



# Sustainable Extraction and Biomedical Applications of Chitosan from Fish Scales, Mushroom Stalks, and Banana Peels

Hasina Jamadar<sup>1</sup>, Pooja Malave<sup>1\*</sup>

<sup>1</sup>Department of Biotechnology, Dr D. Y. Patil Arts, Commerce and Science College, Pimpri, Pune  
Maharashtra, India.

## Abstract :

Chitosan, a biopolymer derived from chitin, has gained prominence due to its biodegradability, biocompatibility, and antimicrobial properties. Traditionally extracted from crustacean shells, chitosan production faces limitations such as allergenicity, seasonal availability, and environmental concerns. This study investigates alternative sources—Labeo rohita fish scales, white button mushroom stalks, and banana peels—for chitosan extraction using acid-alkali chemical treatment. The extracted biopolymers were characterized using Fourier Transform Infrared Spectroscopy (FTIR), solubility testing, and pH analysis. Among the sources, fish scales yielded the highest purity and quantity of chitosan, while mushrooms and banana peels produced chitosan-like compounds with moderate functionality. These materials were further utilized to develop herbal biodegradable bandages and hydrogels, demonstrating promising antimicrobial and wound-healing properties. The findings support the valorization of agro-waste and seafood by-products for sustainable biomedical innovations.

## 1.0 Introduction

Chitosan is a linear polysaccharide composed of  $\beta$ -(1 $\rightarrow$ 4)-linked D-glucosamine and N-acetyl-D-glucosamine units, obtained through the partial deacetylation of chitin. It is the second most abundant natural polymer after cellulose and has been widely recognized for its biodegradability, biocompatibility, antimicrobial activity, and wound-healing potential, making it valuable in biomedical, pharmaceutical, and agricultural fields (Timilsina, 2025).

Traditionally, chitosan is extracted from crustacean shells; however, this approach is limited by seasonal availability, high processing costs, allergenic potential, and challenges in marine waste disposal (Baral et al., 2024). These drawbacks have prompted the search for alternative, sustainable, and non-crustacean sources such as fungi, agricultural residues, and fishery by-products.

Among alternative resources, fish scales are particularly promising. *Labeo rohita* fish scales contain collagen and mineralized chitin, which can be efficiently deacetylated to yield high-quality chitosan (Timilsina, 2025; Baral et al., 2024). Similarly, fungal sources such as white button mushrooms (*Agaricus bisporus*) offer a renewable and non-allergenic supply of chitin, which can be converted into chitosan for biomedical applications (Almeida et al., 2025; Akila & Priya, 2012). Recent studies have also demonstrated the valorization of mushroom waste into chitin oligosaccharides with antioxidant and immunomodulatory potential (Windels et al., 2025).

Banana peels, a major agricultural by-product, do not yield pure chitosan but contain polysaccharides and polyphenolic compounds that can be chemically modified to form chitosan-like molecules. These derivatives possess antimicrobial, antioxidant, and biodegradable film-forming properties, making them suitable for green biomaterial applications (Musa et al., 2019). The use of banana peels not only reduces agricultural waste but also aligns with circular economy principles of waste valorization and sustainable resource management.

In recent years, chitosan-based composites integrated with herbal extracts such as neem and turmeric have been developed into bandages and hydrogels with enhanced antimicrobial and wound-healing effects. These combinations demonstrate the synergy of traditional medicinal plants with modern biopolymer technology, thereby supporting low-cost biomedical product development (Sathiyavimal et al., 2019).

The present study investigates the extraction of chitosan from *Labeo rohita* fish scales and mushroom stalks, and evaluates banana peels for the production of chitosan-like compounds using acid–alkali chemical treatment. The extracted products were characterized by Fourier Transform Infrared Spectroscopy (FTIR), solubility testing, and pH analysis. A comparative evaluation of yield, purity, and functional properties across the three sources was performed, followed by their application in the preparation of herbal biodegradable bandages and hydrogels. This study highlights the potential of waste-derived chitosan for sustainable biomedical innovations, particularly in wound care.

## 2.0 Materials and Methodology

### 2.1 Sample Collection and Preparation

Fresh *Labeo rohita* fish scales were collected from a local fish market in Pune, Maharashtra, while white button mushroom (*Agaricus bisporus*) stalks and banana peels were obtained from vegetable vendors. All raw materials were thoroughly washed under running tap water to remove adhering impurities, followed by rinsing with distilled water. The cleaned samples were cut into small pieces, dried in a hot air oven at 60 °C until constant weight, and ground into a coarse powder for further processing.

## 2.2 Extraction of Chitosan

As referred from (Kumari and Rath 2014) and (Ooi, Munawar, and Kiew 2023)

### 2.2.1 Deproteinization

The dried powders were subjected to deproteinization using sodium hydroxide. Specifically, 10 g of each sample was treated with 100 mL of 1 M NaOH (1:10 ratio) in conical flasks. The mixtures were heated at 90 °C for 2 h with occasional stirring. After treatment, residues were washed with distilled water until neutral pH and dried at 60 °C.

### 2.2.2 Demineralization

The deproteinized residues were treated with 1 M hydrochloric acid (1:10 ratio, w/v) at room temperature for 2 h with continuous stirring to remove calcium carbonate and other minerals. Effervescence indicated the release of CO<sub>2</sub> during the reaction. The residues were filtered, washed with distilled water to neutral pH, and oven-dried.

### 2.2.3 Deacetylation

The dried chitin was converted into chitosan by treating with 50% NaOH (w/v) at 100 °C for 3 h under constant stirring. The resulting product was filtered, washed repeatedly with distilled water until pH was neutral, and dried at 60 °C. The dried material was weighed to calculate yield and stored in desiccators until further analysis.

## 2.3 Characterization of Extracted Chitosan

### 2.3.1 Fourier Transform Infrared Spectroscopy (FTIR)

The extracted materials were characterized by FTIR to confirm the presence of functional groups corresponding to chitin and chitosan. Dried powders were mixed with KBr, pelletized, and scanned over the range of 4000–400 cm<sup>-1</sup>.

### 2.3.2 Solubility Test

The solubility of extracted chitosan was evaluated by dissolving 1% (w/v) samples in 1% acetic acid solution. The degree of solubility was recorded as clear, turbid, or insoluble after 24 h.

### 2.3.3 pH Analysis

Aqueous suspensions (1% w/v) of the extracted materials were prepared in distilled water, and pH was measured using a digital pH meter to assess acidity/basicity relative to standard chitosan.

### 2.3.4 Formulation of Herbal Biodegradable Bandages and Hydrogels

The extracted chitosan and chitosan-like compounds were blended with aloe vera gel and turmeric extract for bioactivity enhancement. Bandages were fabricated by coating sterile cotton gauze with chitosan–herbal mixtures and drying at room temperature. Hydrogels were prepared by crosslinking chitosan solutions with 1% glutaraldehyde under mild stirring, followed by the incorporation of herbal extracts.

## 2.4 Antimicrobial Assay

The antimicrobial activity of bandages and hydrogels was tested against *Escherichia coli* and *Staphylococcus aureus* using the agar well diffusion method. Zones of inhibition were measured after 24 h incubation at 37 °C.

## Results

The extraction of chitosan was successfully carried out from three different raw materials: *Labeo rohita* fish scales, white button mushrooms, and banana peels. Sequential processing through deproteinization, demineralization, and deacetylation yielded chitosan in varying quantities depending on the source.

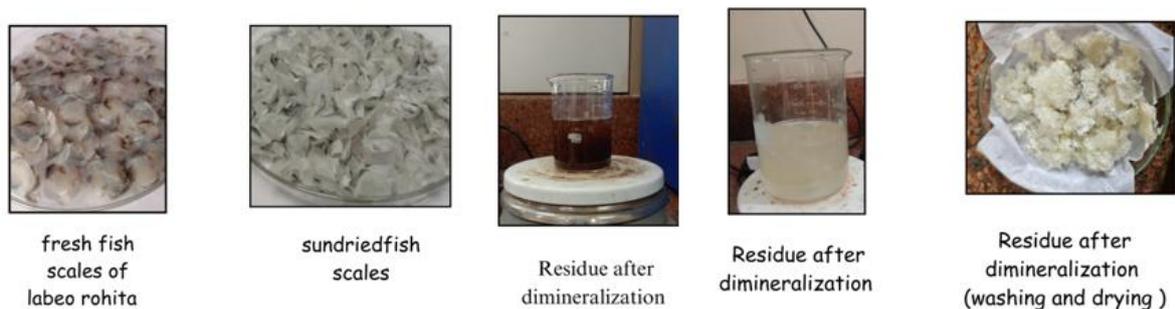


Figure 1: Demineralization and deproteinization of Fish scales



Figure 2: Demineralization and deproteinization of Banana peels

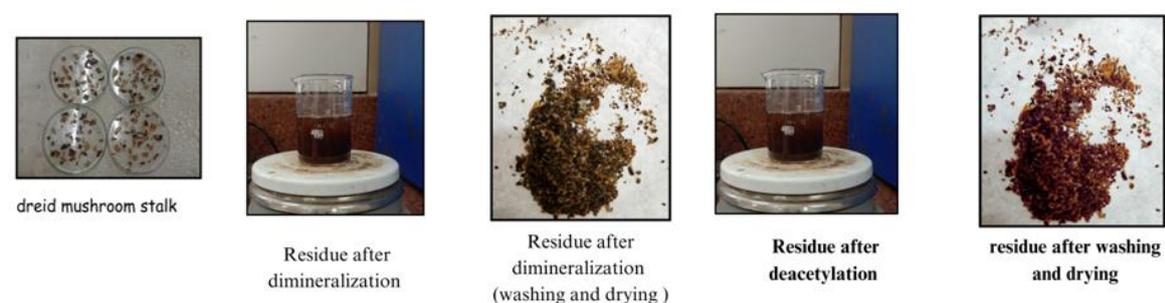
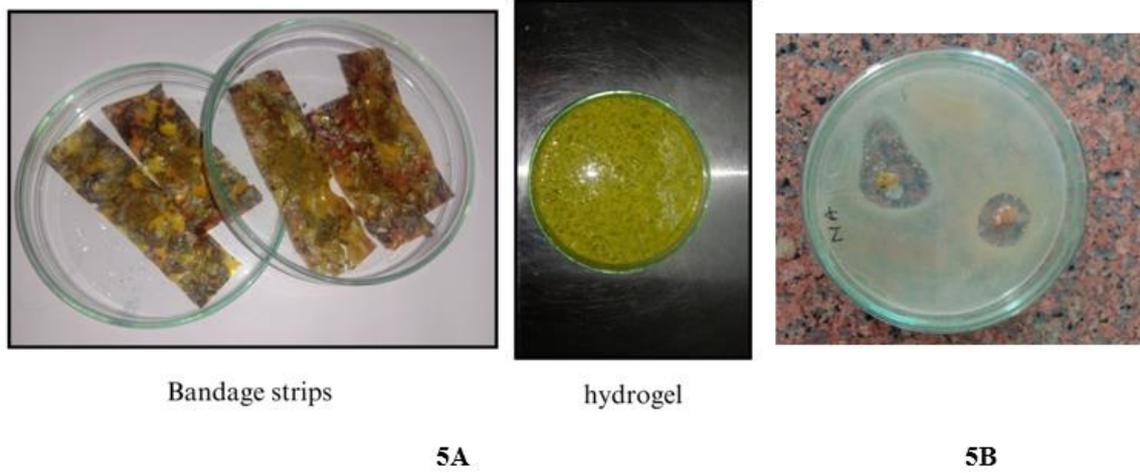


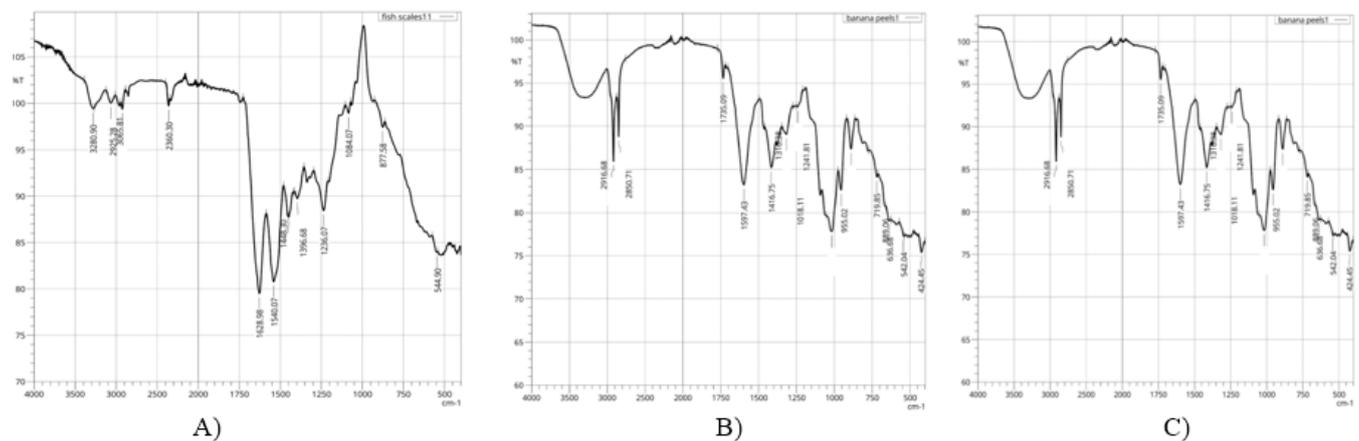
Figure 3: Demineralization and deproteinization of Mushroom Stalks

From fish scales, the highest yield of chitosan was obtained, with the deproteinized sample (12 g) reducing to approximately 5.9 g after the complete extraction process. White button mushrooms, though initially yielding 7.7 g of dried stalk waste, produced only 0.55 g of chitosan after processing, representing a lower overall conversion compared to fish scales. Banana peels provided an intermediate output, with 11.6 g of dried sample resulting in approximately 1.93 g of final product.



**Figure 5: 5a-Preparation of chitosan-based bandage strips and hydrogel from waste-derived biomaterials, 5b- Antibacterial activity of Bandage against *E.coli***

FTIR analysis of the extracted samples confirmed the presence of characteristic functional groups of chitosan in fish scales and mushrooms, particularly the peaks corresponding to  $-OH$ ,  $-NH_2$ , and  $C-O$  stretching vibrations, which are indicative of chitosan's structural identity. In the case of banana peels, FTIR spectra revealed the presence of chitosan-like compounds but lacked some of the distinct peaks associated with fully deacetylated chitosan, confirming that the product was not true chitosan.



**Figure 6: FTIR analysis of A) Fish scales B) Banana peels C) Mushroom stalks**

## Discussion

The comparative analysis clearly demonstrated that *Labeo rohita* fish scales represent a superior source of chitosan, as reported in many studies, when compared to mushrooms and banana peels. This can be attributed to their structural composition, which includes a significant proportion of chitin embedded in collagen and mineralized layers. Following effective deproteination and demineralization, fish scales consistently yielded a higher quantity of chitosan with clear FTIR confirmation.

Mushrooms, while producing a lower yield than fish scales, remain a viable and sustainable alternative source of chitosan. The fungal cell walls are known to contain chitin, but in lower concentrations relative to crustaceans and fish-derived sources. The relatively small yield in this study (0.55 g from 7.7 g of initial material) aligns with existing literature suggesting that fungal-derived chitosan often requires larger quantities

of raw material to achieve sufficient yield. Nonetheless, mushroom-derived chitosan holds advantages, particularly being non-allergenic, renewable, and free from seasonal restrictions, making it valuable for biomedical applications.

Banana peels did not yield true chitosan but instead produced chitosan-like compounds with partial similarity in functional groups. This finding is consistent with prior reports that banana peels contain polysaccharides and polyphenolic compounds that can mimic some of the biological properties of chitosan, such as antimicrobial and antioxidant activity, but lack the complete structural identity of chitosan. While banana peels may not serve as a direct source of chitosan, their potential in creating biodegradable films, coatings, or chitosan-substitute materials should not be overlooked. Moreover, their use highlights the broader goal of agricultural waste valorization, converting otherwise discarded materials into functional biomaterials.

The successful application of the extracted chitosan into the preparation of a wound healing bandage further validated its biomedical potential. Fish scale-derived chitosan, with its higher yield and purity, proved most suitable for this purpose due to its film-forming ability and antimicrobial activity. Mushroom-derived chitosan, although limited in yield, may also contribute effectively in similar applications where smaller-scale production is sufficient. Banana peel-derived chitosan-like compounds, while not identical, still provide opportunities for eco-friendly material development, particularly in combination with other polymers.

## Conclusions

Overall, this study underscores the feasibility of utilizing non-crustacean sources for chitosan extraction. Fish scales emerged as the most promising source in terms of yield and quality, mushrooms offered a renewable alternative with biomedical relevance, and banana peels, while not producing pure chitosan, demonstrated potential in sustainable biomaterial development. These findings support the dual objective of advancing biomedical innovation while simultaneously contributing to environmental sustainability by valorizing organic waste streams.

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