



# Physicochemical properties and characterization of chitosan synthesized from fish scales, crab and shrimp shells



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## ABSTRACT

Chitosan is derived from different starting materials such as fish scales, shrimp and crab shells by the process of deacetylation of chitin, which is carried out using 40% KOH at 90 °C for 6 h. Prepared chitosan was characterized by Fourier transforms infrared spectroscopy, X-ray powder diffraction, Scanning electron microscope and Thermogravimetric analysis. Further the physicochemical properties of chitosan like Fat binding capacity (FBC), water binding capacity (WBC), solubility, average molecular weight, ash content, moisture and degree of deacetylation of chitosan were also studied. Crystalline index (%) values of commercial, shrimp, crab and fish chitosan were found to be 96, 82, 88 and 84% respectively. The presence of amino group was confirmed from the FTIR spectra of chitosan synthesized. TGA results demonstrated the lower thermal stability of chitosan. Relatively smoother surface and nano-fiber structures were observed from SEM analysis. The degree of deacetylation of chitosan from different sources such as shells of fish, shrimp and crab were found to be 75%, 78%, and 70% respectively. In a similar way the WBC and FBC of fish, shrimp and crab shells were found to be 492, 358 and 138% and 226, 246 and 138% respectively.

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## 1. Introduction

Chitosan is deacetylated derivative of chitin, which is one of the most abundant polysaccharides occurring in nature next to cellulose [1]. Commercially, chitin and its derivatives are extracted from shrimp shell, crab shell, crayfish and krill shell. In recent years, some studies have emphasized that insects, mushrooms, coral and crustacean resting eggs can be alternative sources of chitin. Around 20,000 orthoptera species have been acknowledged in the recent past, but these organisms have been highly ignored with regards to chitin extraction [2]. Chitin is found on the outer surface of the arthropod body, in the cell wall of mushrooms and in cell structures of algae and yeast [3]. Despite of chitin's extensive application in areas such as biotechnology, agriculture, and medicine, very few sources (shrimp, crab and kill) have been cited as commercial chitin resources. Recent studies reveal that chitin and its derivatives have new applications in the areas such as stent coatings, sensors, wound dressings and cosmetics etc (Fig. 1). Considering the wide application of chitin and its derivatives, there is an increasing demand for unconventional chitin sources [4]. Crustacean waste from crab shells is composed of chitin which forms a chitin protein

complex with proteins. Fish scales, shrimp and crab shells are composed of different components such as proteins (15–50%), minerals (30–50%), and chitin (15–30%) [5,6]. Production of chitin uses the basic raw materials which are cuticles of various crustaceans, principally crabs, and shrimps. In fishery waste chitin existing is closely associated with proteins, minerals, lipids, and pigments. They all have to be quantitatively removed to achieve the highest purity of chitin necessary for biological applications. Chitin is an amino polysaccharide, built of a long polymer chain consisting of N-acetyl glucosamine units connected by  $\beta$ -1,4-glycoside bonds [7]. In order to separate chitin from crustacean shells, chemical processing steps such as demineralization and deproteinization are applied using strong acids and bases to remove calcium carbonate and proteins, respectively. However, the use of these chemicals may cause partial deacetylation of chitin and hydrolysis of the polymer resulting in final inconsistent physiological properties [6]. This research article covers a detailed study of chitin and chitosan extracted from three different sources such as shrimp (*Crangon crangon*), fish (*Labeo rohita*) and crab (*Scylla Serrata*) shells. Uniform conditions were used for the deproteinization and demineralization processes in the extraction of chitin and synthesis of chitosan from fish. Shrimp and crab waste. Physicochemical properties of chitin and chitosan (extracted from different sources) were determined and the materials were characterized using different techniques.

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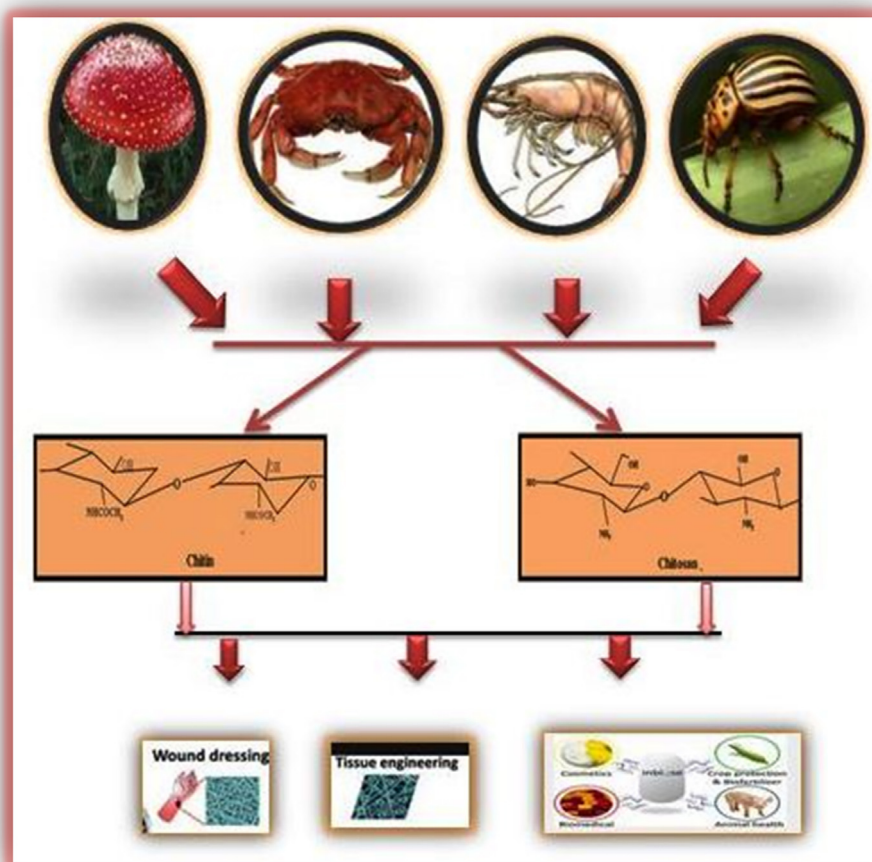


Fig. 1. Different sources of chitin in nature and applications.

## 2. Material and method

Chitosan synthesis involves three major steps such as deproteinization, demineralization and deacetylated. Fishery waste

(*Labeo rohita*), shrimp shells (*Crangon crangon*), and crab shells (*Crangon crangon*) were treated with 3% NaOH for 30 min at a temperature of 80 °C. The protein matter was depleted from the scales and the sample mass was washed with distilled water over and over again till the sample reaches neutrality. The deproteinized samples were dried at room temperature and then treated with 3% HCl (demineralization) for 30 min at room temperature to yield chitin. The excess HCl present in the chitin sample was removed by thorough washing, with distilled water and the sample was dried at room temperature. Chitin samples were deacetylated to form chitosan by treating with 40% KOH for 6 h at a temperature of 90 °C.

## 3. Physico-chemical properties of chitosan from different sources

### a Solubility

The solubility of chitosan depends on its biological origin, molecular weight and degree of deacetylation. For measuring solubility

of chitosan, chitosan was dissolved in 1% acidic acid and subjected to centrifugation. The undissolved solid mass was separated and dried in an oven and weighed. The solubility data of the samples was calculated using the following expression:

$$\% \text{ solubility} = \frac{(\text{Initial weight of tube + chitosan}) - (\text{Final weight of tube + chitosan})}{(\text{Initial weight of tube + chitosan}) - (\text{Initial weight of tube})} \times 100 \quad (1)$$

### b Fat binding capacity

Fat-binding capacity of prepared chitosan was calculated using the equation proposed by Wang and Kinsella [8]. For FBC, 0.5 g of chitosan sample was taken in a 50 ml centrifuge tube which was weighed initially and 10 ml of soya oil was taken followed by mixing on a vortex mixer for 1 min to disperse the sample. The contents were left at ambient temperature for 30 min with intermediate shaking (for a period of 5 s) after every 10 min and then centrifuged at 3200 rpm for 25 min. After the centrifugation, the supernatant was decanted, and the tube was weighed again and FBC was calculated using the following relationship:

$$\text{FBC}(\%) = \frac{\text{fat bound (g)}}{\text{initial sample weight (g)}} \times 100 \quad (2)$$

### c Water binding capacity

Water binding capacity of chitosan was determined by using the method reported by Wang and Kinsella [8]. For water binding capacity (WBC) 0.5 g of chitosan sample was taken in a centrifuge tube of 50 ml which was weighed initially and 10 ml of water was

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