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Valorization of Shrimp Processing Waste by Green Methodology and its Application in Chitosan-PEG Composite Film

A.K. Sanivarapu[1,*], B.K. Babu[2], N.V.V.D. Prasad[3], K. Satish Kumar[4]

Abstract

Shrimp processing waste is mostly unidentified marine by product, presents considerable environmental pollution which also offers a rich but not fully exploited sources of biopolymers. The current work presents a sustainable method to convert shrimp processing waste into chitosan a biopolymer by green methodology. Isolation of chitin to chitosan from shrimp processing waste was carried through four green methodology steps i. Demineralization using citric acid ii. Deproteinization using papain iii. Decolorization using hydrogen peroxide and iv. Deacetylation using Deep Eutectic Solvent (DES). The produced chitosan was analyzed by UV-Visible, IR, EDS and SEM techniques. The SEM and EDS images of commercial chitosan are compared with chitosan synthesized from shrimp processing waste. The results confirmed that chitosan from shrimp processing waste is of high quality with structural and morphological properties comparable to commercial chitosan. A composite of Chitosan –PEG (Poly Ethylene Glycol) film was prepared by the solvent casting method using chitosan extracted from shrimp shell waste and commercial chitosan. These films are analyzed by FT-IR analysis and thickness. FT-IR analysis of both commercial chitosan and chitosan from shrimp processing waste with PEG confirmed the presence of characteristic functional groups and revealed that intermolecular hydrogen bonding interaction between both Chitosan and PEG. These results suggest successful incorporation of PEG into the chitosan matrix. Chitosan from shrimp processing waste – PEG film exhibited a greater thickness in comparison to the commercial chitosan-PEG film. These findings demonstrate that chitosan obtained from shrimp processing waste is an alternative way to produce eco-friendly biodegradable films. In addition, the physicochemical properties suggest that the composite film can be suitable for applications in food packaging, wound healing, biomedical devices and water purification. Hence this study highlights the conversion of sea food processing waste into biomaterial.

Keywords: Shrimp processing waste, chitosan, biopolymer, green methodology, deep eutectic solvent (DES), Composite of Chitosan-PEG film.

***Author for Correspondence**
A.K. Sanivarapu

[1]Associate Professor, Department of Chemistry, Aditya University, Suramplaem, Andhra Pradesh, India
[2]Professor, Department of Engineering Chemistry, AU College of Engineering, Visakhapatnam, Andhra Pradesh, India
[3,4]Research Scholar, Department of Chemistry, Aditya University, Suramplaem, Andhra Pradesh, India

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INTRODUCTION

The Shrimp processing sector in India generated a large volume of Shrimp processing waste, when thrown into sea or land it causes air, water and soil pollution [1–3]. Out of several important byproducts chitin after cellulose is considered as the second rich source of biopolymer in nature. Chitin which is chemical known as N-acetylglucosamine polymer is having a linear structure having characteristics like biodegradability, biocompatibility, non-toxicity and thermal stability [4–7]. Another important byproduct of chitin is chitosan which is obtained on deacetylation. Chitosan is also naturally occurring cationic biopolymer and commercially important

due to its properties such as non-toxicity, adsorption, physiological compatibility and chelating capacity [8]. Hence chitin and chitosan are widely used in cosmetic, food, agricultural, tissue engineering [9], waste water treatment [10] and packaging material applications [11].

The concept of green methodology in waste valorization refers to the conversion of shrimp processing waste into value added products using Environmentally sustainable processing methods. As chitosan is a valuable biomaterial there is necessity in developing sustainable technologies for converting waste into valued byproduct. In this study chitosan is produced from shrimp processing waste which minimize the use of harsh chemicals and utilize ecofriendly reagents such as citric acid (demineralization), papain enzyme(deproteinization), hydrogen peroxide (decolorization), and a deep eutectic solvent (DES) (deacetylation). Production of chitosan from shrimp processing waste aligns with sustainable bioeconomy, waste valorization and reducing environmental burdens [12, 13].

Pure chitosan possesses few limitations in direct industrial and clinical applications as it is p^H dependent, soluble only in acetic acid, rigid crystalline structure, high glass transition temperature. Chitosan on fabrication into a hydrophilic polymer such as PEG which is water soluble and flexible and acts as an ideal structural modifier. This improves the mechanical ductility and flexibility of the composite as highly mobile ether chains of PEG are inserted between the rigid chitosan strands. Composite of Chitosan and PEG as hydrogels, beads and film are used in drug delivery, wound dressing and structural tissue engineering scaffolds. Hence, this composite material bridges the gap between bioactivity of natural polymers and mechanical strength of synthetic polymers [14, 15, 16].

EXPERIMENTAL MATERIALS AND METHODS

Synthesis

The raw shrimp processing waste was treated in two stages which includes

1. Sample collection
2. Isolation of chitin and chitosan by green methodology.

Isolation of chitin and chitosan is carried in four stages a. Demineralization with citric acid b. Deproteinization with papain enzyme c. Decolorization using hydrogen peroxide d. Deacetylation with Deep Eutectic Solvent [17–19].

1. Sample collection and preparation: shrimp shell waste was collected and brought to laboratory. Shrimp shell was washed thoroughly with water to get rid of impurities. The cleaned shells were dried in sunlight for three days. These dried wastes were crushed into small pieces in a mixer and stored in air tight container.
2. Extraction of chitin and chitosan by green method: 10g of the dried powder was used for the extraction process which is carried in four stages: Demineralization, Deproteinization, Decolorization and Deacetylation.
    a. *Demineralization by citric acid:* It is the first step of the chemical process involving the stripping of Calcium carbonate and other minerals present in shrimp shell waste. 10g of powdered shrimp was treated with 2% citric acid solution agitated at standard temperature. The treated sample is washed with water to maintain neutral P^H.
    b. *Deproteinization using papain:* It is the second step in the process which involves the removal of protein from the shells. The treated shrimp shell was now treated with papain solution (0.5–1% w/v) at alkaline pH under continuous stirring at temperature of 60^0C. The resulting precipitate was washed to neutrality using copious amounts of distilled water and dried overnight in hot air oven to get chitin.
    c. *Decolorization using hydrogen peroxide:* The residue which is obtained is decolorized by treating with 15% Hydrogen peroxide at 60^0 C in stirring conditions for 1 hour. The resulting precipitate was washed to neutrality using copious amounts of distilled water and dried overnight in hot air oven to get chitin.
    d. *Deacetylation using DES:* Dried powder is treated with 10ml of Deep Eutectic solvent

(choline chloride and urea in a molar ratio of 1:2) under stirring conditions at temperature of 80^0C for 6hours.The resulting precipitate was washed to neutrality using copious amounts of distilled water and dried at 50^0C for one day. The obtained chitosan is stored in air tight container.

SYNTHESIS OF CHITOSAN-PEG COMPOSITE FILM

Composite films were prepared from commercial chitosan and chitosan from shrimp processing waste, incorporating polyethylene glycol (PEG) by Solvent casting technique which is simple, cost effective and widely used [20–22]. Chitosan powder was dissolved in 1% acetic acid solution under continuous stirring for 24 hours at room temperature. PEG and glycerol were added to the solution as plasticizers. The mixture was stirred for another 1-2 hours for blending. The solution was degassed and slowly poured into a petri dish ad dried in hot air oven at 40^0C for 24 hours. Finally, the composite films were carefully peeled from the petri dish.

RESULTS AND DISCUSSION

The physicochemical properties of the synthesized chitosan summarized in Table 1 comparison with those of commercial chitosan. The synthesized chitosan exhibited a degree of deacetylation (DD) of 95.58%, which is considerably higher than the minimum value (>75%) generally reported for commercial chitosan. Synthesized chitosan was completely soluble in 1% acetic acid as commercial chitosan. The synthesized chitosan achieved a yield of 60.7% which is higher than the commercial chitosan obtained from shell waste. The moisture content of the synthesized chitosan was found to be 14%, which is within the acceptable range of 5–15% for commercial chitosan. The ash content of the synthesized chitosan was 11%, which is considerably higher suggests the presence of residual minerals like calcium and phosphorus in the sample.

UV-VISIBLE SPECTRA OF CHITOSAN FROM SHRIMP PROCESSING WASTE (SAMPLE)

The UV-visible spectrum of chitosan from shrimp processing waste as shown in Figure 1 was recorded in the wavelength of 200-400nm range. A peak was observed at 277nm, which is attributed due to the $n \rightarrow \pi^*$electronic transition of the amide (–CONH–) and amino ($–NH_2$) functional groups in the polymer chain [23, 24].

IR SPECTRA OF CHITOSAN FROM SHRIMP PROCESSING WASTE (SAMPLE)

The IR spectrum of chitosan from shrimp processing waste as shown in Figure 2 has peak at 3365 cm^{-1} correspond to the stretching vibration of O–H/N–H stretching. The band observed at 2877 cm^{-1} correspond to asymmetric stretching of CH_2. Band at 1648 cm^{-1} and 1457 cm^{-1} correspond to Amine I C = O stretching and Amine II NH_2 bending. Band at 1070 cm^{-1} is attributed due to stretching vibration of C – O groups [25].

EDS ANALYSIS

EDS analysis of Commercial Chitosan from LOBA CHEMIE as shown in Figure 3 contain Carbon - 46.30%, Oxygen – 48.75% and Calcium – 4.96% indicating the presence of organic constituents and some amount of Calcium confirming the presence of calcium-based minerals.

Table 1. Physicochemical characteristics of synthesized chitosan.

S.N.	Parameter	Synthesized Chitosan	Typical purified Chitosan
1	Degree of deacetylation	95.58%	>75%
2	Solubility in 1% acetic acid	Soluble	Soluble
3	Yield	60.7%	15–40% (from shell waste)
4	Moisture content	14%	5–15%
5	Ash content	11%	<1–2%

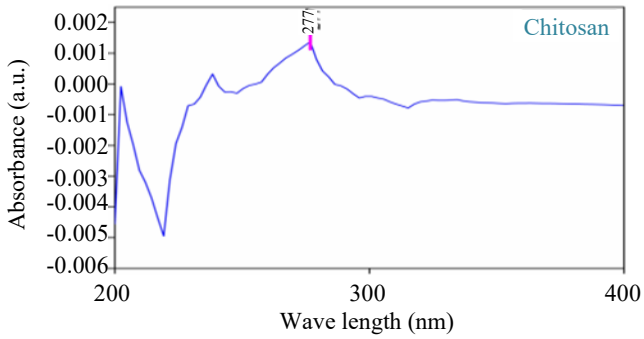

Figure 1. UV-spectra of Chhitosan (Sample).

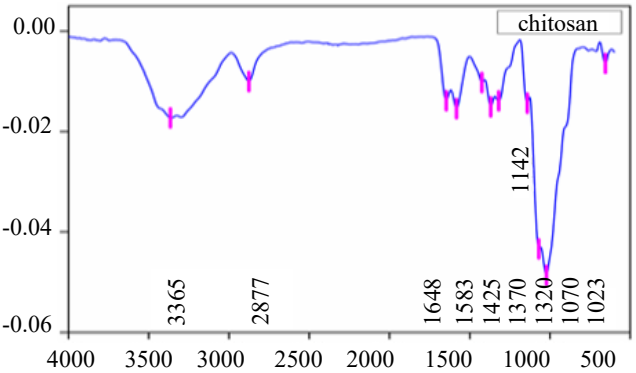

Figure 2. IR spectra of Chitosan (Sample).

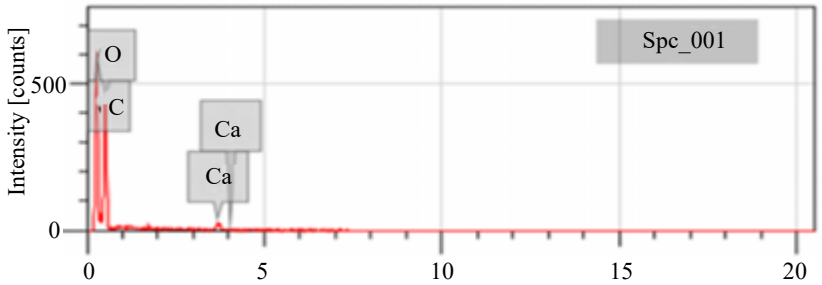

Figure 3. EDS of Chitosan (LOBA CHEMIE).

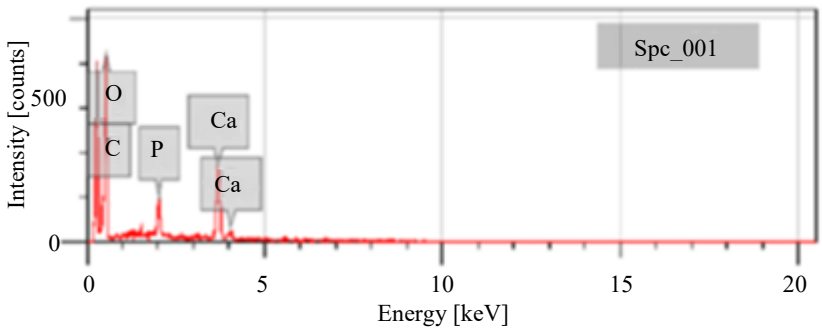

Figure 4. EDS of Chitosan (Sample).

The elemental composition as determined by EDS analysis, reveals that Chitosan from shrimp processing waste (Sample) as shown in Figure 4 primarily consists of Carbon, Oxygen, Calcium and phosphorus with weight percentages of 28.18%, 40.55%, 26.07% and 5.20% respectively. This elemental composition is consistent with the molecular structure of Chitosan, which primarily consists of Carbon chains with oxygen and phosphorus with some amount of calcium content within the chitosan matrix.

SEM ANALYSIS

SEM image of commercial chitosan as shown in Figure 5 showed a rough and compact surface with interconnected fibrous structures with pores which indicates uniform film formation and good compatibility among the film constituents.

SEM image of chitosan from shrimp processing waste (Sample) as shown in Figure 6 shows an irregular and agglomerated surface morphology with rough textured regions and dense particle clusters. The heterogeneous morphology indicates the presence of inorganic mineral phases dispersed within the organic matrix. The bright regions observed in the micrograph may correspond to calcium-rich domains, which is consistent with the EDS results confirming Ca and P elements.

CHARACTERIZATION OF COMPOSITE FILMS BY FTIR AND THICKNESS ANALYSIS

The FTIR spectrum of composite prepared from chitosan commercial and PEG as shown in Figure 7 showed absorption bands at 3359, 2879, 1645, 1558, 1413, 1039 cm^{-1} and the composite prepared using chitosan from shrimp processing waste (Sample) and PEG as shown in Figure 8 showed bands at 3361, 2881, 1645, 1558, 1409, 1041 cm^{-1}. Based on above two FTIR spectrum values, we conclude that both are giving approximately values corresponding to O–H/N–H stretching, C–H stretching, amide I, amide II, and C–O–C stretching vibrations, respectively. The composite film exhibited slight shifts in the amide and C–N stretching regions compared with standard/commercial chitosan, indicating intermolecular hydrogen-bonding interactions between chitosan and polyethylene glycol (PEG). The preservation of major absorption bands demonstrated that the fundamental chemical structure of chitosan remained intact after composite formation.

Thickness measurements revealed that the commercial chitosan–PEG film exhibited a thickness of 0.02 mm whereas shrimp-derived chitosan–PEG film possessed a thickness of 0.08 mm**.** Although, similar quantities of materials were used during film preparation, the higher thickness of the shrimp-derived film may be attributed to differences in molecular weight, degree of deacetylation, viscosity, and residual biopolymeric constituents present in the extracted chitosan. The increased thickness indicates a greater solid content and stronger polymer network formation during solvent evaporation.

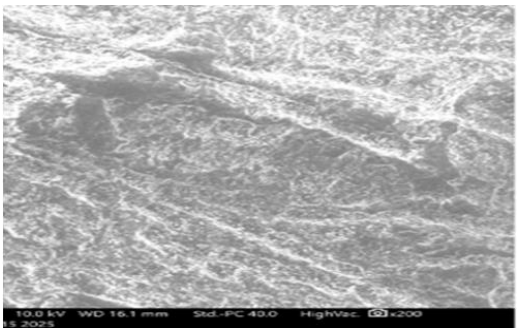

Figure 5. SEM image of Chitosan (LOBA CHEMIE).

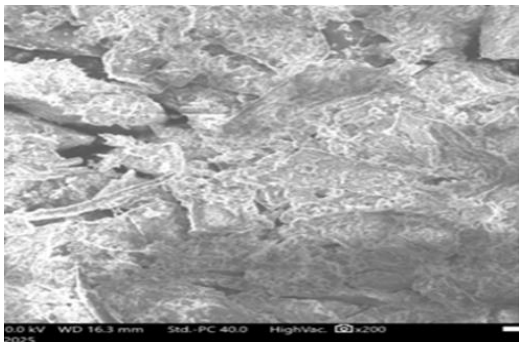

Figure 6. SEM image of Chitosan (Sample).

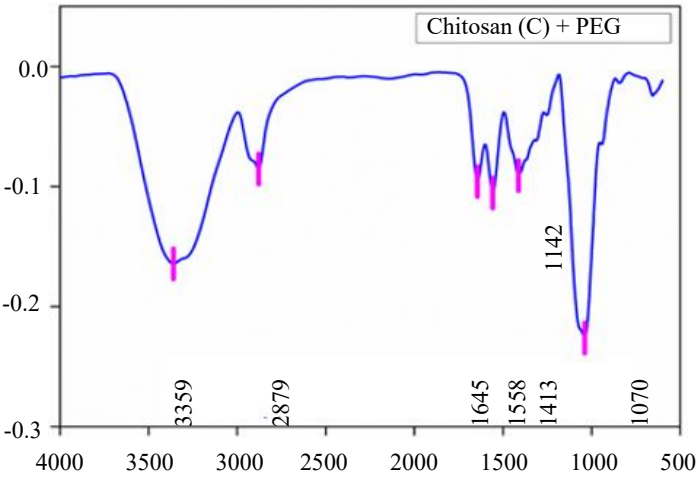

Figure 7. IR-Chitosan (LOBA CHEMIE) and PEG.

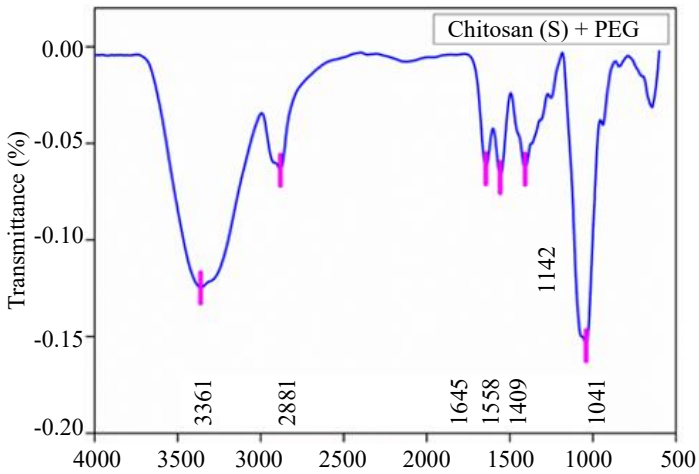

Figure 7. IR-Chitosan (Sample) and PEG.

Visual observation of the films showed the formation of transparent, continuous, and self-supporting films, indicating good compatibility between chitosan and PEG. The presence of abundant hydroxyl and amino groups, together with PEG ether linkages, suggests enhanced hydrophilicity, flexibility, film-forming ability, and potential biocompatibility of the composite material.

Overall, the results demonstrate that shrimp shell waste can serve as an effective and sustainable source of chitosan for the fabrication of biodegradable chitosan–PEG composite films. Chitosan–PEG composite films possess excellent biodegradability, biocompatibility, flexibility, antimicrobial potential, and film-forming ability, making them attractive materials for food packaging, biomedical, wound-healing, and environmental applications. Chitosan-PEG composite film is used in sustainable food packaging materials due to their biocompatibility and moisture retention [26–28]. In wound dressing due to their biocompatibility and moisture retention [29, 30]. In water purification and waste water treatment membranes for absorption of dyes and heavy metals [31]. In drug delivery due to their biocompatibility and biodegradable [32].

CONCLUSION

Chitosan was successfully synthesized from shrimp processing waste by green methodology where the extraction of chitin and chitosan is carried in four stages a. Demineralization with citric acid b. Deproteinization with papain enzyme c. Decolorization using hydrogen peroxide d. Deacetylation with Deep Eutectic solvent. In this study, the extraction procedure presents a sustainable solution to one of

the industry major waste disposal challenges. Further UV, IR, EDS, SEM analysis confirmed that the extracted chitosan is effective in achieving standard quality, which can be applied widely used for a wide range of applications in various fields.

Chitosan extracted from shrimp shell waste was successfully blended with PEG to produce transparent composite films. FTIR analysis confirmed the presence of characteristic chitosan and PEG functional groups and indicated hydrogen-bonding interactions between the two polymers. The presence of calcium and phosphorus in the extraction of chitosan did not affect in the film formation. These elements remain as dispersed minerals in micro-domains within the organic polymer matrix. This is confirmed by the IR-spectra of Chitosan –PEG composite where the functional groups remain unaltered, showing that the mineral constituents remained inert within the polymer matrix during the film blending.

The shrimp-derived chitosan–PEG film exhibited a higher thickness (0.08 mm) than the commercial chitosan–PEG film (0.02 mm), suggesting enhanced polymer network formation. The results demonstrate that shrimp shell waste-derived chitosan is a promising sustainable alternative to commercial chitosan for biodegradable film applications used in food packaging, wound healing, biomedical devices, water purification. Hence this study highlights the conversion of sea food processing waste into biomaterial.

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