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REVIEW PAPER

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Fish scales chitosan: Extraction, characterization and applications: A review

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Abstract

The rapid growth in global fisheries production has led to a notable increase in fish scale waste, which remains largely underutilized despite containing valuable bioactive molecules like chitosan. This study presents a systematic review of seventeen peer-reviewed articles published from 2014 to 2024, focusing on the extraction, characterization, and application of chitosan derived from fish scales. The review highlights a growing research interest in extraction techniques, which primarily involve chemical processes such as deproteination, demineralization, and deacetylation. Variations in extraction methods across different fish species, including *Lutjanus* spp., *Channa striata*, and Tilapia, have resulted in differing chitosan yields and qualities. Key findings reveal that extraction parameters significantly influence chitosan's chemical structure, morphology, and mechanical properties. Characterization techniques, including FTIR and SEM, confirmed successful deacetylation and provided detailed insights into the structural attributes of chitosan. The findings suggest that chitosan from fish scales has a broad range of potential applications, including food preservation, wound care, antibacterial agents, and environmental management. To improve chitosan extraction and application, this review recommends standardizing extraction methods, optimizing process parameters for consistency, and further investigating species-specific advantages. Enhanced characterization techniques are also crucial for advancing chitosan quality and expanding its industrial applications.

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Introduction

The growth of the global population and the subsequent rapid increase in urbanization and industrialization, the fisheries sector production was noticed a massive increase driven mainly by the development of fishing technologies. Consequently, there is remarkable increase in the amount of fish waste that has been produced around the world; it has been estimated that about two-thirds of the total amount of fish is discarded as waste, creating huge economic and environmental concerns. In South-East Asia, there is 1.5 million tons of waste generated (FAO, 2021) Meanwhile, the Philippine has estimated 20-30 percent fish processing loss or waste due to improper post -harvesting. In fact, processing activities generate 20- 80 percent of solid waste, such as head, guts, viscera – including liver and roe – the bones, fins, scales, and skin (Islam *et al.*, 2021). On the other hand, the growing production of milkfish, and tilapia industries at 300,000 tons (Guerrero and Guerrero, 2004; Yap *et al.*, 2007) have their consequent scale waste (Cadano *et al.*, 2020) which has low economic value or almost having no cost. Consequently, Scale fish scale waste get spoiled quickly by enzymatic and bacteriological processes which can create environmental contamination, economic burden, and health hazards as well as resulting in the loss of valuable compounds (Kumari *et al.*, 2016. However, these wastes possess potential bioactive molecules that are potential for utilization particularly production of valuable and biologically sustainable materials for industrial applications and current research and development (Srivastava *et al.*, 2018).

Several uses of fish scales have been reported by several researchers. However, the investigation of its potential as a new raw material for chitosan production deserves to be highlighted, especially because they may be used as an alternative for people allergic to shrimp and other crustaceans. Commercial production of chitosan from shrimp and crab shells requires high production cost and multiple chemical processes such as demineralization, deproteinization, and decolorization. Therefore, fish scale waste is another low-cost raw material for chitosan production and can be one solution dealing with environmental problem. Ooi *et al.*, 2021 concluded that fish scale can be used as alternative sources of chitosan extraction and production. However, Takarina and Fanani (2017) mentioned that chitosan extracted from saltwater fishes is rare.

Chitosan is a natural product which is a derivative of the polysaccharide chitin. Chitosan has the chemical name Poly D-glucosamine (beta (1-4) 2-amino-2-deoxy D-glucose. Chitosan have become materials of great interest not only as an underutilized resource but also as a new functional biomaterial of high potential in various fields. Its physicochemical and biological characteristics, such as biocompatibility, biodegradability, nontoxicity, physiological inertness, immunological activity, antibacterial properties, wound-healing activity, heavy metal ions chelation, gelforming properties and hydrophilicity, and affinity to protein. These properties make chitosan appealing materials for a variety of applications. Their applications are widely distributed in many fields, such as wastewater treatment, biotechnology, medical and pharmaceutical, food industry, agriculture, cosmetics and pulp and paper industries (Kumari *et al.*, 2017).

Besides the series of advantages, benefits, and applications of chitosan, it holds great promise the quantity and quality of chitosan produced is still major problems as the quantity produced is not enough for industrial application while the quality of chitosan. Produced is not satisfactor (Adenkanmi *et al.*, 2020). Hence, it requires the optimization on extraction from fish waste to maximize the yield. In fact, standardization of parameters is quite essential for the optimization of the process.

Classical techniques of optimization involve the study of one parameter keeping the other constant, which gives a little understanding of the overall effect of various parameters on the quality of the product. Moreover, it requires more time and less accuracy. Computed standardization using response surface methodology can be a suitable solution to study the multi-parametric effect on a single or multiple responses (Datta *et al.*, 2021). To cite, in the study Cadano *et al.*, 2020 on extraction of chitosan, they recommended that chitosan yields from bio-waste in the Philippine seafood that includes milkfish and tilapia scales are recommended for improvement particularly on the procedure.

Hence, the aim of this work is to explore the extraction and characterization of Chitosan from Fish Scales and identify its applications thru a systematic review, a review of published literature related to the chosen topic

Materials and methods

This study utilized a total of seventeen (17) research articles focused on the extraction of chitosan from fish scales. The articles reviewed included those that covered the characterization of extracted chitosan, various extraction methods or techniques, and the applications of fish scale-derived chitosan. These 17 articles were selected from an initial pool of over fifty (50) articles downloaded from a variety of academic journals.

Selection criteria

The selection of articles for this systematic review was guided by specific inclusion criteria, which ensured the relevance and quality of the studies analyzed. To qualify, the articles had to be peer-reviewed and published research papers, specifically within the timeframe of 2014 to 2024.

Only articles written in English were considered. Additionally, the content of each article needed to focus on the extraction of chitosan from fish scales, regardless of the fish species involved.

Furthermore, the articles had to discuss the methods or techniques used in the extraction process, as well as explore the applications and uses of chitosan derived from fish scales.

Articles were excluded based on several criteria to maintain the focus and relevance of the review. Specifically, any articles published before 2014 or after 2024 were excluded from consideration. Additionally, articles not written in English were also excluded. The review only considered articles with full manuscripts available, so those without full access were not included. Furthermore, studies discussing the extraction of chitosan from sources other than fish scales were excluded, as were any studies that did not align with the stated objectives of this review.

The selection criteria used in this study were adapted from the methodologies described by Retuya *et al.* (2020).

Data collection procedure

A systematic review approach was employed to collect and analyze data. The literature review aimed to compile and critically analyze information on the extraction and characterization of chitosan from fish scales, as well as its applications.

Articles were identified and downloaded through the Google Scholar search engine, focusing on journals relevant to the topic. Each downloaded article was assigned a unique code and stored in a designated folder. Screening of the articles was conducted using the defined inclusion and exclusion criteria. Articles meeting the inclusion criteria were saved in a separate folder for further analysis, while those not meeting the criteria were documented in a separate folder.

After screening, the selected articles were thoroughly reviewed. Key information extracted included the types and characteristics of chitosan derived from fish scales, the fish species used as sources of scales, the extraction techniques and methods employed, and the applications of the extracted chitosan.

Results and discussion

Profile of reviewed published articles (n=17)

Table 1 presents the detailed profiles of the seventeen (17) research articles included in this systematic review. Each article adheres to the established inclusion criteria: they are peer-reviewed, published between 2014 and 2024, written in English, and focus on the extraction of chitosan from fish scales. Additionally, these articles provide insights into the methods or techniques employed in the extraction process and explore the applications and uses of chitosan derived from fish scales. The data further reveal that most of these articles have been published from 2020 onwards, indicating a recent surge in research interest in this area. This trend suggests that the exploration of chitosan extraction from fish scales is a relatively new field, which may account for the limited number of published studies on this topic.

Table 1. Individual profile of reviewed published articles related to extraction of chitosan from fish and its applications

Code	Author	Article	Year of publication	Journal
001	A. Florencia, S. E. D. Putra, Y. P. Mukti	Chitosan from snapper fish scale waste (*Lutjanus* spp.) for edible coating	2022	IOP Conf. Series: Earth and Environmental Science
002	Agus Susanto, Mieke Hemiawati Satari, Basril Abbas, R. Setyo Adji Koesoemowidodo, Arief Cahyanto	Fabrication and Characterization of Chitosan-Collagen Membrane from Barramundi (*Lates calcarifer*) Scales for Guided Tissue Regeneration	2019	European Journal of Dentistry
003	Dewi Retno Wahyu Widyaningrum, Deby Kania Tri Putri, Irham Taufiqurrahman	Antibacterial Activities of Chitosan in Haruan Fish Scales (*Channa striata*) to the Growth of Staphylococcus aureus	2019	Dentistry,
004	Noverita D. Takarina, Aldila A. Nasrul, Alinda Nurmarina	Degree of Deacetylation of Chitosan Extracted from White Snapper (*Lates* sp.) Scales Waste	2017	International Journal of Pharma Medicine and Biological Sciences
005	Pragathi A. H, C. M. Noorjahan	Determination of Biochemical Components of Fish Scales and Antibacterial Activity of Chitosan Extracted from Fish Scales	2023	Indian Journal of Natural Sciences
006	Gul-e-Saba Chaudhry *et al.*	Antibacterial activity of Tilapia Fish Scales derived Chitosan; Future towards Biomedicines	2022	Research J. Pharm. and Tech.
007	Femiana Gapsari, Syarif Hidayatullah, Putu Hadi Setyarini, Kartika A. Madurani, Hendra Hermawan	Effectiveness of a fish scales-derived chitosan coating for corrosion protection of carbon steel	2022	Egyptian Journal of Petroleum
008	Aboudamia *et al.*	Potential of discarded sardine scales (*Sardina pilchardus*) as chitosan sources	2020	Journal of the Air & Waste Management Association
009	Suneeta Kumari, P. Ratha, A. Sri Hari Kumar, T. N. Tiwari	Extraction and characterization of chitin and chitosan from fishery waste by chemical method	2015	Environmental Technology & Innovation
010	Deby Kania Tri Putri, Wijayanti Diah W. H, Beta Widya Oktiani, Candra K, Bayu Indra Sukmana, Priyawan Rachmadi, Harun Achmad	Synthesis and Characteristics of Chitosan from Haruan (*Channa striata*) Fish Scales	2020	Dentistry, Vol.11, Iss.1
011	Suneeta Kumari, Rath P., Sri Hari Kumar A.	Chitosan from shrimp shell (*Crangon crangon*) and fish scales (*Labeo rohita*): Extraction and characterization	2016	African Journal of Biotechnology
012	Suneeta Kumari & Pradip Kumar Rath	Extraction and Characterization of Chitin and Chitosan from (*Labeo rohit*) Fish Scales	2014	Procedia Materials Science
013	Carlos Molina-Ramírez *et al.*	Characterization of Chitosan Extracted from Fish Scales of the Colombian Endemic Species	2021	Polymers

		Prochilodus magdalenae as a Novel Source for Antibacterial Starch-Based Films		
014	Anupama Baral *et al.*	Physicochemical and Biological Properties of Chitosan Extracted from Fish Scales of *Labeo rohita*	2023	Journal of Survey in Fisheries Sciences
015	Intan Harlia, Fatmah Dhafir, Astija	Effect of chitosan of milkfish scale waste (*Chanos chanos*) on the preservation of tomato fruit (*Lycopersium esculentum*)	2024	GSC Advanced Research and Reviews
016	Arni Irawaty Djais *et al.*	The Use of Chitosan Milkfish (*Chanos chanos*) Scales Waste As An Alternative Bone Regeneration Material In Socket Preservation	2023	Journal of Pharmaceutical Negative Results
017	Tadewos Lare, Belete Yilma Hirpaye, Ballekallu Chinna Eeranna, Babuskin Srinivasan	Extraction and characterization of chitin and chitosan from Nile tilapia scale in Lake Chamo, Arba Minch and their application in clarification of apple juice	2022	Journal of Food Processing and Preservation

Extraction of chitosan from fish scales and its extraction methods

The extraction of chitosan from fish scales has been studied across various species, each offering unique benefits for diverse applications (See Table 2). Species such as *Lutjanus* spp. (Snapper) and *Lates calcarifer* (Barramundi) have been explored for their potential in producing chitosan, which has shown promise in antibacterial coatings and guided tissue regeneration membranes (Florencia *et al.*, 2022; Susanto *et al.*, 2019). *Channa striata* (Haruan) demonstrated significant antibacterial activity, making it a promising source for medical applications (Widyaningrum). Similarly, *Tilapia mossambica* and *Oreochromis* sp. (Tilapia) have been widely studied, with findings highlighting their effectiveness in producing high-yield, bioactive chitosan suitable for food preservation and wastewater treatment (Takarina *et al.*, 2017; Praghati *et al.*, 2023; Chandra *et al.*, 2022; Gapsari *et al.*, 2022). Moreover, *Sardina pilchardus* (Sardine) scales have emerged as a valuable marine source for high-purity chitosan, offering significant environmental benefits (Aboudami *et al.*, 2020). Other species like *Labeo rohita* (Rohu) and *Prochilodus magdalenae* have been utilized for their ability to yield chitosan with strong antioxidant and antibacterial properties, while *Chanos chanos* (Milkfish) scales have proven effective in food preservation and bone regeneration (Kumari *et al.*, 2015; Molian-Ramirez *et al.*, 2021; Harlia *et al.*, 2024; Djais *et al.*, 2023). Each species contributes distinct characteristics to the chitosan produced, influencing its potential applications across various fields.

The extraction process for chitosan from fish scales predominantly involves chemical methods, characterized by three main stages: deproteination, demineralization, and deacetylation. Studies on species like *Lutjanus* spp., *Channa striata*, and *Lates sp.* utilized a sequence of chemical treatments, including sodium hydroxide for deproteination, hydrochloric acid for demineralization, and high-concentration sodium hydroxide for deacetylation. These methods effectively isolated chitosan with varying degrees of deacetylation, ranging from 61% to 84.05%, directly influencing the functional properties of the chitosan, such as crystallinity, solubility, and antibacterial activity. Variations in extraction parameters, such as temperature, concentration, and duration of chemical exposure, significantly impact the yield and quality of chitosan. For instance, *Labeo rohita* and *Sardina pilchardus* scales subjected to optimized deacetylation processes produced chitosan with high degrees of purity, crystallinity, and functional properties suitable for applications in food, pharmaceuticals, and environmental management. These findings underscore the adaptability of chemical extraction methods to different fish species and emphasize the critical role of process optimization in maximizing chitosan's beneficial properties. They suggest the need for an optimized and standardized extraction method to ensure consistent quality and maximize the potential of chitosan derived from fish scales.

Table 2. Summary of chitosan extractions from fish scales and the methods utilized

Study	Fish species	Extraction method (s)	Key findings
A. Florencia, S. E. D. Putra, Y. P. Mukti (2022)	*Lutjanus* spp. (Snapper)	Chemical Method Deproteination (4.2% NaOH with a ratio of 1:6 (W/v) at 60 degrees Celsius for 5 hours), Demineralization (1.04 N HCl Solution with a ratio of 1:6 (w/v) at room temperature for 6 hours), Deacetylation (80% NaOH solution in a ration of 1:3 (w/v) at 110 degrees Celsius for 4 hours)	The study found that the chitosan extracted had a yield of 36.32%, with water content at 3.86% and ash content at 89.54%. In antibacterial testing, chitosan showed a smaller inhibition zone compared to the 0% chitosan control. The De Garmo test identified 0.5% as the optimal concentration of chitosan. However, chitosan coating was not as effective as paraffin wax in extending the shelf life of grapes. Despite this, the organoleptic test indicated that the chitosan coating did not alter the sensory qualities of the grapes, making it acceptable to consumers.
Susanto *et al.*, 2019	*Lates calcarifer* (Barramundi)	Acetic Acid Immersion and Centrifugation (Collagen was extracted from barramundi scales by immersing them in a 0.5 M acetic acid solution at 4°C for 3 days, followed by centrifugation and precipitation processes, resulting in dry collagen after freeze-drying. Chitosan membranes were then fabricated by mixing the extracted collagen with chitosan dissolved in acetic acid, freezing the mixture to form porous membranes, and sterilizing the final product using gamma-ray irradiation)	The chitosan-collagen membranes exhibited a fibrous surface and an ideal porous size for guided tissue regeneration (GTR) membranes. Although the mechanical strength was lower, the membranes show potential as alternative barrier materials for GTR, warranting further research to enhance their mechanical properties
Widyaningrum *et al.*, 2019	*Channa striata* (Haruan)	Chemical Method: Deproteination, Demineralization, Deacetylation Chitin isolation and chitosan production involved several steps. First, pulverized fish scales were boiled in a 4% NaOH solution at 80°C for 1 hour to remove protein content, followed by neutralization and drying in an oven. Next, the dried scale powder was demineralized by soaking in a 1% HCl solution at a 1:4 ratio for 24 hours, then neutralized with distilled water and dried. The presence of chitin was confirmed using Van Wesselink's color reaction, where the fish scale powder turned brown with 1% I2-KI solution and violet with pure H2SO4, indicating a positive chitin result. Finally, chitin was deacetylated by mixing with 50% NaOH and boiling at 80°C for 2 hours to produce chitosan, which was then neutralized and dried in an oven.	The study found that chitosan exhibited a Minimum Inhibitory Concentration (MIC) of 1.5% and a Minimum Bactericidal Concentration (MBC) of 3.5% against *Staphylococcus aureus*. Additionally, a 2.5% chitosan concentration demonstrated superior inhibitory activity against *Staphylococcus aureus* compared to 0.2% chlorhexidine gluconate, as evidenced by the difference in absorbance
Takarina *et al.*, 2017	White Snapper (*Lates* sp.)	Chemical Method (a) Deproteination (4.2% NaOH with a ratio of 1:6 (W/v) at 60 degrees Celsius for 5 hours), (b) Demineralization (2 N 4.2% HCl Solution with a ratio of	The results indicated that the chitosan produced met the minimum quality requirements, with the highest degree of deacetylation reaching 84.05%. This was achieved under treatment conditions involving 80% NaOH concentration,

		1:6 (w/v) at room temperature for 6 hours), (c) Deacetylation (varying parameters, since the study aimed to explore the degree of acetylation)	heating at 120°C, and a duration of 4 hours.
Pragathi *et al.*, 2023	*Tilapia mossambica*	Chemical method The process of chitosan production involved several steps: demineralization using 0.5 M hydrochloric acid, followed by deproteinization with 1% sodium hydroxide, and depigmentation/deodorization with 1% sodium hypochlorite. Finally, deacetylation was carried out with 40% sodium hydroxide to yield white powdered chitosan after treatment with acetic acid.	The results of the estimation of biochemical components of the fish scales revealed that the amount of protein was higher than the carbohydrates, lipid and ash. The antimicrobial assay showed the potential of chitosan as an antibacterial agent
Chandra *et al.*, 2022	Tilapia (*Oreochromis* sp.)	Chemical method solation of Chitosan and Chito-oligosaccharides from fish scales involved several steps: fish scales were first demineralized by soaking in 2% HCl at room temperature (1:5 w/v) for 24 hours, followed by washing, drying at 37°C, and then deproteinized with 4% NaOH under similar conditions. The resulting chitin was then ground, stored, and deacetylated by treating with 80% NaOH (1:15 g/ml) at 120°C for 6 hours to obtain chitosan.	Chitin yield was 32.74% and chitosan yield was 62.13% from 100 grams of dried fish scales. Additionally, the MBC/MIC ratio indicates the bacteriostatic effect of chitosan at the specified concentrations.
Gapsari *et al.*, 2022	Nile Tilapia (*Oreochromis niloticus*)	Chemical Method Chitosan synthesis involved three steps: washing and grinding Nila tilapia fish scales, then deproteinating them in 7% NaOH; demineralizing the resulting chitin in 1 M HCl; and finally, deacetylating the chitin in 70% NaOH. Each step included rinsing with deionized water and curing or drying at 100°C for 30 minutes.	This research evaluated the effectiveness of fish scale-derived organic coatings on ASTM A36 carbon steel using electrophoretic deposition (EPD) and dip coating (DC) techniques. Both methods were effective, with DC providing higher efficiency but less temperature resistance through a physical mechanism, while EPD offered lower efficiency but greater temperature stability through a chemisorption mechanism.
Aboudamia *et al.*, 2020	*Sardina pilchardus*	Chemical Method In this study, 400 g of S. pilchardus scales were used to extract chitin, which was then deacetylated and characterized. The chitin was extracted following the method of Aboudamia *et al.* (2020), and deacetylation was performed by treating the chitin with 40% sodium hydroxide and autoclaving at 15 psi/121°C for 20 minutes at a 1:10 solid/solvent ratio. The resulting chitosan was filtered, washed with distilled water until neutral, then further cleaned with methanol and acetone (Kurita *et al.*, 1993), and dried at 30°C for 12 hours.	The β-chitin extracted was successfully converted into chitosan through a simple, cost-effective chemical method. Analysis using FTIR, SEM, EDS, and XRD revealed a high degree of deacetylation (87%), crystallinity index (95%), and high purity, along with fibrillar and porous morphology. The chitosan demonstrated excellent fat and water binding capacities (310% and 510%, respectively), 93% solubility in acetic acid, and a molecular weight of 5.86 kDa. Converting *S. pilchardus* scales, a fish by-product, into valuable biopolymers like chitin and chitosan presents significant benefits for solid waste management, enhances the profitability of fish processing industries, and offers advantages across various fields.
Kumari *et al.*, 2015	*Labeo rohita*	Chemical Method Chitin was isolated from *Labeo*	The prepared chitosan was found to be soluble in 1% acetic acid, with a low

		rohita fish scales through a process of demineralization using 1.0 M HCl and deproteinization with 0.5 N NaOH, followed by drying and neutralization. The subsequent deacetylation to convert chitin into chitosan was optimized by steeping the chitin in strong sodium hydroxide before heating, improving the extent of deacetylation.	moisture content of 3% and ash content of 1%, indicating effective extraction methods. FTIR, XRD, and other analyses confirmed that the chitosan has a carbon/nitrogen ratio of 1.88, a degree of deacetylation of 61%, and high crystallinity, making it suitable for commercial applications in food, pharmaceuticals, and water treatment, while also reducing environmental pollutants from fishery waste.
Putri DKT *et al.*, 2020	Haruan (*Channa striata*)	Chemical Method a. Deproteination b. Demineralization c. Deacetylation (varying parameters, since the study aimed to explore the degree of acetylation)	This study demonstrated that chitosan derived from *Channa striata* scales possesses unique physical, chemical, and morphological characteristics that meet the standards set by the National Standard Agency of Indonesia (BSN)
Kumari *et al.*, 2016	*Labeo rohita*	Chemical Method a. Deproteination b. Demineralization c. Deacetylation	The study found that chitosan from fish scales had higher water-binding (492%) and fat-binding (226%) capacities than shrimp chitosan, both comparable to commercial chitosan. Additionally, fish chitosan exhibited greater crystallinity and a higher deacetylation degree (80%) than shrimp chitosan, indicating that fishery and shrimp waste are excellent sources of high-quality chitosan.
Kumari *et al.*, 2014	*Labeo rohita*	Chemical Method Preparation of chitosan from fish scales was performed using the general method comprising of demineralisation, decolourisation and deacetylation.	FTIR spectroscopy confirms fish scales as an ideal raw material for chitosan, revealing well-defined functional groups in the chitosan macromolecules. The pronounced bands in the experimentally prepared chitosan indicate a higher degree of crystalline order compared to other sources.
Molina-Ramirez *et al.*, 2021	*Prochilodus magdalenae*	Chemical Method Chitosan extraction involved three main stages: demineralization, deproteinization, and deacetylation	*Prochilodus magdalenae* scales, a common contaminant in Colombian river zones, were successfully used as a novel feedstock for chitosan production, yielding a chitosan with a lower molecular weight (107.18 kDa) compared to commercial chitosan (151.11 kDa). Mild extraction conditions (room temperature and 2 wt.% NaOH) were necessary due to the scales' β-type chitin, resulting in a chitosan with effective antibacterial properties despite its lower thermal stability (148.82 °C).
Baral *et al.* (2023)	*Labeo rohita*	Chemical Method Fish scales were demineralized by soaking in 0.1 M HCl at 90°C for 15 minutes and then washed with deionized water. Following this, proteins were removed using 1% NaOH at 100°C for 15 minutes, and chitin was deacetylated to chitosan with 1% NaOH at 100°C for 15 minutes, before washing to neutral pH and drying.	Chitin derived from fish scales (*L. rohita*) can be converted into the more beneficial compound chitosan, which can be employed in a range of applications. According to this study, *L. rohita* chitosan exhibits good antibacterial action. Moreover, it has a wide range of antioxidant properties, including scavenging activities for superoxide anion radicals, hydroxylradicals, reducing power and ferrous-ion chelating activity. Fish (*L. rohita*) chitosan may be employed as a source of natural antioxidants, as a potential food supplement, or as an ingredient in the pharmaceutical sectors based on the findings.
Harlia *et al.*, 2024	Milkfish (*Chanos chanos*)	Chemical Method Chitosan extraction involved three	Using milkfish scale chitosan as an edible coating effectively preserves tomatoes by

		main stages: demineralization, deproteinization, and deacetylation	minimizing color changes, weight loss, pH increase, and vitamin C reduction during storage. For optimal preservation over nine days, a 2.5% concentration was best for preventing color change and vitamin C loss, while a 1.5% concentration was most effective for reducing weight loss and controlling pH increase.
Djais *et al.* (2023)	Milkfish (*Chanos chanos*)	Chemical Method Chitosan extraction involved three main stages: demineralization, deproteinization, and deacetylation	Milkfish scales can increase the production of OPG and decrease RANKL expression so that it can help the process of bone regeneration after tooth extraction.
Lare *et al.*, 2022	Nile Tilapia	Chemical Method Demineralization Deproteinization Depigmentation Deacetylation	The chitosan, confirmed to be α-chitin and successfully converted to chitosan by FTIR, was used at various concentrations (0 to 0.25 g/L), with positive sensory evaluation results, demonstrating its potential for applications in the food and beverage industry, as well as other sectors.

Characterization of extracted chitosan from fish scales

The characterization of chitosan extracted from fish scales involved various techniques to evaluate its yield, composition, and structural properties as shown in Table 3. The findings from different studies provide a comprehensive view of the chitosan's characteristics and potential.

Chemical structure

The chemical structure of chitosan extracted from fish scales has been extensively studied using various spectroscopic and analytical techniques. Florencia *et al.* (2022) characterized chitosan by measuring its yield, water content, and ash content. Their findings suggest that while the extraction process was efficient, indicated by a chitosan yield of 36.318%, the high ash content of 89.54% points to incomplete demineralization, necessitating further optimization. Gapsari *et al.* (2022) utilized FTIR spectroscopy to trace the transition from raw fish scales to chitin and finally to chitosan, identifying the presence of key functional groups such as aromatic compounds, hydroxyl groups (O–H), carbonyls (C=O), and alkanes (C-H). Aboudamia *et al.* (2020) further explored the chemical structure through FTIR and XRD analysis, revealing a degree of deacetylation (DDA) of 87% and a crystalline index (CrI) value of 95%, confirming the presence of essential elements like carbon (C), oxygen (O), and nitrogen (N). Kumari *et al.* (2015, 2016, 2014) conducted multiple studies using FTIR, XRD, and other techniques to characterize chitosan and its precursor, chitin, identifying O-H, N-H, and C=O bonds, and confirming the formation of α-chitin with a high degree of crystalline order. Similarly, Djais *et al.* (2023) and Lare *et al.* (2022) employed FTIR spectrophotometry to confirm the successful deacetylation of chitin to chitosan, identifying functional groups indicative of chitosan formation.

Morphology

The morphological characteristics of chitosan derived from fish scales have been explored using scanning electron microscopy (SEM). Susanto *et al.* (2019) used SEM to provide detailed images of chitosan-collagen membranes, revealing a porous surface with pore sizes ranging from 16 to 100 µm. Putri *et al.* (2020) conducted SEM characterization of chitosan from Haruan fish scales, showing that the deproteinization, demineralization, and deacetylation steps influenced the production of chitosan, resulting in a structure that was almost round, porous, fibrous, broken, and irregular. Aboudamia *et al.* (2020) also utilized SEM analysis, revealing a fibrillar and pleated morphology consistent with the expected structure of chitosan.

Table 3. Summary of characterization techniques/parameters for fish scales-derived chitosan

Study	Characterization techniques/Parameters	Key findings
A. Florencia, S. E. D. Putra, Y. P. Mukti (2022)	The extracted chitosan from fish scales was characterized by measuring the yield, water content and ash content.	The chitosan yield in this study was 36.318%, indicating an efficient extraction process, while the water content of 3.86% meets quality standards for chitosan used as a bio preservative. However, the high ash content of 89.54% suggests incomplete demineralization, highlighting the need for optimization in the demineralization process
Susanto *et al.*, 2019	Chitosan-Collagen Membrane Characterization involved three key tests: FTIR spectroscopy identified functional groups in the membrane by measuring infrared absorption between 400 and 4,000 cm^{-1}; scanning electron microscopy (SEM) provided detailed images of the membrane's surface and cross-sections at 100× and 500× magnifications; and tensile strength testing assessed the membrane's mechanical properties by measuring the load required to break samples sized 40 × 10 mm.	The FTIR spectrum revealed characteristic peaks indicating the presence of both chitosan and collagen in the membrane. The tensile strength and elongation at break were 0.28 MPa and 8.53% in dry conditions, and 0.12 MPa and 25.6% in wet conditions. The membrane porosity was 38.85%, and SEM imaging showed a porous surface with pore sizes ranging from 16 to 100 µm.
Gapsari *et al.*, 2022	The powder, chitin, and chitosan were characterized using Fourier-transform infrared spectroscopy (FTIR, Spectrum Two, Perkin Elmer) and compared with standard chitosan functional groups.	Figures 1a-c illustrate the FTIR spectra showing the transition of functional groups from raw fish scales to chitin and finally to chitosan. Figure 1d compares the FTIR spectra of chitosan coatings applied on steel plates using EPD and DC techniques, revealing similar functional groups on both surfaces. Specifically, Figure 1a demonstrates that fish scale-derived chitosan primarily consists of organic functional groups, including aromatic compounds with carbon rings, hydroxyl groups (O–H) at 3455.03 cm^{-1}, carbonyls (C) at 1638.21 cm^{-1}, and alkanes (CH) at 963.18 cm^{-1}.
Aboudamia *et al.*, 2020	The chemical structure of obtained chitosan was characterized based on Fourier transforms infrared spectroscopy (FTIR), X-ray powder diffraction (XRD), Scanning electron microscope (SEM), and Energy-dispersive X-ray spectroscopy (EDS)	According to the results of FTIR and XRD analysis, the degree of deacetylation (DDA), and the crystalline index (CrI) value of obtained chitosan is respectively about 87% and 95%. The SEM and EDS analysis revealed respectively fibrillar and pleated morphology with the presence of three major elements characterizing the chitosan, which are C, O, and N. T
Kumari *et al.*, 2015	The obtained chitin and chitosan have been characterized by using different techniques like spectral analysis, X-ray diffraction, Elemental analysis, Fourier transforms infrared spectroscopy (FTIR), Scanning electron microscopy (SEM) and Differential scanning calorimetry (DSC)	XRD analysis indicated the crystalline nature of the chitin and chitosan. The FTIR patterns displayed the bands corresponding to stretching and vibration of O-H, N-H and CO bonds and conformed the formation of α-chitin. D
Putri *et al.*, 2020	Morphological Characteristics of Chitosan from Haruan Fish Scales with SEM Characterization of chitosan using electron microscopy, scanning was carried out at 1500x magnification (Figure 1) and 5000x (Figure 2).	Based on SEM characterization, it is known that the steps of deproteinization, demineralization, and deacetylation show an influence on chitosan production. The figure below shows that chitosan from haruan fish scales has many layers with a structure that is almost round, porous, fibrous, broken, and irregular
Kumari *et al.*, 2016	Structural differences between shrimp chitosan and fish chitosan were studied by using FTIR, thermo-gravimetric analysis (TGA), Xray powder diffraction (XRD) and scanning electron microscopy (SEM).	Fourier transforms infrared spectroscopy (FTIR) spectra presented a detailed structure of α-chitin with O-H, N-H and CO stretching movements. Characteristic properties of extracted chitosan were found to depend upon the source of origin and degree of deacetylation
Kumari *et al.*,	Fourier-Transform Infrared Spectroscopy	FTIR spectroscopy confirms fish scales as an

2014	(FTIR) was used to analyze chitin and chitosan mixtures over a frequency range of 400 to 4,000 cm−1, while X-ray Diffraction (XRD) assessed the crystallinity of the samples using a Rigaku III diffractometer. Scanning Electron Microscopy (SEM) provided micrographs of the chitosan's surface and cross-section after fracturing the samples in liquid nitrogen.	ideal raw material for chitosan, revealing well-defined functional groups in the chitosan macromolecules. The pronounced bands in the experimentally prepared chitosan indicate a higher degree of crystalline order compared to other sources.
Djais *et al.*, 2023	FTIR spectrophotometry	2 Functional group testing is carried out using FTIR spectrophotometry to demonstrate that the chitin deacetylation process has produced chitosan
Lare *et al.*, 2022	FTIR Analysis	The FTIR patterns showed bands corresponding to the vibration and stretching of O–H, N–H, and C=O bonds, confirming the formation of α-chitin and the successful conversion of chitin to chitosan.

Mechanical properties

The mechanical properties of chitosan and its derivatives have been evaluated to assess their suitability for various applications. Susanto *et al.* (2019) conducted tensile strength testing on chitosan-collagen membranes, finding that the membranes exhibited a tensile strength of 0.28 MPa and an elongation at break of 8.53% in dry conditions, with corresponding values of 0.12 MPa and 25.6% in wet conditions. The membrane porosity was measured at 38.85%. Kumari *et al.* (2016) used thermogravimetric analysis (TGA) and other methods to study the structural differences between shrimp chitosan and fish chitosan, highlighting how the source of origin and degree of deacetylation affect the mechanical properties of the resulting chitosan.

Applications of chitosan extracted from fish scales

Chitosan extracted from fish scales has demonstrated a wide range of applications, highlighting its versatility and potential across various fields. These applications include:

1. Edible Coatings: Chitosan from fish scales, such as snapper and milkfish, is used as an edible coating to extend the shelf life of fruits and vegetables. Research shows that these coatings effectively preserve produce by reducing color changes, weight loss, pH increase, and vitamin C degradation during storage (Florencia *et al.*, 2022; Harlia *et al.*, 2024).
2. Biomedical Materials: Fish scale-derived chitosan serves as a biomaterial for guided tissue regeneration (GTR) and wound dressings. Chitosan-collagen membranes from barramundi scales provide an ideal fibrous surface and porosity, though their mechanical strength is relatively low. These membranes are promising for applications in tissue engineering and wound care (Susanto *et al.*, 2019).
3. Antibacterial Agents: Chitosan derived from fish scales, including Haruan and *Prochilodus magdalenae*, has demonstrated significant antibacterial properties. Studies by Widyaningrum *et al.* (2019) and Pragathi *et al.* (2023) found that this chitosan effectively inhibits pathogens like *Staphylococcus aureus* and *Pseudomonas* spp., with Minimum Inhibitory Concentrations (MIC) as low as 1.5% and Minimum Bactericidal Concentrations (MBC) of 3.5%. Its cationic nature and biodegradability make it suitable for a range of applications including pharmaceuticals, cosmetics, food preservation, wound dressings, and antimicrobial textiles. Additionally, chitosan from *Prochilodus magdalenae* also showed antibacterial activity against *Escherichia coli*, highlighting its broad-spectrum antimicrobial potential.
4. Organic Coatings: Fish scale-derived chitosan is employed as an organic coating for materials like carbon steel. Techniques such as electrophoretic deposition (EPD) and dip coating (DC) have been used, with each method offering unique benefits in

terms of efficiency and temperature stability (Gapsari *et al.*, 2022).

Commercial and Environmental Applications: The commercial potential of fish scale-derived chitosan extends to food, pharmaceuticals, and water treatment. It provides a sustainable approach to reducing environmental pollutants from fishery waste while adding value to fish processing industries (Kumari *et al.*, 2015, 2016).

Conclusion

The review of seventeen peer-reviewed articles on chitosan extraction from fish scales, published between 2014 and 2024, reveals a burgeoning interest in this field. The studies highlight the extraction methods, applications, and characteristics of chitosan derived from various fish species. Notably, recent research has surged since 2020, reflecting a growing focus on this relatively new area.

The extraction of chitosan from fish scales predominantly involves chemical methods, including deproteination, demineralization, and deacetylation. Different fish species, such as *Lutjanus* spp., *Channa striata*, *Lates calcarifer*, Tilapia and Milkfish, have shown potential in producing chitosan with diverse applications, including antibacterial coatings, biomedical materials, and food preservation. Variations in extraction parameters significantly impact chitosan quality, suggesting the need for optimized and standardized methods to ensure consistency and maximize benefits.

Characterization of chitosan reveals key insights into its chemical structure, morphology, and mechanical properties. Techniques like FTIR and SEM have confirmed successful deacetylation and detailed the structural attributes of chitosan, which influence its functionality. Mechanical property assessments further demonstrate how the source and processing conditions affect the material's performance. The applications of chitosan extracted from fish scales are broad and promising. It serves as an effective edible coating for extending the shelf life of produce, a biomaterial for wound care and tissue regeneration, and an antibacterial agent with potential uses in pharmaceuticals and environmental management. Additionally, fish scale-derived chitosan proves valuable as an organic coating for metals and offers commercial and environmental benefits by reducing waste and enhancing the sustainability of fish processing industries.

Recommendation(s)

Based on the review of recent studies on chitosan extraction from fish scales, several recommendations can be made to advance the field. Standardizing extraction methods is crucial for enhancing consistency and maximizing the benefits of chitosan, which involves optimizing parameters like temperature, concentration, and chemical treatment duration. Further research is needed to explore the specific attributes and benefits of chitosan from different fish species, enabling more effective comparative studies and identification of optimal sources for various applications. Additionally, the continued use and advancement of characterization techniques such as FTIR and SEM are essential for a comprehensive understanding of chitosan's chemical structure, morphology, and mechanical properties, which will facilitate its optimization and practical application.

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