

Enhanced shelf life of trout fillets using an edible coating containing the polysaccharide extracted from Loquat (*Eriobotrya japonica*) seed

Mojtaba Nouri^a, Homa Baghaei^{a,*}, Mahdi Kashaninejad^b, Abdorreza Mohammadi nafchi^a

^a Department of Food Science and Technology, Da.C., Islamic Azad University, Damghan, Iran

^b Department of Food Process Engineering, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan, Iran

ARTICLE INFO

Keywords:

Fish fillet
Shelf life
Microbial evaluation
Chemical evaluation
Polysaccharide
Loquat (*Eriobotrya japonica*)

ABSTRACT

This study aimed to evaluate the effect of an edible coating enriched with polysaccharides extracted from loquat (*Eriobotrya japonica*) seeds on the microbial, chemical, and sensory quality of trout fillets during 12 days of cold storage. The highest polysaccharide yield (48.2 %) was achieved by applying microwave treatment at 540 W for 15 min. The extract exhibited remarkable antioxidant properties, including 57 % 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 64 % hydroxyl radical (HO) scavenging activity, as well as strong ferric reducing antioxidant power (0.7 absorbance at 700 nm), suggesting its excellent potential to delay oxidative spoilage. To formulate the coating, different ratios of gelatin and loquat seed polysaccharide were tested (G1-P0, G1-P0.5, and G1-P1). Increasing the polysaccharide content resulted in a significant reduction in the growth rate of total and psychrophilic bacteria during storage. Moreover, lower pH values were observed in treatments with higher polysaccharide concentrations. Samples with more polysaccharide also showed reduced levels of chemical spoilage indicators, including peroxide value (PV), thiobarbituric acid reactive substances (TBA), free fatty acids (FFA), total volatile basic nitrogen (TVB-N), and trimethylamine (TMA), all of which are known markers of lipid oxidation and protein degradation in trout fillets. Sensory evaluations confirmed that fillets coated with polysaccharide-rich formulations had significantly better scores in odor, color, and overall acceptability compared to uncoated samples. This study introduces a novel application of loquat seed polysaccharides in edible coatings, offering enhanced preservative effects compared to traditional gelatin-based coatings for trout fillets.

1. Introduction

Recently, consumers have shown a great tendency to fish consumptions due to its rich content of nutrients such as proteins, omega-3 polyunsaturated fatty acids, and vitamins (Bruno et al., 2019). On the other hand, there is a perpetual challenge involved in the utilization of fresh fish as they spoil rapidly due to some biological and chemical interactions including lipid oxidation, autolytic reactions as well as microbial breakdown, resulting from the presence of volatile nitrogen, free fatty acids, and free amino acids, followed by the production of secondary compounds such as aldehydes and ketones (Hamzeh and Rezaei, 2012). Even though the level of deterioration depends on several primary factors, such as the initial quality of fish, harvesting and transportation conditions, and storage circumstances, the above interactions commonly occur in all seafood products, leading to a decline in quality and a shortened shelf life (Jasour et al., 2015).

Generally, there are several common methods to preserve fish

products during storage. Among them, refrigeration is considered the most effective method, as it delays enzymatic and microbial activities by lowering the temperature. However, this method cannot prevent certain undesirable changes, such as fatty acid oxidation and the growth of aerobic microorganisms during storage (Ojagh et al., 2010).

Another critical challenge in preserving fresh fish is the application of illegal chemical preservatives, which have been proven hazardous to human health (Li et al., 2019). Nowadays, due to the rising consumer awareness about the adverse effects of traditional preservation techniques and an increasing demand for high-quality, fresh, and safe products, these methods are no longer acceptable to the public. Hence, the employment of novel and safe preservation techniques appears essential to meet consumer expectations. In this regard, edible coating, which is one of the most efficient methods for food products, has attracted scientists' attention to overcome the mentioned problems (Kazemian-Bazkiaee et al., 2020). Edible coatings are thin layers of edible material that are applied to the surface of food as protective or

* Corresponding author.

E-mail address: Homa.baghaei@iau.ac.ir (H. Baghaei).

<https://doi.org/10.1016/j.fufo.2025.100693>

Received 2 May 2025; Received in revised form 7 June 2025; Accepted 21 June 2025

Available online 5 July 2025

2666-8335/© 2025 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

functional coatings, primarily intended to enhance shelf life, quality, and safety (Aguirre-Joya et al., 2018).

In this context, biodegradable edible coatings have emerged as a green and cost-effective alternative for extending the shelf life of both fresh and processed foods (Brasil et al., 2012). This coating system not only protects food products from spoilage but also enhances their nutritional quality by preserving existing vitamins and enabling the delivery of additional nutrients, as it is typically formulated from natural compounds (Suhag et al., 2020). Edible coatings function by creating a barrier to oxygen and moisture, minimizing water loss, and maintaining the aroma, taste, texture, and nutrient content of the product, thereby improving its overall quality and consumer appeal (Körge et al., 2020).

Various natural polymers, including proteins, lipids, and polysaccharides, have been widely used in the production of edible coatings due to their excellent biodegradability, availability, and safety for both human health and the environment (Campos et al., 2011). Among these biopolymers, polysaccharides stand out as natural macromolecules composed of monosaccharide units linked through glycosidic bonds (Díaz-Montes, 2022). These natural polysaccharides can be derived from diverse sources, including plants, animals, marine algae, and microorganisms such as yeasts, fungi, and bacteria (Ren et al., 2019; Sharma, 2021). Particularly, plant-based polysaccharides are well recognized for their antimicrobial and antioxidant properties, making them highly suitable for applications in food preservation systems, especially as components of edible coatings (Ren et al., 2019).

In recent years, increasing attention has been paid to the use of natural compounds in improving the quality, safety, and shelf life of aquaculture products. For instance, Anshar et al. (2023) demonstrated that the incorporation of natural feeds such as *Daphnia* sp. and synbiotics significantly enhanced the growth performance and survival rate of *Marosatherina ladigesi*, highlighting the value of biologically active inputs in aquaculture. Similarly, Manzoor et al. (2023) emphasized the necessity of controlling microbial infections such as *Edwardsiella tarda* in tilapia through early detection and biosecurity measures. Moreover, Alsulami and El-Saadony (2024) reported that bacterial zinc nanoparticles improved immune response and reduced microbial load in Nile tilapia, further supporting the role of bioactive materials in enhancing fish health.

Among plant-derived sources, loquat (*Eriobotrya japonica*) seeds have recently attracted attention as a novel and underutilized material rich in polysaccharides with notable antioxidant and antimicrobial properties, suggesting their potential for application in functional edible coatings (He et al., 2022).

Loquat (*Eriobotrya japonica*) is a native to southeastern China and it was introduced to the Japanese in ancient times (Liu et al., 2016). Different parts were found to be beneficial for human health and treating some diseases such as diabetes, cancer, cholesterol, infections, as well as cardiovascular and digestion problems as they are rich in fiber, potassium, manganese, different vitamins along with antioxidant compounds such as flavonoids, vitamin C, phenols, polysaccharides, etc. (Lee et al., 2015). There are around 1–5 brown seeds inside all fruits of Loquat which are discarded as waste while they abundantly contain some valuable compounds such as polysaccharides (Y. Liu et al., 2016). Thus, this wasted seed can be used as a natural source of polysaccharides which can potentially be potential enough to be used in food packaging to improve the food quality extending their shelf life. Polysaccharides from natural sources can be obtained by using some common methods including the usage of organic solvents, heating, and boiling (Shi, 2016). Applying these traditional techniques not only parallels the consumption of too much time, energy, and solvents but also leads to the production of a polysaccharide with lower functional and antioxidant properties which is not reasonable for industry (Okolie et al., 2019). Therefore, as the final properties of a polysaccharide can be affected by the method of extraction, novel techniques such as microwave, supercritical fluid, and ultrasound have been recently introduced, since they can improve the efficiency (Ren et al., 2019). To our knowledge, this is

the first study to apply microwave-extracted loquat (*Eriobotrya japonica*) seed polysaccharide in a gelatin-based edible coating aimed at preserving trout fillets.

Rainbow trout (*Oncorhynchus mykiss*) are widely farmed around the world due to their resistance to environmental changes during breeding (Dursun and Erkan, 2014). Additionally, this species is abundantly used for producing processed products such as trout fillets due to a growing demand for the utilization of processed trout (Heidari and Rezaei, 2022). Due to this reason, use of a novel protection method such as edible coating seems to be necessary to provide safe and high-quality trout fillets. Until now, some investigations have been conducted regarding the utilization of edible coatings prepared by using natural compounds such as polysaccharide for preserving edible products but most of them have been limited to fruits and vegetables (Salehi, 2020; Tahir et al., 2019). Moreover, there is no original study related to producing an edible coating based on loquat polysaccharide for improving the quality of trout fillets during storage. The aim of this study was the analysis of antioxidant properties of loquat seeds polysaccharide extracted by microwave method which was further used in preparing a gelatin-based edible coating. The quality of trout fillets was subsequently analyzed during storage while they were coated in prepared edible coatings.

2. Materials and methods

2.1. Chemicals

Chitosan, glycerol, methanol, ethanol, butanol, acetic acid, thiobarbituric acid reagent, nutrient agar medium, PCA medium, DPPH, ferrous sulfate, sodium salicylate, ascorbic acid, potassium ferricyanide, iron (III) chloride (FeCl₃), chloroform, potassium iodide, potassium carbonate, trichloroacetic acid (TCA), and NaOH were purchased from Merck Company (Merck, Frankfurter Str. 250, 64,293 Darmstadt, Germany).

2.2. Sample preparation and polysaccharide extraction

Loquat was collected from a garden located in Gorgan, Iran, and then transferred to the food science laboratory, Gorgan, Iran. After cleaning and washing, the seeds were separated from the fruits and then crushed to obtain a uniform powder. The sample was then allowed to dry at room temperature (25 °C). The dried sample was defatted based on the Soxhlet method by using petroleum ether solvent (Jiang et al., 2014). The resultant powder was weighed and kept at room temperature (25 °C) for further utilization.

Polysaccharide extraction from Loquat seeds was conducted according to the method of Thirugnanasambandham et al. (2015) with a slight modification. The recovery process was carried out by using a microwave (MARS6, CEM, The USA) equipped with a 1-liter round bottom flux, reflux, and condenser. The sample was mixed with distilled water at a ratio of 20 % (w/v), corresponding to 20 g of loquat seed powder per 100 mL of distilled water, and subjected to microwave treatment at three power levels (180, 360, and 540 W) for 15 min which were then named microwave-assisted polysaccharide (MASP-180, MASP-360, and MASP-540). The resultant solution was concentrated using a rotary and then mixed with 95 % ethanol (V/V) and kept at 4 °C for 48 h. After centrifugation (6000 rpm for 30 min), the precipitate was collected and washed using acetone. Finally, the sample was dried by employing a freeze dryer and stored for further testing.

2.3. Polysaccharide characterization

2.3.1. Recovery yield

The recovery yield of extracted polysaccharide was measured based on both wet and dry weight by applying the following equation (Bougatef et al., 2019):

$$\text{Dry weight based yield (\%)} = \frac{\text{Dry weight of polysaccharide}}{[(\text{Wet weight of seed} - \text{Moisture content})] \times 100}$$

2.3.2. Fourier transform infrared (FTIR)

FTIR spectroscopy of the extracted polysaccharide was analyzed based on the method of Alboofetileh et al. (2019) with a minor modification. The polysaccharide sample (2 mg) was mixed with potassium bromide powder (300 mg) and compressed into a tablet with a thickness of 1 mm. The FTIR peaks of the polysaccharide were obtained within the frequency range of 400–4000 cm⁻¹ by utilizing FTIR spectra (TENSOR 27, Bruker, Germany) with an accuracy of 0.11 cm.

2.3.3. Antioxidant properties

2.3.3.1. DPPH radical scavenging activity. The DPPH radical scavenging activity of the extracted polysaccharide was evaluated using the method described by Chen et al. (2012). Polysaccharide solutions at different concentrations (0.5–3 mg/mL) were prepared by mixing the samples with ethanol, followed by incubation at 25 °C for 40 min. Absorbance was measured at 517 nm using a UV/Vis spectrophotometer (Model 2100, Japan), and ascorbic acid (vitamin C) was used as a positive control. The DPPH radical scavenging activity (%) was calculated as follows:

$$\text{DPPH radical scavenging activity(\%)} = \frac{(A_c - A_s)}{A_c} \times 100$$

Ac and As are demonstrated as the absorption of the control group and the absorption of the polysaccharides

2.3.3.2. Hydroxyl radical scavenging activity. Hydroxyl radical scavenging activity of the recovered polysaccharide was calculated based on the method of Xue et al. (2018) with a slight modification. Different concentrations (0.05–0.3 mg/mL) of polysaccharide were prepared by diluting the polysaccharides in ethanol. Specifically, 1 mL of each concentration was then mixed with 1 mL of ferrous sulfate (2 mM) and 0.5 mL of sodium salicylate (20 M). After adding 0.5 mL of H₂O₂ (6 mM) to the prepared solutions, DMSO solution at a ratio of 1:10 was used to facilitate the dissolving process. The final solutions were then kept at 37 °C for 50 min. The absorbance of the samples was recorded at a wavelength of 562 nm using a spectrophotometer (UV/vis 2100, Japan) while ascorbic acid (vitamin C) was used as a positive control. Hydroxyl radical scavenging activity of the polysaccharides was evaluated based on the following equation:

$$\text{Hydroxyl radical scavenging activity(\%)} = \frac{(A_c - A_s)}{A_c} \times 100$$

Ac and As are demonstrated as the absorption of the control group and the absorption of the polysaccharides

2.3.3.3. Ferric-reducing antioxidant power (FRAP). Ferric-reducing antioxidant power (FRAP) of the extracted polysaccharide was measured according to the procedure of Chew et al. (2008) with a slight modification. Here, 1 mL of different concentrations (0.05–0.3 mg/mL) of polysaccharide was mixed with 2 mL of 0.2 M phosphate buffer (pH=6.6) and 2 mL of potassium ferricyanide (1 % w/v). The mixture was then heated to 50 °C for 20 min. After immediate cooling, 10 % TCA was added to the mixture in order to stop the reaction. The sample was then centrifuged at 3000 rpm for 10 min. The collected supernatant was mixed with 2 mL of deionized water and 0.4 mL of 1 % iron (III) chloride (FeCl₃). After 10 min, the absorbance of the samples was recorded at the wavelength of 700 nm using a spectrophotometer (UV/vis 2100, Japan), while ascorbic acid (vitamin C) was used as a positive control.

2.4. Fish fillets' preparation and coating process

Fresh rainbow trout was provided from a fish pond located in Gorgan, Iran. The purchased fish were transferred to the laboratory while they were kept in an ice jacket. Upon arrival, the head and tail were immediately removed, with the fish being manually filleted and washed using cold distilled water. The prepared fillets were then kept in the refrigerator (4 °C) until the time of coating. The coating process was carried out according to the method of Johns and Stead (2002), in which gelatin (2 %) was prepared for coating, while glycerol was used as a plasticizer. The extracted polysaccharide was added to the prepared gelatin coating at three different ratios (1:0, 1:0.5, and 1:1) which were then named G1-P0, G1-P.5, and G1-P1. The fish fillets were first separated and then dipped into the prepared coating solution, while an uncoated fillet was used as a control sample.

2.5. Common tests quality measurement of coated fillets

2.5.1. Microbial evaluation

2.5.1.1. Total microorganism counting test (TC). Total Microorganism Counting Test (TC) was performed to count the total amount of bacteria, yeasts, and molds. Suspension and dilution preparation of this microbial test was carried out according to standard 356 and related standards as follows. Initially, the sample was diluted in a physiological serum under a sterile condition in order to prepare 1–10 dilutions. The prepared solutions were then shaken thoroughly for 15 s in order to become homogenous and uniform. The culture medium was molten agar which was cooled at 45 °C. The resultant solutions were then transferred into plates and 15 mL of culture medium was poured into the plates. All plates were then mixed under sterile conditions to ensure uniform distribution of the culture medium and the sample. Following the culture medium solidification, the plates were turned upside down and incubated at 30 °C for 72 h. Next, all colonies that were grown in the culture medium were carefully counted by the colony counter. After the total counting, the plates with two consecutive dilutions, which had at least 15 colonies and at most 300 colonies, were selected for counting and their average was calculated according to the following formula:

$$\text{Average colony counting(CFU / g - 1)} = \frac{\text{Number of colonies} \times \text{dilution factor}}{\text{Volume of culture plate}}$$

2.5.1.2. Psychrophilic bacteria counting. The counting of the amount of psychrophilic bacteria was conducted using the similar method of counting total bacteria mentioned in the previous section. The only difference was that the culture medium used in this test was PCA and it was kept at 7 °C for 1 week.

2.5.2. Chemical evaluation

2.5.2.1. pH measurement. The pH value of the fish fillets was analyzed by mixing 10 g of minced fillets with 90 mL distilled water and allowing them to get homogenized at a speed of 1000 rpm. The pH value was measured using a pH meter (Lenzkriech, testo 205, Germany).

2.5.2.2. Peroxide value (PV). The peroxide value (PV) of the fish fillets was calculated according to the method of Rezaei and Shahbazi (2018). Here, 30 mL of the prepared solvent which was a mixture of acetic acid, chloroform, and 0.5 mL saturated potassium iodide was added to the 3 g of oil sample and allowed to be stirred for 1 min. After stirring, 0.5 mL of 1 % starch was added to the solution. In the presence of peroxide, a purple ring was formed in the upper part of the container, which was then titrated with 0.01 normal sodium thiosulfate until the solution became colorless. Finally, the amount of peroxide was calculated in milliequivalents of active oxygen per kilogram of sample according to

the following equation:

$$PV = \frac{[(S - B) \times N \times 1000]}{W}$$

S and B demonstrate the volume of sodium thiosulfate for titration and control sample (mL), N shows the normality of sodium thiosulfate solution, and W denotes the oil sample weight (g)

2.5.2.3. Thiobarbituric acid (TBA). The level of thiobarbituric acid in the coated fillets was measured according to the method of [Natseba et al. \(2005\)](#). Specifically, 200 mg of fish paste was mixed with butanol in a 25 ml flask and let them get mixed thoroughly. Then, 5 ml of the sample solution was mixed with 5 ml of the reagent solution (thiobarbituric acid in butanol) and placed in a steam bath with a temperature of 95 °C for 2 h. The test tubes containing the samples were cooled using cold water for 10 min. The sample's absorbance was recorded at a wavelength of 530 nm. The amount of thiobarbituric acid was calculated using the following equation:

$$TBA = \frac{(50 \times A_{532})}{m}$$

Where, 50 is a constant number based on the flask volume (25 mL) and cell length (1 cm), A_{532} represents the sample absorbance, and m demonstrates the weight of the sample (mg).

2.5.2.4. Free fatty acid (FFA). The amount of free fatty acids in fish fillets was calculated according to the procedure of [Senphan and Benjakul \(2015\)](#). At first, 0.1 g oil sample and 5 mL isooctane solvent were poured into 15 mL test tubes which were allowed to mix completely. Then, 5 g copper acetate was mixed with 100 mL distilled water with the pH adjusted at 6–6.2 using pyridine, aiming at producing copper acetate-pyridine reagent (5 %). Thereafter, 1 mL of copper acetate-pyridine reagent (5 %) was added to the solution and allowed to be shaken for 90 S. The supernatant was taken and its absorbance was read at 715 nm. Palmitic acid in isooctane solution at the concentrations of 0–5–5 µmol/mL was used for preparing the standard curve.

2.5.2.5. Total volatile base nitrogen (TVB-N). The total volatile basic nitrogen (TVB-N) of fish fillets were calculated based on the protocol of [Siang and Kim \(1992\)](#). Firstly, an inner ring (containing the mixture of boric acid solution and the indicator mixture) and an outer ring (containing fish tissue extract and saturated potassium carbonate solution) were prepared. For making the inner ring, Boric acid was provided by adding 200 mL of ethanol (100 %) to a mixture of 10 g of boric acid and distilled water. The indicator mixture was obtained by mixing 0.01 g of bromocresol green and 0.02 g of methyl red with 10 mL of ethanol. The boric acid solution and the indicator mixture were then mingled together and distilled water was used to expand the volume of the solution to 1000 mL. The resultant mixture was then homogenized to become more uniform. In order to produce the outer ring, the saturated potassium carbonate was prepared by dissolving 60 g of potassium carbonate in 50 mL of distilled water followed by heating for 10 min. The final solution was then cooled and filtered using filter paper. Next, 2 g of fish tissue and 8 mL of trichloroacetic acid (8 %) were mixed and homogenized for 1 min to prepare the testing sample. The mixture was placed at room temperature for 30 min and was stirred manually at different times. The sample was filtered and trichloroacetic acid (4 %) was employed to increase the volume of the solution to 10 mL where the final extract was stored at –20 °C. The control group contained only 10 mL of trichloroacetic acid 94 %). After making the needed solutions, the fish tissue (1 mL) and the inner ring solution (1 mL) were respectively added to the outer and inner ring of the Conway flask while saturated potassium carbonate (1 mL) was placed on the opposite side of the outer ring. The poured flask was closed and rotated clockwise and then heated at 37 °C for 1 h using an oven. Finally, the amount of inner ring was recorded by an insulin syringe after using hydrochloric acid (0.02 N) for

titration. The amount of fish fillet TVB-N was calculated based on the following equation:

$$TVBN \text{ (mgN / 100g)} = \frac{(V_S - V_B)(N_{HCL} \times A_n) \times V_E \times 100}{W_S}$$

VS and VB represent the consumption volume of hydrochloric acid (0.02) for the main sample and control sample, NHCL denotes the normality of consumption of hydrochloric acid, AN demonstrates the atomic mass of nitrogen (equation 14), WS is the weight of the fish tissue sample, and VE reflects the volume of trichloro acetic acid used for fish tissue extract.

2.5.2.6. Trimethylamine (TMA). The level of trimethylamine (TMA) in fish fillets was calculated based on the AOAC method ([AOAC, 1995](#)). In order to prepare the sample, 10 g of fish tissue was mixed with 30 ml of trichloroacetic acid (7.5 %) and then homogenized for 2 min until a milky solution was obtained. The homogenized tissue was centrifuged at 2500 rpm for 10 min where the supernatant was used as fish tissue extract. The standard solution was prepared by adding 1, 2, and 3 mL of TMA standard solution to 4 mL of distilled water, while an empty tube was also considered as a control. The standard curve was drawn by determining the amount of light absorption. The amount of TMA was calculated by determining the light absorption in the unknown sample and using the prepared standard curve.

2.5.3. Sensory evaluation

The sensory evaluation of fish fillets was carried out according to the method of [Ojagh et al. \(2010\)](#) with a slight modification. In this method, six semi-trained people (four males and two females aged from 25 to 39) were used for sensory analysis. The quality of raw fish kept in the refrigerator was measured by considering three quality parameters including odor (5, extremely desirable; 1, off-odors), discoloration (5, no discoloration; 1, extreme discoloration), and overall acceptability (5, extremely desirable; 1, extremely unacceptable). A sensory score of below 4 is considered as a shelf life rejection. However, the small size of the sensory panel ($n = 6$) represents a limitation of this study. Future research should consider using a larger and more diverse panel to improve the reliability and generalizability of sensory results.

2.6. Statistical analysis

Statistical analysis was performed using IBM SPSS Statistics for Windows, Version 18.0 (IBM Corp., Armonk, NY, USA). Prior to conducting the one-way analysis of variance (ANOVA), the assumptions of normality and homogeneity of variance were tested and confirmed. Duncan's multiple range test was used for post-hoc comparisons at a significance level of $P < 0.05$. Results are presented as mean \pm standard deviation (SD), based on triplicate measurements. Microsoft Excel 2016 (Microsoft Corp., Redmond, WA, USA) was used to prepare graphs. Assumptions of normality and homogeneity of variance were not explicitly tested; however, the experimental design and distribution of data did not suggest significant violations.

3. Results and discussion

3.1. Polysaccharide characterization

3.1.1. Recovery yield

The recovery yield of the extracted polysaccharide was improved by increasing the power of the microwave, in which the lowest recovery yield was obtained by applying the microwave with 180 W (33.4 ± 0.77 %), while the superior yield was obtained by applying the microwave with the power of 540 W (48.2 ± 0.53 %), which was chosen as the optimum treatment for edible coating ($P < 0.05$). The microwave method can improve the recovery yield by breaking down the outer

layer of plants by diminishing the emulsion viscosity, resulting from a higher temperature caused by higher microwave power. Yield can also be improved due to enhanced ion movement, which results from the rearrangement of electrical charges around the molecules (Thirugnanasambandham et al., 2015). In this study, the polysaccharide yield was higher than that of *Artemisia sphaerocephala* (31.8 %) extracted by microwave, but lower than that of mung bean hulls (60 %) obtained using the same method (Wang et al., 2009; Zhong et al., 2012).

3.1.2. Fourier transform infrared (FTIR)

FTIR analysis is usually performed to examine the types of glycosidic bonds, monosaccharides, and functional groups in polysaccharides. The FTIR spectrum of the recovered polysaccharide is displayed in Fig. 1. The absorption peak observed at 3428 cm^{-1} corresponds to the stretching vibrations of O–H bonds, which are characteristic of hydroxyl groups forming the main framework of polysaccharides (Li et al., 2017). The peak detected at 2930 cm^{-1} is attributed to the C–H stretching vibrations, indicating the presence of aliphatic $-\text{CH}_2$ or $-\text{CH}_3$ groups, commonly found in sugar residues of polysaccharides (Li et al., 2020). Another C–H stretching vibration was observed at 2860 cm^{-1} (Thirugnanasambandham et al., 2015). The strong peaks at 1659 cm^{-1} and 1759 cm^{-1} are associated with the presence of carboxyl groups, which contribute to the antioxidant activity of polysaccharides (Zhu et al., 2012). The peak at 1433 cm^{-1} indicates the presence of O–H or C–H bending vibrations (Wu et al., 2019). Peaks at 1066 , 1111 , and 1290 cm^{-1} correspond to C–C and C–O stretching vibrations. Additionally, the peak at 609 cm^{-1} is indicative of the anomeric structure in pyranose rings of sugars. Similar absorption peaks have been reported for polysaccharides extracted from loquat (*Eriobotrya japonica*) cultivars and leaves (Fu et al., 2019, 2020).

3.1.3. Antioxidant properties

3.1.3.1. DPPH radical scavenging activity. DPPH radical is a stable nitrogen-centered radical with hydrogen-accepting capability, commonly used to evaluate the antioxidant properties of bioactive compounds. The DPPH radical scavenging activity of the extracted polysaccharide at different concentrations (0.05–0.3 mg/mL) is shown in Fig. 2A. At all tested concentrations, ascorbic acid exhibited significantly higher scavenging activity than the polysaccharide. However, at concentrations of 0.1 and 0.2 mg/mL, the difference between the antioxidant activities of ascorbic acid and the polysaccharide was minimal.

The scavenging ability of the polysaccharide increased with increasing concentration. The highest DPPH radical scavenging activity was recorded at 0.3 mg/mL ($57 \pm 1.77\%$), while the lowest was observed at 0.05 mg/mL ($22 \pm 3.1\%$) ($P < 0.05$).

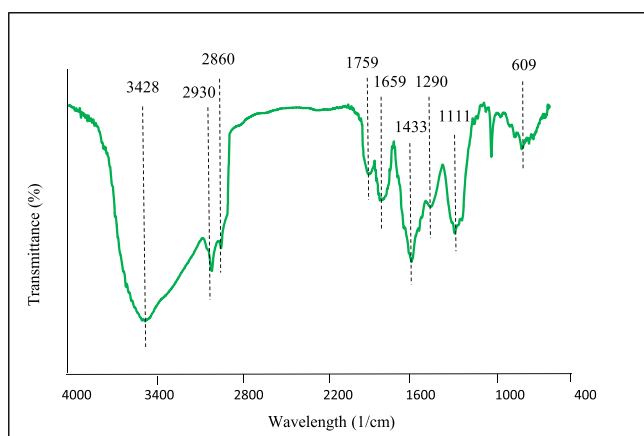


Fig. 1. Fourier transform infrared spectroscopy (FTIR) of the polysaccharide extracted from loquat seed using microwave (540 W).

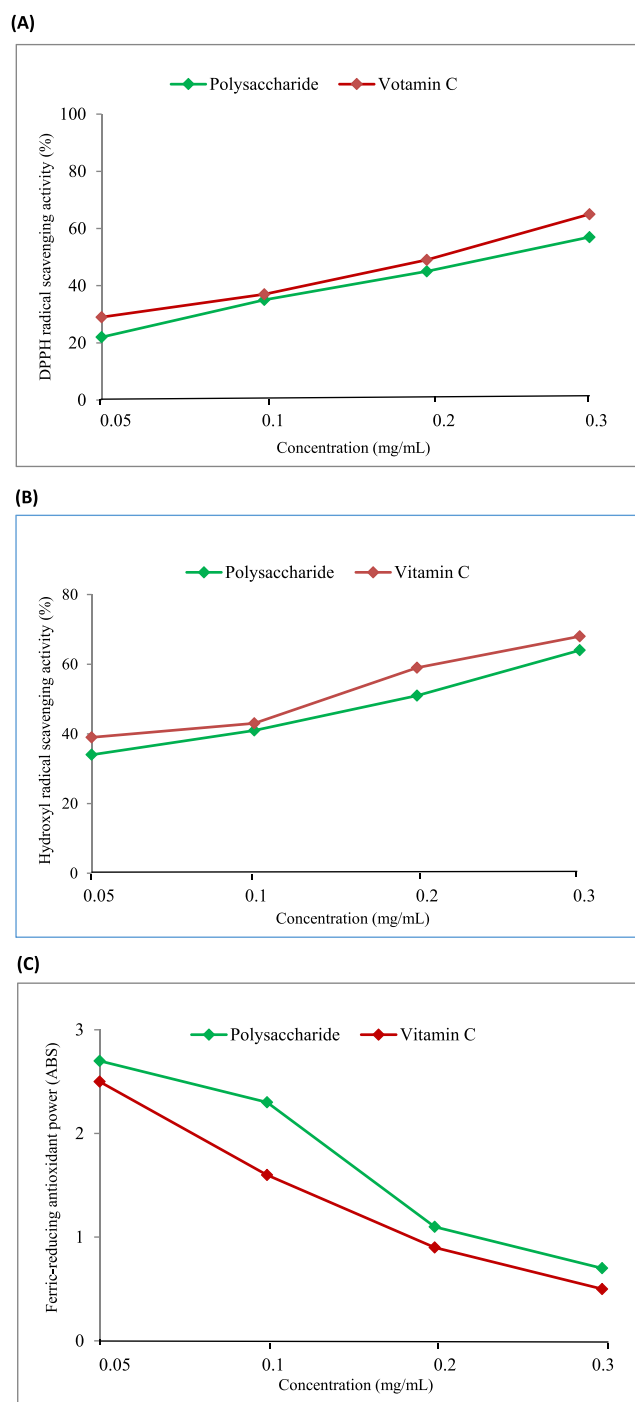


Fig. 2. DPPH radical scavenging activity (A), hydroxyl radical scavenging activity (B), and ferric reducing antioxidant power (FRAP) (C) of the polysaccharide extracted from loquat seed using microwave (540 W). Vitamin C was used as standard.

These findings suggest that the antioxidant activity of polysaccharides may be closely related to their monosaccharide composition. Monosaccharides act as reducing agents by donating hydrogen atoms to free radicals, thus forming stable molecules (Jiao et al., 2011). It has been reported that mannose content is positively correlated with antioxidant activity, whereas glucose content shows a negative correlation (Meng et al., 2017).

Another influential factor is the molecular weight of polysaccharide, in which the antioxidant properties are enhanced by reducing the molecular weight. Fan et al. (2009) reported that the DPPH radical

scavenging activity of *Dendrobium denneanum* polysaccharide improved as the concentration increased. An almost similar DPPH inhibiting activity was also reported by Yu et al. (2019), where they recovered the polysaccharide from *Chlorella vulgaris* with (65.1 %, at 0.4 mg/mL) DPPH scavenging activity. In addition, Rostami and Gharibzadeh (2016) reported an almost similar DPPH scavenging activity (67.0–74.1 %, at 0.2 mg/mL) for the polysaccharide extracted from *Zizyphus jujuba* Mill.

3.1.3.2. Hydroxyl (OH) radical scavenging activity. The hydroxyl (OH) radical scavenging activity of the extracted polysaccharide is represented in Fig. 2B. As observed, ascorbic acid showed a superior ability to inhibit hydroxyl radicals compared to the extracted polysaccharide. However, at concentrations of 0.1 and 0.3 mg/mL, the OH radical scavenging activities of both ascorbic acid and the polysaccharide were found to be almost similar. The scavenging activity of the polysaccharide increased significantly with increasing concentration ($P < 0.05$), with the highest activity observed at 0.3 mg/mL (64 ± 1.83 %) and the lowest at 0.05 mg/mL (34 ± 2.58 %).

These findings are consistent with previous studies on natural polysaccharide-based coatings. For instance, Zhang et al. (2020) reported that chitosan derivatives exhibited concentration-dependent hydroxyl radical scavenging activities, with certain derivatives achieving up to 100 % scavenging at higher concentrations. Similarly, Rizfa et al. (2020) demonstrated that sodium alginate extracted from *Sargassum* species showed enhanced antioxidant activity, including hydroxyl radical scavenging, when subjected to thermal and chemical modifications. These studies support the potential of natural polysaccharide-based coatings in enhancing antioxidant properties and extending the shelf life of food products.

The higher OH scavenging activity of the polysaccharide at a higher concentration might be attributed to the higher mannose content, which can provide the necessary hydrogen to reduce the level of free radicals (Meng et al., 2017). In a comparable study, Liu et al. (2013) reported that the OH-inhibiting ability of *Lycium ruthenicum* polysaccharide was enhanced by elevating the concentration.

The OH radical scavenging activity of the extracted polysaccharide in this study was higher than that of the OH scavenging activity of the polysaccharide extracted from *Potentilla anserina* (50.1 %, at 10.0 mg/mL). Ren et al. (2017) and Yu et al. (2019) reported an almost similar OH scavenging activity of the polysaccharide extracted from *S. thunbergii* (68.7–72.4 %, at 0.8 mg/mL) and *C. vulgaris* (56.2 %, at 0.4 mg/mL), respectively.

3.1.3.3. Ferric reducing antioxidant power. Ferric reducing antioxidant power of the extracted polysaccharide is presented in Fig. 2C. Ascorbic acid demonstrated a higher ferric-reducing ability than the polysaccharide at all tested concentrations, though the values were comparable at 0.05 mg/mL. Interestingly, the ferric-reducing power of the polysaccharide decreased with increasing concentration ($P < 0.05$). The highest ferric reducing capacity was observed at 0.05 mg/mL (2.7 ± 0.35), while the lowest was recorded at 0.3 mg/mL (0.7 ± 0.24).

This inverse trend may be attributed to molecular aggregation at higher concentrations, which can hinder the exposure of functional hydroxyl and carboxyl groups involved in electron donation.

The reducing power of the polysaccharide could be linked to its capacity to donate hydrogen atoms and convert Fe^{3+} to Fe^{2+} , thereby breaking the free radical chain and limiting peroxide formation.

It has been demonstrated that microwave-assisted extraction can enhance the antioxidant capacity of polysaccharides by modifying their structure and disrupting the cell wall (Kaufmann and Christen, 2002).

Compared to previous studies, the polysaccharide in this study showed higher ferric reducing power than those extracted from *Ulva prolifera* (0.27 ABS, at 2 mg/mL) and *Passiflora edulis* Sims (0.32 ABS, at 0.25–1.5 mg/mL) (Xiong et al., 2019; Yuan et al., 2018).

3.2. Common tests quality measurement of coated fillets

3.2.1. Microbial evaluation

3.2.1.1. Total and psychrophilic bacteria counting. Total microbial counts increased over storage time, but fillets treated with the G1-P1 coating exhibited significantly lower microbial loads compared to other treatments, highlighting its superior antimicrobial efficacy. The average number of total microorganisms during storage is outlined in Table 1A. The spoilage threshold of 4.69 Log CFU/g has been introduced for fish fillets (Jonušaite et al., 2021). Accordingly, all samples remained within acceptable microbial limits up to day 7 of storage, although statistically significant differences were observed between the control and the treated samples ($P < 0.05$).

As expected, microbial load increased in all samples over time. By day 12, the control group exceeded 8.5 Log CFU/g, whereas the G1-P1 treatment maintained significantly lower levels, under 8.0 Log CFU/g, indicating that the coating effectively delayed microbial spoilage Table 1B.

Psychrophilic bacteria also increased over time, but their growth was notably inhibited in the G1-P1 treatment compared to the control and other coating groups. The microbial limit for psychrophilic bacteria is defined as 6 Log CFU/g (Jonušaite et al., 2021). Only the control sample and the gelatin-only treatment (G1-P0) surpassed this limit by the end of storage. By day 12, G1-P1 exhibited the lowest psychrophilic count (~ 5.5 Log CFU/g), remaining within acceptable limits, while the control sample exceeded 6.4 Log CFU/g.

The consistently lower levels of total and psychrophilic microorganisms in the G1-P1 treatment confirm the antimicrobial properties of the extracted polysaccharide, likely through disruption of bacterial membranes and enzyme inhibition. These findings are in agreement with studies by Wang et al. (2018) and Guerreiro et al. (2015), which showed similar microbial reductions using polysaccharide-based edible coatings.

3.2.2. Chemical evaluation

3.2.2.1. pH measurement. The pH levels of all samples increased during storage, but fillets treated with the G1-P1 coating showed the slowest rise, remaining below the spoilage threshold throughout the period. A pH value of 7.5–7.8 is generally considered the spoilage level in fish fillets. The pH variations during storage are illustrated in Fig. 3A. The increase in pH over time in all treatments is primarily attributed to the accumulation of volatile nitrogenous compounds such as ammonia and trimethylamine, produced by bacterial and endogenous enzymatic activities (Zhou et al., 2021; Li et al., 2022).

Similar studies using natural edible coatings like chitosan and alginate have also reported slower pH increases during storage, indicating their effectiveness in delaying spoilage. For example, Wang et al. (2020) demonstrated that chitosan coatings significantly inhibited pH rise by limiting bacterial growth, while Garcia et al. (2021) showed that

Table 1A

Total microorganism counting test (TC) of trout fillets during 12 days of storage at 4 °C (log CFU/g⁻¹).

Storage time	C	G1-P0	G1-P00.5	G1-P1
Day 0	3.4 ± 0.09^a	3.4 ± 0.09^a	3.4 ± 0.09^a	3.4 ± 0.09^a
Day 1	3.8 ± 0.1^a	3.7 ± 0.08^{ab}	3.6 ± 0.07^b	3.3 ± 0.1^c
Day 4	4.5 ± 0.08^a	4.3 ± 0.16^a	4.2 ± 0.27^b	4.1 ± 0.24^c
Day 7	5.6 ± 0.31^a	5.5 ± 0.11^a	5.5 ± 0.13^a	5.2 ± 0.19^b
Day 10	7.1 ± 0.31^a	6.9 ± 0.25^b	6.9 ± 0.17^b	6.7 ± 0.15^c
Day 12	8.7 ± 0.17^a	8.3 ± 0.21^b	8.7 ± 0.37^c	7.8 ± 0.24^d

C, G1-P0, G1-P00.5, and G1-P1 are known as control and gelatin and polysaccharide contained coating (with the ratio of 1:0, 1:0.5, and 1:1). Data provided as mean \pm standard deviation. The significant difference is shown by superscript letters ($P < 0.05$).

Table 1B

Psychrophilic bacteria counting of trout fillets during 12 days of storage at 4 °C (log CFU/g).

Storage time	C	G1-P0	G1-P0.5	G1-P1
Day 0	2.7 ± 0.09 ^a	2.7 ± 0.09 ^a	2.7 ± 0.09 ^a	2.7 ± 0.09 ^a
Day 1	3.2 ± 0.1 ^a	3.1 ± 0.12 ^b	3 ± 0.57 ^c	2.9 ± 0.14 ^d
Day 4	3.9 ± 0.13 ^a	3.6 ± 0.06 ^b	3.4 ± 0.24 ^c	3.5 ± 0.24 ^d
Day 7	4.6 ± 0.51 ^a	4.2 ± 0.27 ^b	3.9 ± 0.13 ^c	3.5 ± 0.21 ^d
Day 10	5.6 ± 0.18 ^a	5.1 ± 0.22 ^b	4.6 ± 0.07 ^c	3.9 ± 0.19 ^d
Day 12	6.4 ± 0.25 ^a	6.1 ± 0.29 ^b	5.9 ± 0.31 ^c	5.5 ± 0.16 ^d

C, G1-P0, G1-P0.5, and G1-P1 are known as control and gelatin and polysaccharide contained coating (with the ratio of 1:0, 1:0.5, and 1:1). Data provided as mean ± standard deviation. The significant difference is shown by superscript letters ($P < 0.05$).

alginate-based coatings preserved fish fillet freshness by reducing pH increase. These findings are consistent with the present study, confirming the potential of natural polysaccharide-based coatings to extend the shelf life of fish products.

The control group (treatment C) exhibited a significantly higher pH increase over 12 days compared to polysaccharide-containing treatments. No significant differences in pH were observed on days 1 and 4 ($P > 0.05$), but significant differences appeared on days 7, 10, and 12 ($P < 0.05$). By day 12, the control sample reached a pH of 7.6, approaching spoilage, whereas G1-P1 remained at 6.9, indicating superior preservation performance.

The lower pH in G1-P1 may be due to the oxygen- and moisture-barrier properties of the coating, which limit microbial metabolism and formation of basic amines (Pastoriza et al., 1996). Additionally, the antibacterial and antioxidant activities of the polysaccharide likely contributed to reduced microbial growth and lipid oxidation.

These findings are supported by Volpe et al. (2015), who observed lower pH increases in trout fillets coated with carrageenan. Similarly, Wang et al. (2018), reported that an edible coating containing tartary buckwheat polysaccharide and nisin helped maintain lower pH in tilapia fillets.

3.2.2.2. Peroxide value (PV). Peroxide values increased during storage in all samples, but treatments containing polysaccharide, especially G1-P1 and G0-P1, exhibited significantly lower oxidation levels compared to the control.

Peroxides and hydroperoxides, which are primary products of lipid oxidation in fish, serve as reliable indicators of spoilage. Fig. 3B shows the peroxide values (PV) of fish fillets coated with different formulations over 12 days of storage at 4 °C. Based on the classification by Varlik (1993), values below 2 mmol O₂/kg are "very good", up to 5 mmol/kg are "good", and 8–10 mmol/kg mark the limit of acceptability.

All treatments showed a gradual increase in PV until day 10, followed by a slight decrease on day 12 due to the degradation of unstable peroxides into secondary oxidation products.

From day 4 onward, significant differences ($P < 0.05$) were observed among treatments. The control group exhibited the highest PVs throughout storage, while G1-P1 consistently maintained the lowest values, followed closely by G0-P1. G1-P0 showed moderate performance, indicating that polysaccharide addition had a more pronounced antioxidant effect than gelatin alone.

By day 12, the peroxide values of G1-P1, G0-P1, G1-P0, and the control were approximately 3.0, 3.4, 3.7, and 4.1 meq/kg, respectively, all of which remained below the spoilage threshold, but with clear distinctions in oxidative stability. PV reflects early lipid oxidation by measuring hydroperoxides. By limiting PV formation, edible coatings preserve freshness and delay rancidity, directly extending fish shelf-life.

The superior performance of polysaccharide-containing coatings is attributed to their antioxidant capacity, which reduces free radical formation and delays lipid oxidation (Ojagh et al., 2010). Similar antioxidant effects were reported by Ojagh et al. (2010) and Fadiloğlu and Emir

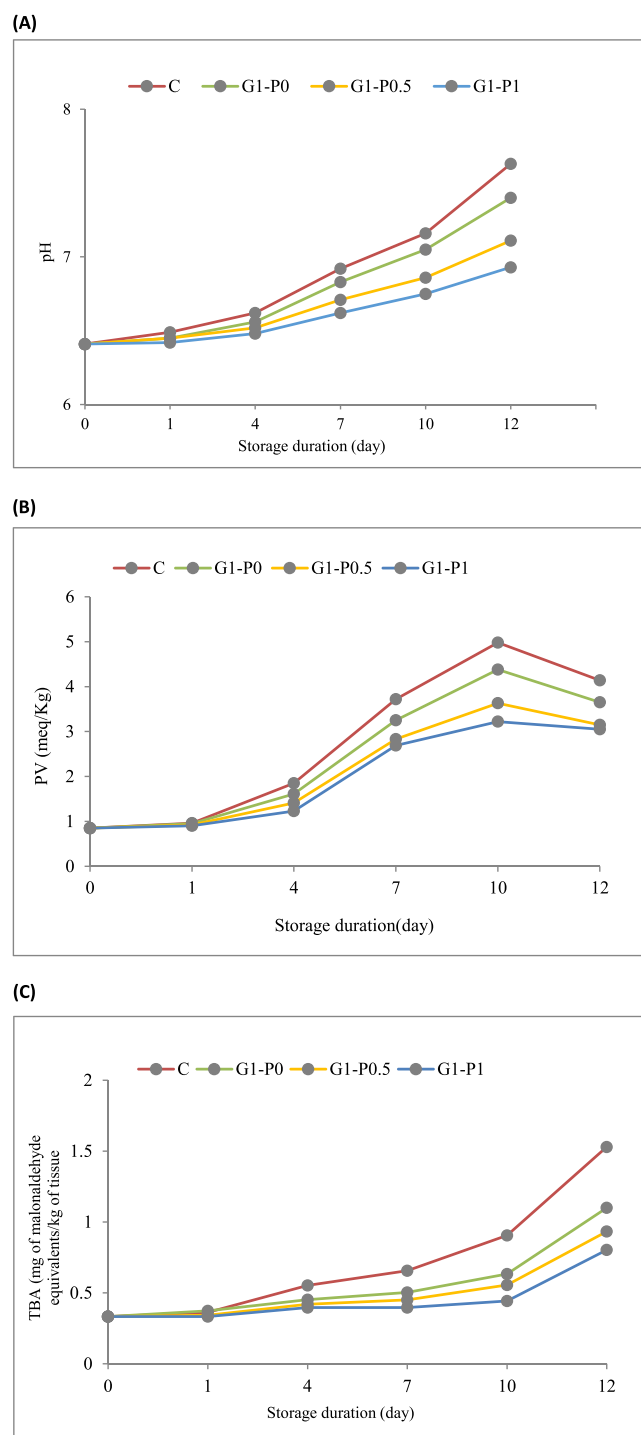


Fig. 3. The level of pH (A), peroxide value (PV) (B), and thiobarbituric acid (TBA) (C) in trout fillets during 12 days of storage at 4°C. C, G1-P0, G1-P0.5, and G1-P1 are known as control and gelatin-polysaccharide-containing coating (with the ratio of 1:0, 1:0.5, and 1:1).

Çoban (2018) using chitosan-based coatings in fish preservation.

3.2.2.3. Thiobarbituric acid (TBA). TBA levels increased throughout storage in all samples, but treatments containing polysaccharide—especially G1-P1 and G0-P1—significantly suppressed malonaldehyde formation compared to the control, indicating reduced lipid oxidation. TBA value reflects the extent of secondary lipid oxidation by quantifying malonaldehyde in fish tissues. As shown in Fig. 3C,

all treatments exhibited a gradual increase in TBA values over 12 days, with a more pronounced rise between days 10 and 12, coinciding with oxidation acceleration.

No significant differences ($P > 0.05$) were observed among treatments during the first two days, but by day 4 and onward, the impact of the coatings became evident ($P < 0.05$). By day 12, the TBA values for G1-P1, G0-P1, G1-P0, and the control were approximately 0.8, 0.9, 1.1, and 1.5 mg/kg, respectively. These values suggest that while all samples remained under the spoilage threshold of 2 mg/kg, G1-P1 and G0-P1 treatments were most effective in delaying oxidative spoilage. TBA quantifies malonaldehyde, a marker of advanced oxidation linked to off-flavors. Lowering TBA maintains sensory and nutritional quality, thus prolonging refrigerated seafood storage.

This protective effect is attributed to the antioxidant capacity of the polysaccharide, which likely neutralizes free radicals and limits the formation of aldehydic compounds. These findings align with Fadiloğlu and Emir Çoban (2018) and Wang et al. (2018), who reported that edible coatings enriched with biopolymers significantly reduce TBA formation in coated fish fillets during cold storage.

3.2.2.4. Free fatty acid (FFA). The accumulation of free fatty acids (FFA) was significantly lower in coated samples particularly G1-P1, which exhibited the lowest FFA values throughout the storage period (3.4 % on day 12 vs. 4.2 % in the control) highlighting the coating's ability to suppress lipid hydrolysis and preserve product freshness.

FFA content reflects the extent of hydrolytic rancidity caused by lipolytic enzymes and microbial activity, leading to off-flavors, texture degradation, and nutritional loss. High FFA levels are a hallmark of spoilage in fatty fish and are closely linked to unpleasant odors and reduced consumer acceptability. By minimizing FFA accumulation, the coatings help maintain the freshness and sensory acceptability of fish during storage. Lower FFA levels are directly associated with extended shelf-life and improved product quality. These results align with the TBA findings, indicating that coatings enriched with antioxidants can delay lipid oxidation and suppress FFA buildup. Similar protective effects of biopolymer-based coatings were reported by Heydari et al. (2015) and Hamzeh and Rezaei (2012) in various fish species.

3.2.2.5. Total volatile basic nitrogen (TVB-N). TVB-N values increased during storage, but the growth was significantly slower in coated samples especially G1-P1 indicating the protective effect of the polysaccharide-based edible films. Total volatile basic nitrogen (TVB-N) are produced by the activity of proteolytic bacteria during chemical and microbial spoilage and are important indicators for assessing the chemical quality of food (Volpe et al., 2015). The acceptable limit of TVB-N in trout fillets is generally reported to be around 25 mg/100 g (Giménez et al., 2002).

In the present study, trout fillets did not exceed the TVB-N limit throughout the storage period, although the control sample approached this limit on day 12. All treatments showed an increasing trend in TVB-N levels during storage, but the increase was much slower in the G1-P1 treatment compared to others. At day 0, no significant differences in TVB-N levels were observed between treatments ($P > 0.05$).

From day 4 onwards, significant differences were detected ($P < 0.05$). The control sample showed the highest TVB-N concentrations, increasing from 10.1 ± 0.09 mg/100 g on day 0 to 23.5 ± 2.81 mg/100 g by day 12. In contrast, the G1-P1 treatment maintained significantly lower TVB-N levels, rising from 10.1 ± 0.09 mg/100 g at day 0 to 14.7 ± 0.88 mg/100 g by day 12. This reduction in TVB-N levels in the G1-P1 treatment can be attributed to the antibacterial and antioxidant properties of the polysaccharide coating, which inhibits bacterial activity and reduces the production of volatile nitrogenous compounds.

These findings align with previous studies; for example, Wang et al. (2018) demonstrated that tilapia fillets coated with a polysaccharide-based edible coating had lower TVB-N levels compared

to uncoated samples, and Heydari et al. (2015) reported that a sodium alginate-based coating reduced TVB-N accumulation in bighead carp during storage.

3.2.2.6. Trimethylamine (TMA). Trimethylamine (TMA) levels progressively increased in all trout fillet samples during refrigerated storage, with significantly lower values in coated samples, particularly those treated with polysaccharide-based formulations. TMA is produced by bacterial enzymatic activity and is associated with the development of

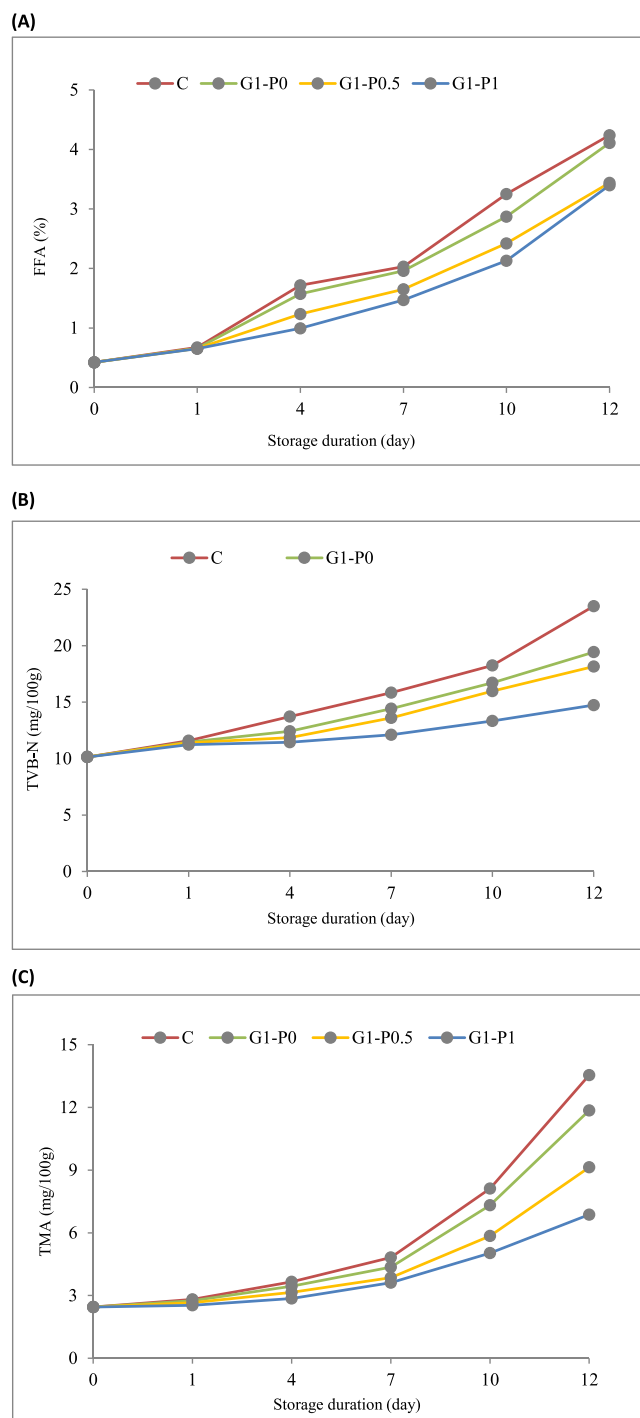


Fig. 4. The level of free fatty acids (FFA) (A), total volatile nitrogen bases (TVB-N) (B), and trimethylamine (TMA) (C) in trout fillets during 12 days of storage at 4°C. C, G1-P0, G1-P0.5, and G1-P1 are known as control and gelatin-polysaccharide-containing coating (with the ratio of 1:0, 1:0.5, and 1:1).

undesirable fishy odors. As shown in Fig. 4C, TMA levels increased steadily over the 12-day storage period. This increase was gradual until day 7, after which a more pronounced rise was observed. No significant differences were detected among treatments until day 7 ($P > 0.05$), but from day 7 onward, significant differences became evident ($P < 0.05$).

Among all treatments, the control group (C) consistently exhibited the highest TMA levels. For instance, on day 12, its TMA concentration reached 13.5 ± 2.01 mg/100 g. In contrast, fillets coated with G1-P1, G1-P2, and G2-P1 formulations demonstrated significantly lower TMA values, indicating a strong inhibitory effect on spoilage-related amine formation. The G1-P1 treatment showed the lowest values overall, with TMA reaching only 6.8 ± 2.74 mg/100 g on day 12, followed closely by G2-P1 at 7.3 ± 1.68 mg/100 g.

These findings highlight the efficacy of polysaccharide-based edible coatings, particularly those with enhanced antioxidant and antimicrobial properties, in delaying TMA production. This is likely due to the inhibition of bacterial enzyme activity and reduced formation of secondary volatile compounds. Similar outcomes have been reported by Günlü and Koyun (2013), who observed suppressed TMA formation in sea bass fillets treated with chitosan-based coatings. Martínez et al. (2018) also found that using edible films containing alginate and chitosan could effectively reduce TMA accumulation during cold storage.

3.2.3. Sensory evaluation

The sensory evaluation results demonstrated that loquat polysaccharide coatings significantly enhanced the sensory quality and shelf life of trout fillets. Tables 2A–2C summarizes the sensory attributes—odor, color, and overall acceptability of coated and uncoated fillets. A sensory score of 4 is regarded as the threshold for consumer acceptance (Fan et al., 2009).

Throughout the 12-day storage, sensory scores declined in all samples, though the rate of decline differed significantly among treatments ($P < 0.05$). Treatments incorporating loquat polysaccharide—especially at higher concentrations exhibited superior odor and overall acceptability scores compared to the control. While the control group fell below the acceptability threshold for odor by day 4, samples such as G1-P1 and G2-P1 maintained odor scores above the threshold until day 12, indicating extended sensory stability.

In terms of color, all samples were acceptable at day 1, with the control group showing slightly better color retention until day 4. However, coated samples such as G1-P2 outperformed the control from day 7 onward, reflecting the protective role of the polysaccharide film against oxidative browning.

The overall acceptability scores declined fastest in the control group, which dropped below the threshold by day 2. In contrast, G1-P1 and G1-P2 retained acceptable levels up to day 7, with a more gradual decline attributed to the antimicrobial and antioxidant properties of the coating.

These findings align with previous studies. Ojagh et al. (2010) demonstrated that a chitosan-based coating extended the shelf life of rainbow trout fillets, and Fadiloğlu and Emir Çoban (2018) showed that chitosan enriched with sumac enhanced the sensory profile of trout fillets during refrigerated storage.

Table 2A

Odor evaluation of trout fillets during 12 days of storage at 4 °C.

Storage time	C	G1-P0	G1-P00.5	G1-P1
Day 0	4.8 ± 0.025^a	4.8 ± 0.025^a	4.8 ± 0.025^a	4.8 ± 0.025^a
Day 1	4.5 ± 0.23^a	4.2 ± 0.64^b	4.5 ± 0.59^a	4.4 ± 0.4^a
Day 4	4 ± 0.15^b	4.5 ± 0.09^a	4.5 ± 0.18^a	4.5 ± 0.38^a
Day 7	3.7 ± 0.25^c	3.9 ± 0.19^b	4 ± 0.17^b	4.2 ± 0.11^a
Day 10	2.5 ± 0.1^c	3.2 ± 0.09^b	3.5 ± 0.08^b	4 ± 0.26^a
Day 12	1 ± 0.21^c	1.5 ± 0.19^b	2.2 ± 0.5^a	2.5 ± 0.24^a

C, G1-P0, G1-P00.5, and G1-P1 are known as control and gelatin and polysaccharide contained coating (with the ratio of 1:0, 1:0.5, and 1:1). (Score 5= extremely desirable; Score 1= off-odors). Data provided as mean \pm standard deviation. The significant difference is shown by superscript letters ($P < 0.05$).

Table 2B

Discoloration of trout fillets during 12 days of storage at 4 °C.

Storage time	C	G1-P0	G1-P00.5	G1-P1
Day 0	4.4 ± 0.014^a	4.4 ± 0.014^a	4.4 ± 0.014^a	4.4 ± 0.014^a
Day 1	4 ± 0.08^a	3.6 ± 0.1^b	3.5 ± 0.14^b	3.3 ± 0.08^c
Day 4	3.8 ± 0.11^a	3.5 ± 0.4^b	3.5 ± 0.24^b	3.4 ± 0.21^b
Day 7	3 ± 0.05^b	2.9 ± 0.11^b	3.2 ± 0.19^a	3.3 ± 0.2^a
Day 10	3 ± 0.28^c	3.1 ± 0.12^b	3.1 ± 0.5^b	3.3 ± 0.22^a
Day 12	1.5 ± 0.23^c	1.8 ± 0.14^b	3 ± 0.14^a	3.1 ± 0.31^a

C, G1-P0, G1-P00.5, and G1-P1 are known as control and gelatin and polysaccharide contained coating (with the ratio of 1:0, 1:0.5, and 1:1). (Score 5= no discoloration; Score 1= extreme discoloration). Data provided as mean \pm standard deviation. The significant difference is shown by superscript letters ($P < 0.05$).

Table 2C

Overall acceptability of trout fillets during 12 days of storage at 4 °C.

Storage time	C	G1-P0	G1-P00.5	G1-P1
Day 0	4.6 ± 0.016^a	4.6 ± 0.016^a	4.6 ± 0.016^a	4.6 ± 0.016^a
Day 1	4 ± 0.14^b	4 ± 0.05^b	4.2 ± 0.07^a	4.2 ± 0.15^a
Day 4	3.5 ± 0.2^c	3.5 ± 0.16^c	3.7 ± 0.12^b	4 ± 0.12^a
Day 7	3.2 ± 0.18^c	3.4 ± 0.15^b	3.5 ± 0.22^b	3.9 ± 0.06^a
Day 10	2.8 ± 0.31^c	3 ± 0.14^b	3.1 ± 0.13^a	3.2 ± 0.14^a
Day 12	1.5 ± 0.05^c	2.2 ± 0.09^b	2.3 ± 0.15^b	2.5 ± 0.14^a

C, G1-P0, G1-P00.5, and G1-P1 are known as control and gelatin and polysaccharide contained coating (with the ratio of 1:0, 1:0.5, and 1:1). (Score 5= extremely desirable; Score 1= extremely unacceptable). Data provided as mean \pm standard deviation. The significant difference is shown by superscript letters ($P < 0.05$).

4. Conclusion

Polysaccharide was successfully extracted from loquat (*Eriobotrya japonica*) seed using the microwave at various powers (180, 360, and 580 W). The superior recovery yield of polysaccharide (48.2 ± 0.53 %) was obtained by applying 580 W microwave result, which was selected for use in the gelatin-based edible coating, as it also exhibited outstanding antioxidant activities including DPPH and HO radical scavenging activity along with excellent ferric-reducing power. Rainbow trout (*Oncorhynchus mykiss*) fillets were coated on gelatin-containing edible coatings plus different ratios of polysaccharide (G1-P0, G1-P00.5, and G1-P1), and kept for 12 days at 4 °C. Use of the highest ratio of polysaccharides (G1-P1) resulted in delaying the microbial spoilage of fillets by diminishing the growth rate of both total and psychrophilic bacteria. The chemical deterioration rate of fish fillets including the production of peroxide (PV), thiobarbituric acid (TBA), free fatty acids (FFA), total volatile basic nitrogen (TVB-N), and trimethylamine (TMA) was also slowed down by elevating the ratio of polysaccharide in the coating, resulting from the antioxidant power of polysaccharide. Additionally, use of a similar ratio of gelatin and polysaccharide in the edible coating (G1-P1) prolonged the sensory acceptance of the trout fillets in three different factors including odor, color, and overall acceptability, which resulted in enhancing the shelf life of trout fillets for human consumption. Accordingly, the gelatin-based edible coating enriched with loquat polysaccharide was suggested as an eco-friendly, economical, and safe preservation method for prolonging the shelf life of rainbow trout fillets in cold storage, which can be further used with commercial aims.

CRedit authorship contribution statement

Mojtaba Nouri: Writing – original draft, Visualization, Resources, Project administration, Methodology, Investigation. **Homa Baghaei:** Project administration, Formal analysis, Data curation, Conceptualization. **Mahdi Kashaninejad:** Writing – review & editing, Software, Methodology, Investigation, Formal analysis. **Abdorrezza Mohammadi**

nafchi: Writing – review & editing, Visualization, Supervision, Software, Resources, Formal analysis.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Ethical statement

This study did not involve any experiments on human participants or animals. Therefore, ethical approval from an institutional review board or animal care and use committee was not required. The research was conducted entirely on food materials, and all experimental procedures were designed and carried out in compliance with institutional policies and internationally accepted standards for responsible research conduct.

Data availability

Data will be made available on request.

References

- Aguiñe-Joya, J.A., De Leon-Zapata, M.A., Alvarez-Perez, O.B., Torres-León, C., Nieto-Oropeza, D.E., Ventura-Sobrevilla, J.M., Aguilar, M.A., Ruelas-Chacón, X., Rojas, R., Ramos-Aguinaga, M.E., 2018. Basic and applied concepts of edible packaging for foods. *Food Packaging and Preservation*. Elsevier, pp. 1–61.
- Alboofetileh, M., Rezaei, M., Tabarsa, M., You, S.G., 2019. Bioactivities of Nizamuddinina zanardinii sulfated polysaccharides extracted by enzyme, ultrasound and enzyme-ultrasound methods. *J. Food Sci. Technol.* 56 (3), 1212–1220. <https://doi.org/10.1007/s13197-019-03584-1>.
- Alsulami, M.N., El-Saadony, M.T., 2024. The enhancing effect of bacterial zinc nanoparticles on performance, immune response, and microbial load of Nile tilapia (*Oreochromis niloticus*) by reducing the infection by trichodina heterodontata. *Pak. Vet. J.* 44 (3), 599–610. <https://doi.org/10.29261/pakvetj/2024.243>.
- Anshar, A.R., Relatami, A.N.R., Amriani, R., Chadajah, A., Rahmi, Tampangallo, B.R., Septiani, R.R., Hoven, I.D., Agustina, K.K., Nurdin, M.A., 2023. The effect of natural (*Daphnia* sp.) and different synbiotic feed on rainbow celebes (*Marosatherina ladiges*) growth performance and survival rate. *Int. J. Vet. Sci.* 12 (5), 730–735.
- AOAC, 1995. official methods of analysis. 16th ed. Washington, DC Association of Official. Off. Methods Anal. AOAC Int. (16th Edn) 6 (11), 382.
- Bougatef, H., Krichen, F., Capitani, F., Amor, I.Ben, Gargouri, J., Maccari, F., Mantovani, V., Galeotti, F., Volpi, N., Bougatef, A., Sila, A., 2019. Purification, compositional analysis, and anticoagulant capacity of chondroitin sulfate/dermatan sulfate from bone of corb (*Sciaenops ocellatus*). *Int. J. Biol. Macromol.* 134, 405–412. <https://doi.org/10.1016/j.ijbiomac.2019.05.036>.
- Brasil, I.M., Gomes, C., Puerta-Gomez, A., Castell-Perez, M.E., Moreira, R.G., 2012. Polysaccharide-based multilayered antimicrobial edible coating enhances quality of fresh-cut papaya. *Lwt* 47 (1), 39–45. <https://doi.org/10.1016/j.lwt.2012.01.005>.
- Bruno, S.F., Ekorong, F.J.A.A., Karkal, S.S., Cathrine, M.S.B., Kudre, T.G., 2019. Green and innovative techniques for recovery of valuable compounds from seafood by-products and discards: a review. *Trends Food Sci. Technol.* 85 (June 2018), 10–22. <https://doi.org/10.1016/j.tifs.2018.12.004>.
- Campos, S., Doxey, J., Hammond, D., 2011. Nutrition labels on pre-packaged foods: a systematic review. *Public Health Nutr.* 14 (8), 1496–1506.
- Chen, R., Li, S., Liu, C., Yang, S., Li, X., 2012. Ultrasound complex enzymes assisted extraction and biochemical activities of polysaccharides from *Epimedium* leaves. *Process Biochem.* 47 (12), 2040–2050. <https://doi.org/10.1016/j.procbio.2012.07.022>.
- Chew, Y.L., Lim, Y.Y., Omar, M., Khoo, K.S., 2008. Antioxidant activity of three edible seaweeds from two areas in South East Asia. *LWT - Food Sci. Technol.* 41 (6), 1067–1072. <https://doi.org/10.1016/j.lwt.2007.06.013>.
- Díaz-Montes, E., 2022. Polysaccharides: sources, characteristics, properties, and their application in biodegradable films. *Polysaccharides* 3 (3), 480–501. <https://doi.org/10.3390/polysaccharides3030029>.
- Dursun, S., Erkan, N., 2014. The effect of edible coating on the quality of smoked fish. *Ital. J. Food Sci.* 26 (4), 370–382.
- Fadıloğlu, E.E., Emir Coban, Ö., 2018. Effects of chitosan edible coatings enriched with sumac on the quality and the shelf life of rainbow trout (*Oncorhynchus mykiss*, Walbaum, 1792) filets. *J. Food Saf.* 38 (6), e12545.
- Fan, Y., He, X., Zhou, S., Luo, A., He, T., Chun, Z., 2009. Composition analysis and antioxidant activity of polysaccharide from *dendrobium denneanum*. *Int. J. Biol. Macromol.* 45 (2), 169–173.
- Fu, Y., Li, F., Ding, Y., Li, H.Y., Xiang, X.R., Ye, Q., Zhang, J., Zhao, L., Qin, W., Gan, R.Y., Wu, D.T., 2020. Polysaccharides from loquat (*Eriobotrya japonica*) leaves: impacts of extraction methods on their physicochemical characteristics and biological activities. *Int. J. Biol. Macromol.* 146, 508–517. <https://doi.org/10.1016/j.ijbiomac.2019.12.273>.
- Fu, Y., Yuan, Q., Lin, S., Liu, W., Du, G., Zhao, L., Zhang, Q., Lin, D.R., Liu, Y.T., Qin, W., Li, D.Q., Wu, D.T., 2019. Physicochemical characteristics and biological activities of polysaccharides from the leaves of different loquat (*Eriobotrya japonica*) cultivars. *Int. J. Biol. Macromol.* 135, 274–281. <https://doi.org/10.1016/j.ijbiomac.2019.05.157>.
- García, M.A., Lopez, P., Martínez, A., et al., 2021. Alginate edible coatings to extend shelf life and maintain quality of refrigerated fish filets. *Int. J. Food Sci. Technol.* 56 (2), 757–765. <https://doi.org/10.1111/ijfs.14732>.
- Giménez, B., Roncalés, P., Beltrán, J.A., 2002. Modified atmosphere packaging of filleted rainbow trout. *J. Sci. Food Agric.* 82 (10), 1154–1159.
- Guerreiro, A.C., Gago, C.M.L., Faleiro, M.L., Miguel, M.G.C., Antunes, M.D.C., 2015. The use of polysaccharide-based edible coatings enriched with essential oils to improve shelf-life of strawberries. *Postharvest Biol. Technol.* 110, 51–60. <https://doi.org/10.1016/j.postharvbio.2015.06.019>.
- Günlü, A., Koyun, E., 2013. Effects of vacuum packaging and wrapping with Chitosan-based edible film on the extension of the shelf life of sea bass (*Dicentrarchus labrax*) filets in cold storage (4 °C). *Food Bioproc. Tech.* 6 (7), 1713–1719. <https://doi.org/10.1007/s11947-012-0833-6>.
- Hamzeh, A., Rezaei, S., 2012. The effects of sodium alginate on quality of rainbow trout (*Oncorhynchus mykiss*) filets stored at 4±2 °C. *J. Aquat. Food Prod. Technol.* 21 (1), 14–21.
- He, R., Zhao, Y., Mu, J., et al., 2022. Extraction, structural characterization and biological activities of polysaccharides from *Eriobotrya japonica* seeds. *Carbohydr. Polym.* 278, 118974. <https://doi.org/10.1016/j.carbpol.2021.118974>.
- Heidari, M.G., Rezaei, M., 2022. Extracted pepsin of trout waste and ultrasound-promoted method for green recovery of fish collagen. *Sustain. Chem. Pharm.* 30 (June), 100854. <https://doi.org/10.1016/j.scp.2022.100854>.
- Heydari, R., Bavandi, S., Javadian, S.R., 2015. Effect of sodium alginate coating enriched with horsemint (*Monarda longifolia*) essential oil on the quality of bighead carp filets during storage at 4 °C. *Food Sci. Nutr.* 3 (3), 188–194.
- Jasour, M.S., Ehsani, A., Mehryar, L., Naghibi, S.S., 2015. Chitosan coating incorporated with the lactoperoxidase system: an active edible coating for fish preservation. *J. Sci. Food Agric.* 95 (6), 1373–1378. <https://doi.org/10.1002/jsfa.6838>. Chitosan coating inco. *Journal of the Science of Food and Agriculture*, 95(6), 1373–1378.
- Jiang, Y., Wang, L., Zhang, L., Wang, T., Yu, L., Ding, C., Yang, R., Wang, X., Zhou, Y., 2014. Characterization, antioxidant and antitumor activities of polysaccharides from *Salvia miltiorrhiza* Bunge. *Int. J. Biol. Macromol.* 70, 92–99.
- Jiao, G., Yu, G., Zhang, J., Ewart, H.S., 2011. Chemical structures and bioactivities of sulfated polysaccharides from marine algae. *Mar. Drugs* 9 (2), 196–223.
- Johns, K., Stead, G., 2002. Supercritical fluids for coatings—From analysis to xenon: a brief overview. *Fluoropolymers 2: Prop.* 35–45.
- Jonusaite, K., Venskutonis, P.R., Martínez-Hernández, G.B., Taboada-Rodríguez, A., Nieto, G., López-Gómez, A., Marín-Iniesta, F., 2021. Antioxidant and antimicrobial effect of plant essential oils and sambucus nigra extract in salmon burgers. *Foods* 10 (4), 776.
- Kaufmann, B., Christen, P., 2002. Recent extraction techniques for natural products: microwave-assisted extraction and pressurized solvent extraction. *Phytochem. Anal.: Int. J. Plant Chem. Biochem. Tech.* 13 (2), 105–113.
- Kazemian-Bazkiaee, F., Ebrahimi, A., Hosseini, S.M., Shojaaee-Aliabadi, S., Farhoodi, M., Rahmatzadeh, B., Sheikhi, Z., 2020. Evaluating the protective effect of edible coatings on lipid oxidation, fatty acid composition, aflatoxins levels of roasted peanut kernels. *J. Food Meas. Charact.* 14, 1025–1038.
- Körge, K., Seme, H., Bajić, M., Likoar, B., Novak, U., 2020. Reduction in spoilage microbiota and cyclopiazonic acid mycotoxin with chestnut extract enriched chitosan packaging: stability of inoculated gouda cheese. *Foods* 9 (11), 1645.
- Lee, H., Kim, Y.-K., Lee, J.-J., 2015. A comparison of nutritional components of loquat (*Eriobotrya japonica* Lindl.) powder in different aerial components. *Korean J. Community Living Sci.* 26 (3), 541–549.
- Li, Q., Li, J., Li, H., Xu, R., Yuan, Y., Cao, J., 2019. Physicochemical properties and functional bioactivities of different bonding state polysaccharides extracted from tomato fruit. *Carbohydr. Polym.* 219 (March), 181–190. <https://doi.org/10.1016/j.carbpol.2019.05.020>.
- Li, X., Wang, L., Li, Z., Zhao, Q., Zhou, Z., Zhang, L., 2020. Extraction, purification, structural characterization and antioxidant activities of polysaccharides from loquat (*Eriobotrya japonica*) seed. *Int. J. Biol. Macromol.* 161, 158–166. <https://doi.org/10.1016/j.ijbiomac.2020.05.179>.
- Li, H., Yu, X., Jin, Y., Zhang, W., 2017. Vibrational spectroscopy characterization and anticoagulant activity of a sulfated polysaccharide from sea cucumber *Athyronidium chilensis*. *Carbohydr. Polym.* 157, 1591–1599. <https://doi.org/10.1016/j.carbpol.2016.11.007>.
- Li, J., Zhang, Y., Wang, Y., et al., 2022. Antimicrobial edible coatings based on polysaccharides for fish preservation: a review. *Food Packag. Shelf Life* 33, 100901. <https://doi.org/10.1016/j.fpsl.2022.100901>.
- Liu, Y., Zhang, W., Xu, C., Li, X., 2016. Biological activities of extracts from loquat (*Eriobotrya japonica* Lindl.): a review. *Int. J. Mol. Sci.* 17 (12), 1983.
- Liu, Z., Dang, J., Wang, Q., Yu, M., Jiang, L., Mei, L., Shao, Y., Tao, Y., 2013. Optimization of polysaccharides from *Lycium ruthenicum* fruit using RSM and its anti-oxidant activity. *Int. J. Biol. Macromol.* 61, 127–134.
- Manzoor, K., Rasool, F., Khan, N., Anjum, K.M., Parveen, S., 2023. Prevalence and molecular detection of *Edwardsiella ictaluri* in cultured tilapia species of fish farms of Punjab in Pakistan and their postmortem examination. *Pak. Vet. J.* 43, 309–314. <https://doi.org/10.29261/pakvetj/2023.015>.
- Martínez, O., Salmerón, J., Epelde, L., Vicente, M.S., de Vega, C., 2018. Quality enhancement of smoked sea bass (*Dicentrarchus labrax*) filets by adding resveratrol and coating with chitosan and alginate edible films. *Food Control* 85, 168–176. <https://doi.org/10.1016/j.foodcont.2017.10.003>.

- Meng, Q., Li, Y., Xiao, T., Zhang, L., Xu, D., 2017. Antioxidant and antibacterial activities of polysaccharides isolated and purified from *Diaphragma juglandis fructus*. *Int. J. Biol. Macromol.* 105, 431–437.
- Natseba, A., Lwalinda, I., Kakura, E., Muya, C.K., Muya, J.H., 2005. Effect of pre-freezing icing duration on quality changes in frozen Nile perch (*Lates niloticus*). *Food Res. Int.* 38 (4), 469–474.
- Ojagh, S.M., Rezaei, M., Razavi, S.H., Hosseini, S.M.H., 2010. Effect of chitosan coatings enriched with cinnamon oil on the quality of refrigerated rainbow trout. *Food Chem.* 120 (1), 193–198. <https://doi.org/10.1016/j.foodchem.2009.10.006>.
- Okolie, C.L., Akanbi, T.O., Mason, B., Udenigwe, C.C., Aryee, A.N.A., 2019. Influence of conventional and recent extraction technologies on physicochemical properties of bioactive macromolecules from natural sources: a review. *Food Res. Int.* 116, 827–839.
- Pastoriza, L., Sampedro, G., Herrera, J.J., Cabo, M.L., 1996. Effect of modified atmosphere packaging on shelf-life of iced fresh hake slices. *J. Sci. Food Agric.* 71 (4), 541–547.
- Ren, B., Chen, C., Li, C., Fu, X., You, L., Liu, R.H., 2017. Optimization of microwave-assisted extraction of *Sargassum thunbergii* polysaccharides and its antioxidant and hypoglycemic activities. *Carbohydr. Polym.* 173, 192–201.
- Ren, Y., Bai, Y., Zhang, Z., Cai, W., Del Rio Flores, A., 2019. The preparation and structure analysis methods of natural polysaccharides of plants and fungi: a review of recent development. *Molecules* 24 (17). <https://doi.org/10.3390/molecules24173122>.
- Rezaei, F., Shahbazi, Y., 2018. Shelf-life extension and quality attributes of sauced silver carp fillet: a comparison among direct addition, edible coating and biodegradable film. *Lwt* 87, 122–133. <https://doi.org/10.1016/j.lwt.2017.08.068>.
- Rizfa, A., Ben Hassine, D., Gharbi, Y., 2020. Thermal and chemical modifications of sodium alginate extracted from *Sargassum* species: effects on antioxidant activity and hydroxyl radical scavenging capacity. *Int. J. Biol. Macromol.* 150, 1234–1242. <https://doi.org/10.1016/j.ijbiomac.2020.01.123>.
- Rostami, H., Gharibzadeh, S.M.T., 2016. Microwave-assisted extraction of jujube polysaccharide: optimization, purification and functional characterization. *Carbohydr. Polym.* 143, 100–107.
- Salehi, F., 2020. Edible coating of fruits and vegetables using natural gums: a review. *Int. J. Fruit Sci.* 20 (S2), S570–S589. <https://doi.org/10.1080/15538362.2020.1746730>.
- Senphan, T., Benjakul, S., 2015. Impact of enzymatic method using crude protease from Pacific white shrimp hepatopancreas on the extraction efficiency and compositions of lipids. *Food Chem.* 166, 498–506.
- Sharma, A., 2021. High entropy alloy coatings and technology. *Coatings* 11 (4), 372.
- Shi, L., 2016. Bioactivities, isolation and purification methods of polysaccharides from natural products: a review. *Int. J. Biol. Macromol.* 92, 37–48.
- Siang, N.C., Kim, L.L., 1992. Determination of TMAON, TMA-N, TVB-N by Conway's microdiffusion methods (1 % boric acid and 0.02 N HCl) pH-8.1-B-8.5. In: *Lab. Manual on Analytical Methods and Procedures for Fish and Fishery Products*. 2nd Mar. Fish. Res. Dep. SEAFDEC, Singapore.
- Suhag, R., Kumar, N., Petkoska, A.T., Upadhyay, A., 2020. Film formation and deposition methods of edible coating on food products: a review. *Food Res. Int.* 136 (March), 109582. <https://doi.org/10.1016/j.foodres.2020.109582>.
- Tahir, H.E., Xiaobo, Z., Mahunu, G.K., Arslan, M., Abdalhai, M., Zhihua, L., 2019. Recent developments in gum edible coating applications for fruits and vegetables preservation: a review. *Carbohydr. Polym.* 224 (March), 115141. <https://doi.org/10.1016/j.carbpol.2019.115141>.
- Thirugnanasambandham, K., Sivakumar, V., Maran, J.P., 2015. Microwave-assisted extraction of polysaccharides from mulberry leaves. *Int. J. Biol. Macromol.* 72, 1–5. <https://doi.org/10.1016/j.ijbiomac.2014.07.031>.
- Varlik, C., 1993. Su urunlerinde kalite kontrol ilke ve yontemleri. *Gida Teknol. Dern.* 17, 16–17.
- Volpe, M.G., Siano, F., Paolucci, M., Sacco, A., Sorrentino, A., Malinconico, M., Varicchio, E., 2015. Active edible coating effectiveness in shelf-life enhancement of trout (*Oncorhynchus mykiss*) filets. *Lwt* 60 (1), 615–622. <https://doi.org/10.1016/j.lwt.2014.08.048>.
- Wang, F., Zhang, H., Jin, W., Li, L., 2018. Effects of tartary buckwheat polysaccharide combined with nisin edible coating on the storage quality of tilapia (*Oreochromis niloticus*) filets. *J. Sci. Food Agric.* 98 (8), 2880–2888.
- Wang, J., Zhang, J., Wang, X., Zhao, B., Wu, Y., Yao, J., 2009. A comparison study on microwave-assisted extraction of *Artemisia sphaerocephala* polysaccharides with conventional method: molecule structure and antioxidant activities evaluation. *Int. J. Biol. Macromol.* 45 (5), 483–492.
- Wang, L., Zhang, D., Ma, H., et al., 2020. Effects of chitosan coating on quality and shelf life of refrigerated fish filets. *LWT - Food Sci. Technol.* 126, 109273. <https://doi.org/10.1016/j.lwt.2020.109273>.
- Wu, D., Zhang, W., Fu, B., Hu, G., 2019. Chemical structure and gas products of different rank coals during pyrolysis: based on in-situ FTIR and TG/MS analysis techniques. *J. Therm. Anal. Calorim.* 136, 2017–2031.
- Xiong, F., Li, X., Zheng, L., Hu, N., Cui, M., Li, H., 2019. Characterization and antioxidant activities of polysaccharides from *Passiflora edulis* Sims peel under different degradation methods. *Carbohydr. Polym.* 218, 46–52.
- Xue, Y., Sui, Q., Brusseu, M.L., Zhang, X., Qiu, Z., Lyu, S., 2018. Insight on the generation of reactive oxygen species in the $\text{CaO}_2/\text{Fe}(\text{II})$ Fenton system and the hydroxyl radical advancing strategy. *Chem. Eng. J.* 353, 657–665.
- Yu, M., Chen, M., Gui, J., Huang, S., Liu, Y., Shentu, H., He, J., Fang, Z., Wang, W., Zhang, Y., 2019. Preparation of *Chlorella vulgaris* polysaccharides and their antioxidant activity in vitro and in vivo. *Int. J. Biol. Macromol.* 137, 139–150.
- Yuan, Y., Xu, X., Jing, C., Zou, P., Zhang, C., Li, Y., 2018. Microwave assisted hydrothermal extraction of polysaccharides from *Ulva prolifera*: functional properties and bioactivities. *Carbohydr. Polym.* 181, 902–910.
- Zhang, Y., Liu, Y., Li, X., et al., 2020. Polysaccharides from loquat (*Eriobotrya japonica*) leaves: impacts of extraction methods on their physicochemical characteristics and biological activities. *Food Chem.* 309, 125711. <https://doi.org/10.1016/j.foodchem.2019.125711>.
- Zhong, K., Lin, W., Wang, Q., Zhou, S., 2012. Extraction and radicals scavenging activity of polysaccharides with microwave extraction from mung bean hulls. *Int. J. Biol. Macromol.* 51 (4), 612–617.
- Zhou, X., Liu, Y., Chen, H., et al., 2021. Effect of natural antimicrobial coatings on the quality and shelf life of fish filets during refrigerated storage. *Food Hydrocoll.* 111, 106329. <https://doi.org/10.1016/j.foodhyd.2020.106329>.
- Zhu, L., Qi, H., Kong, Y., Yu, Y., Xu, X., 2012. Component analysis of extracellular polymeric substances (EPS) during aerobic sludge granulation using FTIR and 3D-EEM technologies. *Bioresour. Technol.* 124, 455–459.