis confirmed that the functional groups were preserved. Smoked trout skin offers a good opportunity for food and other sectors as a sustainable and alternative gelatin source.
Keywords: Smoked trout skin, fish gelatin, functional properties, emulsifying activity, fish by-products

TÜTSÜLENMİŞ ALABALIK DERİLERİNİN JELATİN KAYNAĞI OLARAK DEĞERLENDİRİLMESİ: TEKNOLOJİK VE YAPISAL ÖZELLİKLERİN ANALİZİ

ÖZ
Balık işleme yan ürünleri, jelatin gibi fonksiyonel biyopolimerlerin alternatif kaynağı olarak giderek daha fazla kullanılmaktadır. Bu çalışma, tütsülenmiş alabalık derilerinin jelatin üretimi için potansiyelini değerlendirmeyi ve özelliklerini tütsülenmemiş alabalık jelatini ile karşılaştırmayı amaçlamaktadır. Jelatin, tütsülenmiş alabalık derilerinden ekstraksiyon ile elde edilmiştir. Teknolojik ve yapısal özellikler analiz edilmiş ve tütsülenmemiş alabalık jelatini ile karşılaştırılmıştır. Tütsülenmiş derilerden elde edilen jelatin verimi (%9.89) ve jel kuvveti (45 g), gökkuşağı alabalığı jelatininden daha düşüktür. Buna rağmen, emülsiyon aktivite indeksi daha yüksek, köpük kapasitesi ve protein içeriği ise yakın bir seviyededir. Renk farklılığı ise oldukça belirgin (ΔE:18.93 - çok belirgin) olarak elde edilmiştir. Spektroskopik analizler, fonksiyonel grupların korunduğunu doğrulamıştır. Tütsülenmiş alabalık derisi, sürdürülebilir ve alternatif yeni bir jelatin kaynağı olarak gıda ve diğer sektörler için iyi bir fırsat sunmaktadır.
Anahtar kelimeler: Tütsülenmiş alabalık derisi, balık jelatini, fonksiyonel özellikler, emülsiyon aktivitesi, balık yan ürünleri

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INTRODUCTION

The increasing global demand for sustainable and value-added biomaterials has driven interest in the utilization of fish processing by-products, particularly for gelatin extraction (Joy et al., 2024). Gelatin, a water-soluble protein derived from the partial hydrolysis of collagen, is widely used in the food, pharmaceutical, and cosmetic industries due to its gelling, emulsifying, and stabilizing properties (Karim and Bhat, 2009). While mammalian sources such as bovine and porcine skins have traditionally dominated the gelatin market, concerns regarding religious restrictions, zoonotic disease transmission, and environmental impact have promoted the exploration of marine-derived alternatives (Cebi et al., 2016). Among marine resources, fish skins are considered a viable source for gelatin production owing to their high collagen content and low commercial value (Joy et al., 2024).

Smoking, which uses fire, is one of the oldest preservation methods which includes salting, drying, heating, and smoking processes (Adeyeye, 2019). The smoking process is defined as the penetration of volatile compounds, produced by burning wood, into the surface of meat or fish products. Fish smoking is done in two ways: cold (28–32°C) and hot (70–80°C) smoking (Alasalvar et al., 2011). In cold smoking, the product is not cooked, the proteins do not coagulate, and enzymes and pathogens are not completely inactivated; therefore, it should be kept cold. In hot smoking, these processes take place, and the protective feature is provided (Alasalvar et al., 2011). It is stated that today 40-60% of total meat products and 15% of fish are smoked (Adeyeye, 2019).

Smoked trout is an important luxury food product preferred, especially in European countries, due to its nutritional content and aroma. During the processing of smoked trout, which is produced by applying a maximum temperature of 75°C in Türkiye, a significant amount of fish skin is generated as waste. Kołodziejska et al. (2008) and Zhang and Regenstein (2017) produced gelatin from cold-smoked (max 30°C) salmon skins, with high efficiency of 25% and 22.4%, respectively. With the increase in the processing temperature, the hydrogen bonds in the gelatin molecules are broken by the absorbed energy, which causes the gelatin structure to go into an unregulated state and the protein molecules to open (Ruan et al., 2023). However, it is estimated that the collagen present in the skin during the smoking process is not hydrolyzed into gelatin (Zhang and Regenstein, 2017).

Rainbow trout is a valuable cold-water fish species and was the most produced fish species in Türkiye with 222.486 tons in 2023 (TUIK, 2024). There are studies on the evaluation of rainbow trout waste (Fan et al., 2018; Rajabzadeh et al., 2018; Tabarestani et al., 2010; Üçyol, 2016; Yaghoubzadeh et al., 2020). However, limited research has focused on the use of smoked fish by-products, which may be underutilized despite their abundance in the seafood industry (Kołodziejska et al., 2008; Zhang and Regenstein, 2017). In particular, no study has been found on smoked trout skin gelatin.

Therefore, this study aimed to extract gelatin from smoked trout skins and evaluate its technological (emulsion, foaming, oil binding, color characteristics) and structural properties (FTIR, UV-VIS) in comparison to gelatin obtained from non-smoked trout skin. The findings of this research provide insights into the feasibility of using smoked fish waste as a sustainable gelatin source and contribute to the valorization of seafood processing residues.

MATERIALS AND METHODS

Materials

All reagents and solvents used in the experiments were of analytical purity and used without further purification.

Preparation of fish skins

Smoked trout (*Oncorhynchus mykiss*) skins were procured from Gümüşdoğa Su Ürünleri A.Ş. Fish skins, generated as by-products during the filleting process, were transported to the laboratory under a maintained cold chain. Upon arrival, the skins were rinsed with chilled tap water to remove surface impurities, and the excess water

was drained. Subsequently, the cleaned materials were manually chopped into uniform fragments measuring approximately 0.5 x 0.5 cm. These skins, which are used in the production of gelatin, were kept in the refrigerator (4°C) until the extraction stage.

Methods

Production of smoked trout gelatin

The production of gelatin from fish skins was studied by Zhang and Regenstein (2017). Initially, the fish skins were subjected to an alkaline pretreatment using a 0.01 N NaOH solution at a ratio of 5:1 (v/w) for 90 min. at 4°C. This step was followed by acid treatment with 0.1 N HCl under an identical solvent-to-sample ratio for 40 min. at 4°C. Between each chemical treatment, the samples were rinsed thoroughly with distilled water (5:1 v/w) at ambient temperature. Subsequently, gelatin extraction was conducted in a water bath (Memmert WNB45, Schwabach, Germany) using hot distilled water at 70°C for 4 hours at a 3:1 v/w ratio. Post-extraction, the solid residues were separated by filtration through cheesecloth (4-layered). The resulting gelatin solutions were centrifuged at 7000 xg (50 mL * 6-fixed angle rotor) for 15 min. to remove remaining particulates. The clarified supernatants were then transferred into glass containers and dried in a convection oven (Binder GmbH ED240, Germany) at 60°C for approximately 72 hours. The final gelatin product, obtained as thin sheets, was stored in polyethylene packaging under dry conditions until further analysis. Non-smoked trout gelatin, on the other hand, was described by Tekle (2022).

Gelatin yield (%) = Gelatin weight(g) / skin weight(g) × 100 (1)

Gel strength

A gelatin solution was prepared at a concentration of 6.67% by dissolving it in a 60°C water bath for 30 min. The solutions taken in bloom jars were matured by keeping them at 4°C for 12 hours. In the measurement made with a 5 kg load cell and a 1.27 cm cylindrical probe in the Texture Analyzer, the highest force is given as the Bloom value in g (Tekle, 2016).

Protein and biochemical composition

The protein ratio of gelatins was determined using the Kjeldahl method. A 1 g sample was placed in each Kjeldahl tube, and 12 mL of sulfuric acid (H_2SO_4) and 1 Kjeldahl tablet were added to it. In the combustion unit, the burning process was carried out for about 4 hours until the contents of the tube became clear. After the procedure, 75 mL of pure water was added to the cooled tubes and the distillation stage was started. 75 mL of 33% NaOH solution was automatically added to the tubes from the distillation unit. The conical flask with 25 mL of 4% boric acid solution was placed at the other end of the unit. As a result of the distillation, approximately 150 mL of distillate was collected. The process was completed, and the resulting distillate was titrated with 0.1 N HCl. The amount of crude nitrogen is calculated with the following formula. The amount of nitrogen obtained was calculated using a 5.4 conversion factor; the total amount of protein was then calculated (Yıldız, 2017). Moisture and ash levels of fish skin and gelatin were determined according to AOAC methods (AOAC, 1990).

Total Nitrogen (%) =[(A–B) ×N×0.014/Sample size (g)] ×100 (2)

A: 0.1 N HCl (mL) spent in titration
B: 0.1 N HCl (mL) spent on the witness trial
N: Normality of HCl (0.1 N)

Oil binding capacity (OBC)

The oil binding capacity of smoked trout gelatins was determined by modifying the method specified by Karoud et al. (2019). Approximately 50 mg of sample was accurately weighed into pre-tared centrifuge tubes. Subsequently, 1 mL of sunflower oil was added to each tube, and the mixtures were incubated at room temperature for 1 hour. During this period, the contents were vortexed for 5 seconds at 15-minute intervals using a Velp ZX3 Vortex Mixer (Italy) to ensure uniform dispersion. Following incubation, samples were centrifuged at 7000 xg for 15 min. using a benchtop centrifuge (Andreas Hettich GmbH & Co. KG, Tuttlingen, Germany). The supernatant oil layer was carefully decanted. As a control, only sunflower oil was added to an empty

centrifuge tube under identical conditions. Oil binding capacity is expressed in mL of oil/g protein.

Color Properties

Color parameters including lightness (L*), redness (a*), and yellowness (b*) of the smoked trout gelatin samples were determined using a chroma meter (CR-100, Konica Minolta, Tokyo, Japan). L* measures lightness (white: 100, black: 0), a* measures redness (red: +, green: -) and b* measures yellowness (yellow: +, blue: -). The following equations were used to calculate the Hue, Chroma, and color differences (ΔE*) (Tekle et al., 2024):

$$\Delta E^* = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2} \quad (3)$$

$$\text{Hue} = \tan^{-1} (b^*/a^*)$$

$$\text{Chroma} = [(a^*)^2 + (b^*)^2]^{1/2}$$

Here, ΔL*, Δa*, and Δb* are the differences in L*, a*, and b*, between the two gelatin samples, respectively. L*, a*, and b* values of each sample were measured three times.

FTIR spectroscopy analysis

ATR-FTIR samples of gelatins in powder form were read at a resolution level of 4 cm^{-1} and 16 scans in each spectrum (Bruker Tensor 27 FTIR spectrometer, Ettlingen, Almanya). The recording of the spectra was performed in the mid-infrared region of 4000-600 cm^{-1}. Measurements were made under the same conditions for each sample, and then the three spectra were averaged. Prior to each measurement, the crystal surface was thoroughly cleansed using distilled water followed by absolute ethanol to ensure the removal of any contaminants (Cebi et al., 2016).

UV-VIS absorption spectra

The UV spectra of the gelatin samples were recorded using an UV-VIS spectrophotometer (Thermo Scientific GENESYS 10S UV-VIS, USA). Samples dissolved in distilled water (1 mg/mL) and plain distilled water were used as references. The range of the absorbance curve is between 190 and 400 nm, and the resolution is set at 0.5 nm (Xu et al., 2017).

Emulsion activity and stability

The emulsion activity index (EAI) and stability index (ESI) were determined by Zamorano-Apodaca et al. (2020) with slight modification of the method. 9 mL of gelatin solution (0.1% p/v) was taken and 3 mL of sunflower oil was added to them. The mixture was homogenized at 18000 xg for 1 min using an Ultra-turrax (Daihan HG-15D, Seoul, Korea) device. Immediately and 10 min. after homogenization, 50 µL of emulsion taken from the bottom was diluted to 5 mL with 0.1% dodecyl sulfate sodium salt (SDS). The absorbance of the diluted solutions was read using a spectrophotometer (Thermo Scientific GENESYS 10S UV-Vis, USA) at a wavelength of 500 nm. Absorbance readings (A_0 and A_{10}) were used to calculate EAI and ESI values in the following equations:

$$\text{EAI } (m^2/g) = (2 \times 2.303 \times A_0) / (C \times \varphi) \quad (4)$$

A_0 = Absorbance at a wavelength of 500 nm
C = Initial concentration of protein (g/mL)
φ = The volume of oil in the emulsion

$$\text{ESI (dk.)} = (A_0 \times \Delta t) / \Delta A \quad (5)$$

A_{10}= Absorbance at the end of 10 min.
Δt = 10 min.
$\Delta A = A_0 - A_{10}$

Foam capacity and stability

Foam capacity and foam stability were analyzed Zamorano-Apodaca et al. (2020). 10 mL of gelatin solutions (0.5% p/v) was taken and homogenized at 14000 x g at room temperature for 1 min using the Ultra-turrax (Daihan HG-15D, Seoul, Korea) device. The total foam volume was measured as a result of homogenization after 0 and 10 min. Foam capacity refers to the foam expansion that occurs at the initial time, while foam stability refers to the foam expansion that occurs at the 10th minute. Foam expansion is calculated by the following equation:

$$\text{Foam expansion (\%)} = [(A - B) / B] \times 100 \quad (6)$$

A: Volume at different times (mL)
B: Volume before homogenization (mL)

Statistical analysis

The analyses were performed in three repetitions. The results were evaluated by analysis of variance (ANOVA) using JMP Pro 18 (SAS, NC, USA) program, and the Tukey test.

RESULTS AND DISCUSSION

Yield

The yield of gelatin obtained from smoked trout (*Oncorhynchus mykiss*) skins was determined to be 9.89%. Considering that the gelatin yield obtained from fresh and non-smoked trout skins is 16%, it can be inferred that the smoking process has a negative effect on gelatin yield. Structural changes, such as denaturation and cross-linking in the collagen structure caused by heat treatment and chemical components applied during smoking, may have limited the efficiency of gelatin extraction and reduced the overall yield. Tabarestani et al. (2010) found that the gelatin yield from rainbow trout skin was 9.36%. Zhang and Regenstein (2017), on the other hand, obtained gelatin yields in the range of 2.23% to 22.4% from smoked salmon skin. The average yield of fish gelatins is in the range of 6% to 19% (Karim and Bhat, 2009). The yield values obtained in this study are within the specified range. Therefore, the use of food industry waste, such as smoked trout skins, in the production of gelatin offers an important opportunity.

Protein and biochemical composition

The results of protein and biochemical composition of skin and gelatins are given in Table 1. The protein content of gelatin obtained from smoked trout (*Oncorhynchus mykiss*) skins was determined as 82.43%. Although this value is low compared to the gelatin protein content obtained from non-smoked trout skins, which was determined as 91.29%, both gelatin samples are found to have high protein content. Üçyol (2016) found a protein ratio of 97.14% in gelatin obtained from trout skins. This value in the literature is close to the protein ratio of non-smoked trout gelatin (91.29%) but higher than smoked trout gelatin (82.43%). Since gelatin consists mainly of collagen-derived proteins, its total protein content is directly related to its purity level and functional performance (Gómez-Guillén et al., 2009). The moisture content of both gelatin samples is very close to each other (Table 1). The amount of ash in smoked trout skin (15.37%) was found to be quite high. This is thought to be due to the salting process carried out during the smoking process (Zhang and Regenstein, 2017). Considering the protein and ash ratios of smoked trout gelatin, it is seen that the purification process is successfully carried out in gelatin production.

Table 1. Biochemical composition (%) of smoked and non-smoked trout skin and gelatin

Samples	Protein	Moisture	Ash
Smoked trout skin	22.52±0.17	63.28±0.31	15.37±0.09
Smoked trout gelatin	82.43±0.21	3.28±0.22	2.36±0.70
Non-smoked trout skin	30.40±0.13	56.39±0.27	3.33±0.89
Non-smoked trout gelatin	91.29 ±0.23	3.46±0.21	1.04±0.70

The data are expressed as the mean ± standard deviation of the three measurements

Gel strength

The gel strength of gelatin obtained from smoked trout (*Oncorhynchus mykiss*) skins was determined as 45 g (Table 2). This value is significantly lower than the gel strength of 95.1 g obtained from non-smoked trout skin. Gel strength is a critical parameter that reflects the capacity of gelatin to form a three-dimensional network structure and thus its functional quality. This decrease in gel strength can be associated with the deterioration of collagen structure caused by the smoking process (Zhang and Regenstein, 2017). According to Karim and Bhat (2009), the gel strength of commercial gelatins is expressed as the bloom value and ranges from 100–300 g. Derkach et al. (2020) state that the gel strength generally does not exceed 100 grams in cold-water fish species and only around 200 grams in hot-water fish species. The gel strength values obtained in this study (95.1 g and 45 g) were lower than the

gelatins of hot water fish (grass carp: 267 g and tilapia: 224 g), but similar to the gelatins of cold water fish (salmon: 108 g and cod: 71 g) (Arnesen and Gildberg, 2007; da Trindade Alfaro et al., 2013; Kasankala et al., 2007). The gel strength change in gelatins can be explained by the differences in the imino acid content of the fish species and the gelatin extraction process (Arnesen and Gildberg, 2007). The results show that gelatin produced from smoked fish skins can have a low gel strength and therefore may be of limited use, especially for applications requiring high gel strength. However, it is a very suitable source for use in liquid protein products.

Table 2. Various properties of smoked and non-smoked trout gelatin

	Smoked trout gelatin	Non-smoked trout gelatin
Gel strength	45 g	95.1 g
OBC	1.00 ± 0.07^{g} mL/g	1.69 ± 0.19^{h} mL/g
*L**	68.35 ± 1.51^{i}	81.52 ± 0.48^{j}
*a**	12.57 ± 0.90^{j}	1.58 ± 0.25^{k}
*b**	46.96 ± 0.32^{l}	38.94 ± 0.76^{m}
Chroma	48.62 ± 0.16^{n}	38.97 ± 0.77^{o}
Hue	75.01 ± 1.11^{p}	87.67 ± 0.33^{r}
ΔE	18.93±1.18	

The data are expressed as the mean ± standard deviation of the three measurements. The results were evaluated by analysis of variance (ANOVA) and the Tukey test. Averages with different letters in the same row show a statistically significant effect (*P<0.05*), while the same letters show no statistically significant difference (*P>0.05*). OBC: Oil Binding Capacity

Oil binding capacity

The oil binding capacity of gelatins is a critical functional property in terms of flavor carrier and textural improvement, especially in food systems, and often depends on the hydrophilic–hydrophobic balance, surface properties, and structural integrity of proteins (Ranasinghe et al., 2022). In this study, the oil binding capacity (OBC) of gelatin obtained from smoked trout skins was determined as 1.00 ± 0.07 mL/g (Table 2). This value was significantly lower than the 1.69±0.19 mL/g level measured for non-smoked trout gelatin *(P<0.05)*. It has been noted that the OBC depends on the hydrophobicity of residues in gelatin, the amount of hydrophobic amino acids, and especially the tyrosine (Ranasinghe et al., 2022; Shyni et al., 2014). The smoking process is thought to lead to partial denaturation of collagen in the skin, resulting in a reduction of hydrophobic regions that provide stickiness and structural stability of the protein. This limits the ability of proteins to interact with oil droplets and leads to a decrease in oil binding capacity. Non-smoked trout gelatin, on the other hand, with its more intact protein structure, was able to interact more with lipophilic components exhibited a higher OBC value. These results suggest that the effects of pretreatment techniques such as smoking on functional properties should be carefully evaluated.

Color features

The color characteristics of gelatin samples are critical for assessing their quality and potential applications in various industries, including food and pharmaceuticals. In this study, significant color differences were observed between the gelatin sample obtained from smoked trout and non-smoked trout (Table 2, Figure 1). Smoked gelatin showed a darker appearance with a lower L* value (68.35 ± 1.51), whereas non-smoked trout gelatin showed a higher L* value (81.52 ± 0.48) in lighter tones. In terms of a* value, the high positivity observed in smoked gelatin (12.57 ± 0.90) reveals the prominence of reddish tones; while in non-smoked trout gelatin this value is quite low (1.58 ± 0.25), which indicates more neutral or slightly greenish hues. The b* value was found to be higher in the smoked sample (46.96 ± 0.32), indicating the predominance of yellow

tones. This is because heat processes, such as Maillard reactions, increase pigment formation during the smoking process (Sarabandi et al., 2022). The Chroma value, which indicates color saturation, is higher (48.62 ± 0.16) in smoked gelatin, indicating more vivid and saturated colors, while it is lower in non-smoked trout gelatin (38.97 ± 0.77). The Hue angle is also lower in smoked gelatin (75.01 ± 1.11), indicating a shift in color to a more reddish-yellow hue. On the other hand, in non-smoked trout gelatin, it indicates a more yellowish gradation of 87.67 ± 0.33. If the ΔE value is greater than 3, between 1.5 and 3, and less than 1.5, the color differences are considered as very pronounced, significant, and small differences, respectively (Tekle et al., 2024). The ΔE value, which indicates the total color difference between the two gelatins, was calculated: 18.93 ± 1.18. The value obtained in this study corresponds to a significant difference between gelatin samples according to the CIE color system (Table 2). It has been noted that gelatin colors are affected by lipid oxidation, pigment degradation, and browning reactions (Sarabandi et al., 2022).

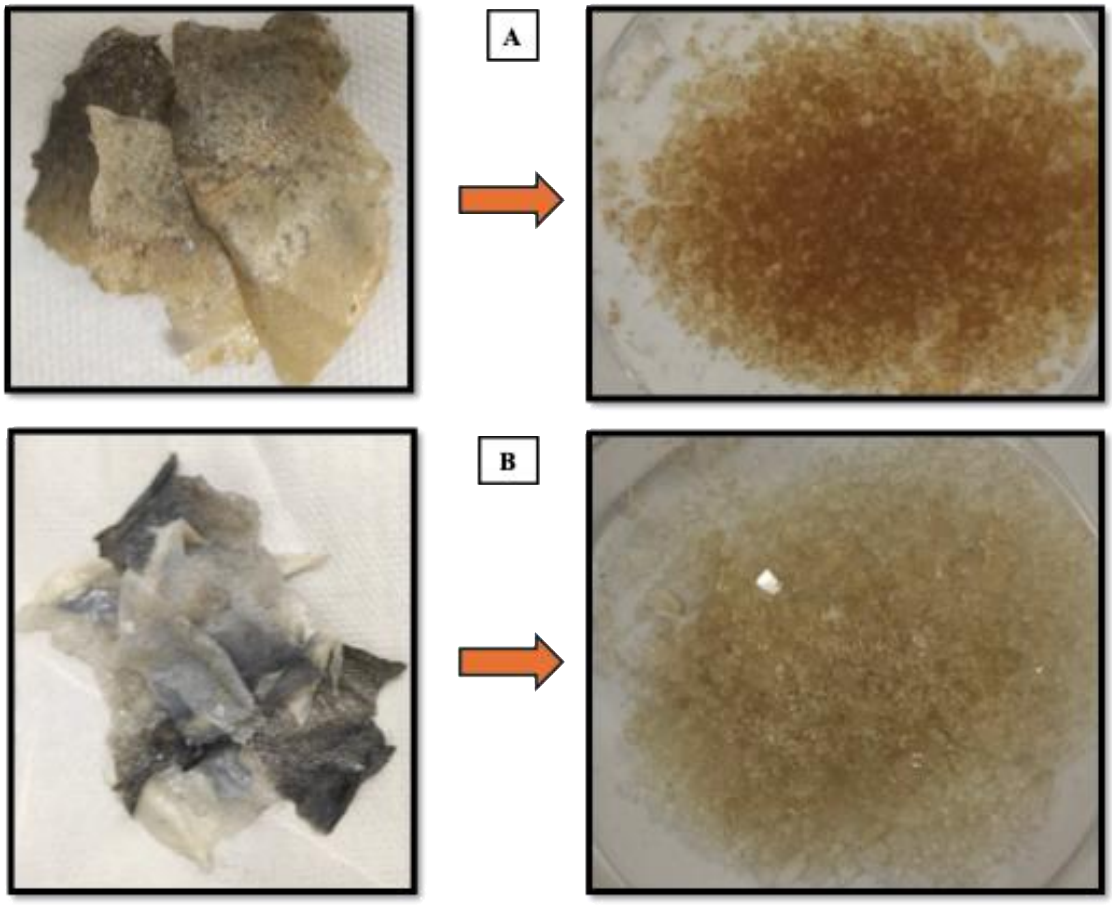

Figure 1. A: smoked trout skin and gelatin, B: non-smoked trout skin and gelatin

FTIR spectroscopy analysis

The FTIR spectra and amide I, II, III, A, and B regions of each gelatin sample are given in Figure 2 and Table 3. FTIR spectrum analyses revealed characteristic bands representing the typical protein structure for both smoked trout skin gelatin and non-smoked trout gelatin. In both samples, the Amide A band was observed at ~3277 cm^{-1}, which corresponds to N-H stretching vibrations and is an indicator of the

structure of hydrogen bonds in proteins. The Amide B bands are similarly in the range of 2932-2934 cm^{-1}, representing CH_2 asymmetric stress vibrations (Muyonga et al., 2004). The Amide I band was recorded as 1629.9 cm^{-1} (smoked) and 1630.57 cm^{-1} (non-smoked), which corresponds to C=O stretching vibrations and provides information about protein secondary structures, especially β-sheet and α-helix structures. Similarly, the Amide II (1523 cm^{-1}) and Amide III (1236-1237 cm^{-1}) bands also refer to N–H bending and C–N stretch vibrations, respectively (Cebi et al., 2016). Similar values in these regions indicate that the smoking process does not lead to any significant chemical changes in the protein structure. In the fingerprint region (1078-1079 cm^{-1}), small vibrations were observed, usually associated with C-O, C-C, and C-N stresses. The spectral similarity of both gelatins suggests that the collagen structure is largely preserved after smoking, but small frequency shifts may show minimal changes at some secondary structure levels. In all spectrum and band regions (Amide A, B, I, II, and III), smoked trout gelatin appears to have higher absorption values (Figure 2). This may be because the gelatin is exposed to greater hydrolysis.

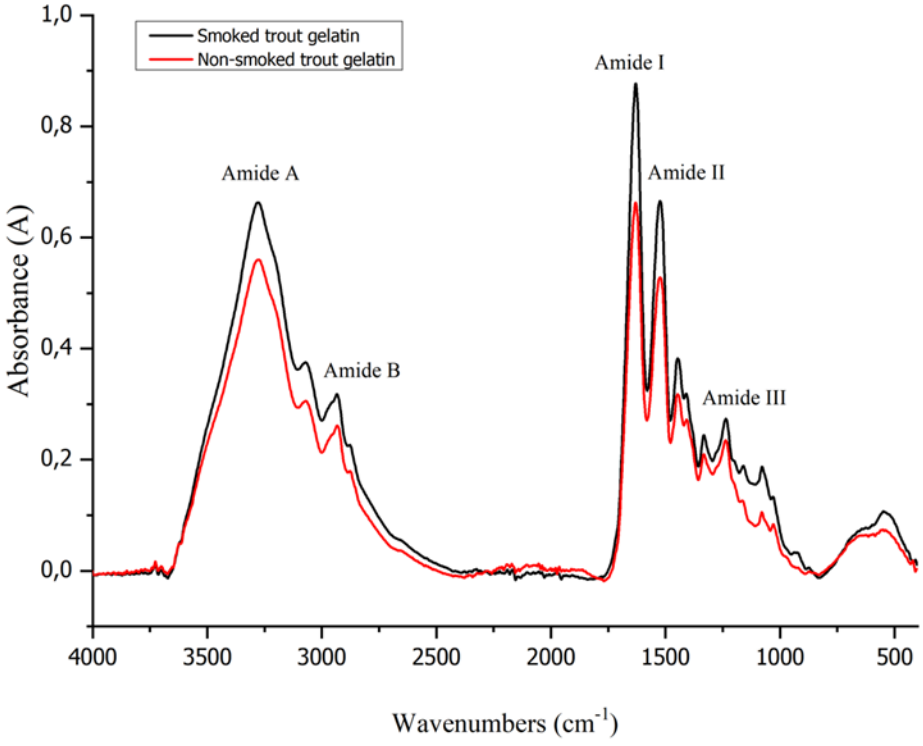

Figure 2. Smoked and non-smoked trout gelatin FTIR spectra and amide regions

Table 3. Location and assignment of the peaks identified in FTIR spectra for smoked and non-smoked trout gelatin

Region	Peak wavenumber (cm^{-1})		Assignment and remarks
	Smoked trout gelatin	Non-smoked trout gelatin	
Amide A	3277.17	3277.73	NH stretch coupled with H-bond
Amide B	2934.33	2932.99	CH_2 antisymmetric and symmetric stretching
Amide I	1629.90	1630.57	C=O stretch/hydrogen bond coupled with COO−
Amide II	1523.31	1523.39	NH bend coupled with CN stretch
Amide III	1236.65	1237.46	NH bend stretch coupled CN stretch
Fingerprint	1078.78	1079.64	C=O skeletal stretch

UV-VIS absorption spectra

The ultraviolet absorption spectra of smoked trout and non-smoked trout gelatin are given in Figure 3. As shown in the figure, both smoked and non-smoked trout gelatin showed a strong absorption in the range of 200-240 nm. Both samples reached a maximum absorbance (λ_max) value of around 220–230 nm, which is typically associated with n→π* transitions in peptide bonds and reflects the characteristics of their protein structure (Xu et al., 2017). These bands usually indicate the presence of conjugated systems belonging to amide groups (Duan et al., 2009). In the smoked trout gelatin sample, a slightly higher maximum absorbance value was observed compared to the non-smoked trout gelatin. This increase can be explained by the partial denaturation of the protein structure during the smoking process, the addition of small molecule chromophores (e.g., phenolic compounds, carbonyl groups) or the effect of free aromatic groups formed as a result of structural deterioration (Cansu, 2024). However, both gelatins showing no significant absorption above 240 nm indicates that no significant amounts of nucleic acids, lipids, or other UV-active impurities are present in the samples. These results suggest that the smoking process largely retains the UV-absorption characteristics of the basic protein backbone, although it leads to some minor structural differences. In addition, since collagen does not contain tryptophan but contains low amounts of tyrosine and phenylalanine, it is reported that it generally absorbs a low amount of UV light at 280 nm (Duan et al., 2009). Therefore, the small peaks obtained around the wavelength of 280 nm in gelatin samples, which is the hydrolyzed form of collagen, confirm that the protein is gelatin and that purification takes place effectively.

Emulsion activity and stability

The emulsion activity index reveals how effectively a protein system can adsorb at the oil-water interface and its capacity to form stable emulsions (Ranasinghe et al., 2022). The emulsion activity index (EAI) and emulsion stability index (ESI) of smoked and non-smoked trout gelatin are given in Figure 4, respectively. The emulsion activity index (EAI) of gelatin obtained from smoked trout (*Oncorhynchus mykiss*) skins was measured to be 240.06 ± 0.18 m^2/g. This value is higher compared to the EAI value of 232.20 ± 0.28 m^2/g obtained from non-smoked trout gelatin (*P<0.05),* revealing the potential of gelatin produced from smoked sources in terms of surfactant properties. ESI, on the other hand, refers to the resistance of an emulsion system to decomposition over time and is related to the durability of the layer formed by proteins at the interface. The ESI value of gelatin obtained from smoked trout skins was determined as 26.81 ±

0.04 min. In contrast, the ESI value from non-smoked trout gelatin was 72.57 ± 0.18 min., indicating significantly lower emulsion stabilization capacity for gelatin from smoked sources (*P<0.05*). EAI and ESI values of skin gelatins of various fish species were specified as 13.49 – 41.87 m^2/g and 8.58 – 51.27 min., respectively (Ranasinghe et al., 2022). The values obtained in this study were found to be significantly higher. In addition, the smoking process may have caused structural changes in the protein chains, weakening the ability of gelatin molecules to form a permanent and stable structure in emulsion systems. In particular, heat treatment and smoke components can reduce the conformational flexibility of proteins by causing changes in collagen structure such as partial denaturation, molecular breakage, or crosslinking (Duan et al., 2018; Karim and Bhat, 2009). This shows that although smoked gelatin can support short-term emulsion formation, it is insufficient to provide long-term stability. The findings show that gelatin obtained from smoked fish skins can be used in emulsion systems in applications that require short duration or low stability. Therefore, considering industrial applications such as shelf life, the use of smoked fish gelatin may be preferred.

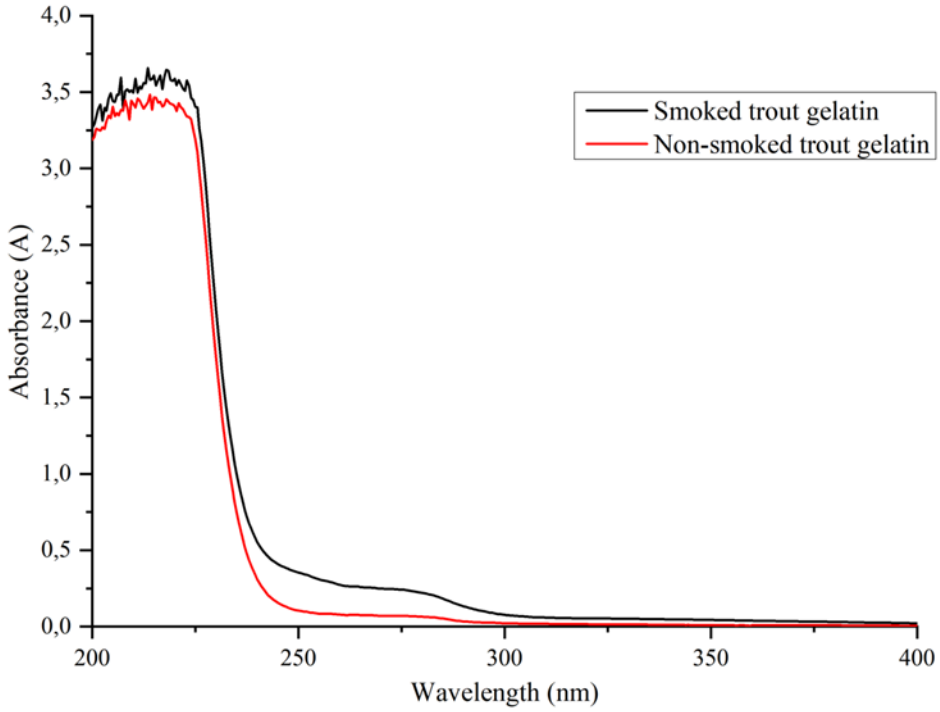

Figure 3. UV-VIS absorption spectra of smoked and non-smoked trout gelatins

Foam capacity and stability

The foaming properties of fish gelatins are important functional properties that can improve the quality of various food products, especially confectionery (Ranasinghe et al., 2022). The foaming capacity (FC) and foam stability (FS) of smoked and non-smoked trout gelatin are given in Figure 5. The foam-forming capacity (FC) of gelatin obtained from smoked trout skins was determined as 85.07 ± 1.15% and the foam stability (FS) was 61.33 ± 10.85%. In contrast, non-smoked trout gelatin exhibited a higher foaming capacity (FC) value (97.73% ± 1.00%) and a slightly higher foam stability (FS) (63.95% ± 15.69%). While there was a statistically significant difference (*P<0.05)* in the FC values of gelatin samples, there was no significant difference in FS values (*P>0.05)*. Ranasinghe et al. (2022) stated that the FC and FS values of various fish gelatins are in the range of 17.4–152.63% and

10.5–147.35%, respectively. The values obtained in this study are within the specified limits. The foam properties of gelatin are closely related to the capacity of proteins to adsorb rapidly on the surface, form films at the interface, and stabilize this film (Shyni et al., 2014). The denaturation that occurs in the collagen structure during the smoking process can limit the participation of proteins in the formation of foam. This may explain the low foam capacity (FC) value compared to non-smoked trout gelatin. However, the foam stability of smoked gelatin was found to be at levels quite close to that of non-smoked trout, indicating that it partially maintain the temporal durability of the film formed on the surface. These results suggest that the smoking process may adversely affect the foam capacity, but acceptable performance in terms of stability can be achieved if the appropriate protein conformation is maintained.

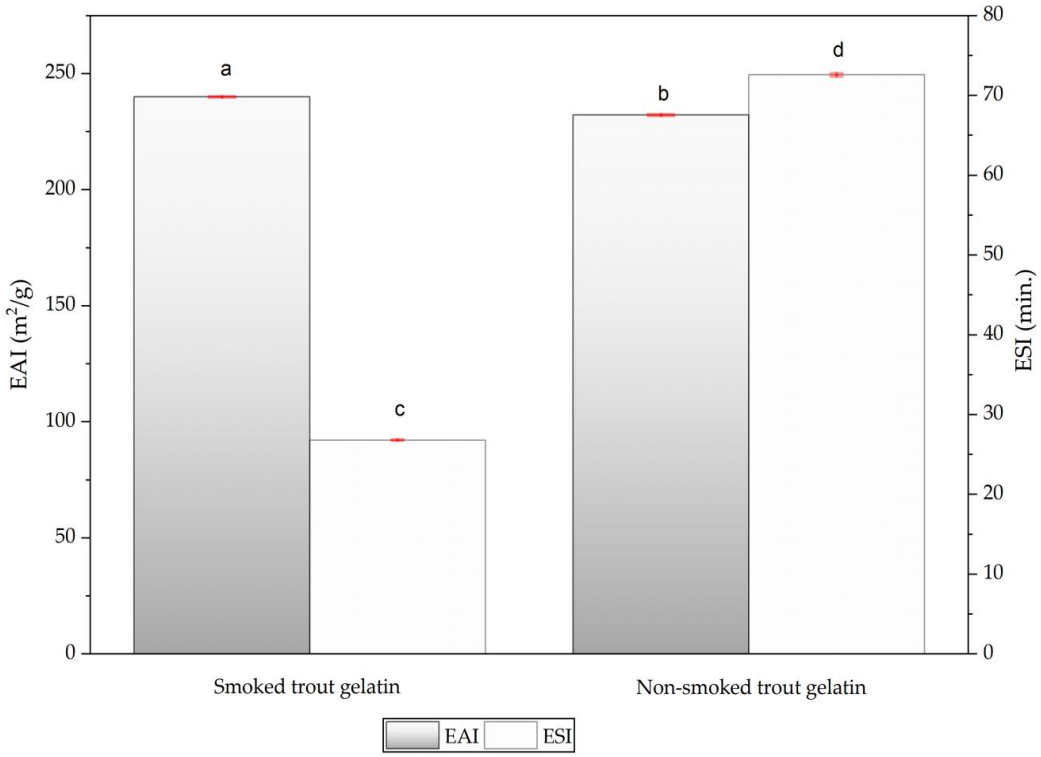

Figure 4. EAI and ESI of smoked and non-smoked trout gelatins

The data are expressed as the mean ± standard deviation of the three measurements. The results were evaluated by analysis of variance (ANOVA) and the Tukey test. Averages with different letters show a statistically significant effect ($P<0.05$), while the same letters show no statistically significant difference ($P>0.05$). EAI: Emulsion activity index, ESI: Emulsion stability index

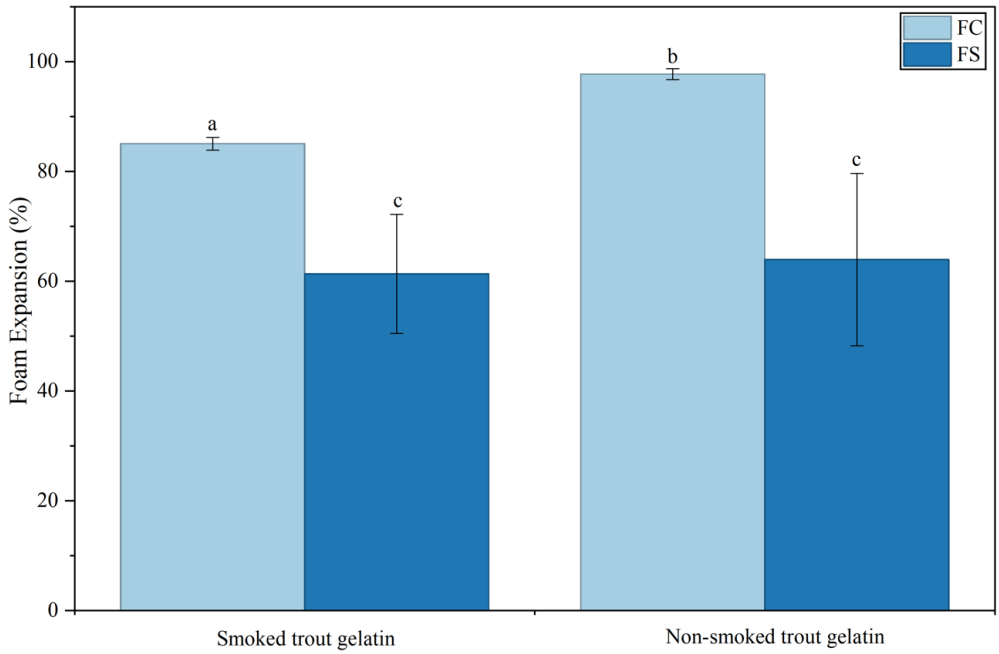

Figure 5. FC and FS of smoked and non-smoked trout gelatin

The data are expressed as the mean ± standard deviation of the three measurements. The results were evaluated by analysis of variance (ANOVA) and the Tukey test. Averages with different letters show a statistically significant effect ($P<0.05$), while the same letters show no statistically significant difference ($P>0.05$). FC: Foam capacity, FS: Foam stability

CONCLUSION

In this study, gelatin was successfully extracted from smoked trout (*Oncorhynchus mykiss*) skins, a significant processing by-product, and its functional and structural properties were systematically compared with those of gelatin obtained from non-smoked trout skins. Although the gelatin yield and gel strength were found to be lower compared to non-smoked trout gelatin, smoked trout gelatin exhibited competitive emulsifying activity, suggesting enhanced surface activity likely due to structural modifications caused by the smoking process. However, its emulsion stability index and oil binding capacity were comparatively lower, indicating limited stabilization ability over time and weaker hydrophobic interactions. Furthermore, smoked trout gelatin exhibited high protein content, considerable foaming capacity, and lightness values distinct from non-smoked trout gelatin. Spectroscopic analyses (FTIR and UV–VIS) confirmed the preservation of characteristic amide bands and aromatic residues, supporting the protein's integrity despite the influence of thermal treatments. Overall, smoked trout skin gelatin holds potential as a partial alternative to conventional fish gelatins, especially in applications requiring enhanced emulsification and acceptable functional behavior. In conclusion, smoked trout skin gelatin presents a sustainable and partially functional alternative to conventional fish gelatin sources. Future studies may explore enzymatic (tansglutaminase etc.) or physical modifications (ultrasound etc.) to further optimize its properties and expand its applicability in food or biomedical fields.

CONFLICTS OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Sefik Tekle: Investigation, methodology, analysis, writing–original draft, review & editing.

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