

Tridax-Procumbens Extraction Impregnated Chitosan Film: Preparation Characterisation and Application

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Abstract

Chitosan and its derivatives has variety of advantages likes in industrial and biomedical fields. In recent years, research has been focused on biodegradable films of natural polymers like chitosan, starch, cellulose and etc. Such polymers may be proteins of plant and biological materials. Natural biopolymeric films with tridaxprocumben have potential advantage over the synthetic products since they are totally biodegradable from biological materials. Tridaxprocumbens extract is used for wounds healing as anticoagulant, antifungal and insect repellent. It is well known for application to treat infection and skin diseases in folk medicines, etc. The present study was focused on impregnation of tridax-procumbens extract into chitosan to develop a antimicrobial films. Further, they assessed was for physicochemical properties through x-ray diffraction and fourier transform infrared (FTIR) spectroscopy studies and antimicrobial activity.

Keywords: Chitosan, chitosan / Tridaxprocumbens blend, Antimicrobial activity

Introduction

In the last few years, there has been growing interest in bio based polymer packaging products made from raw materials and originating from natural, agricultural, marine and livestock raising and renewable sources^[1]. Therefore, chitosan not only refers to a uniquely defined compound but also refers to a family of copolymers with various fractions of acetylated units^[2].

Chitosan, which is the N-deacetylated form of chitin, is derived from chitin by deacetylation. This means that the acetamide groups in chitin are substituted into amino group in chitosan. Chitosan forms complexes with metal ions. As chitosan is a derivative of chitosan, it shares many similar properties with chitin. Like chitin, chitosan has a rigid configuration, easily filmable and spinnable and is insoluble in alkaline solution and some organic solvents. However the main difference between chitin and chitosan is that chitosan is soluble in dilute acids, e.g. formic and acetic acids, and its salt is water-soluble. Thus chitosan finds much wider applications in industry than chitin.

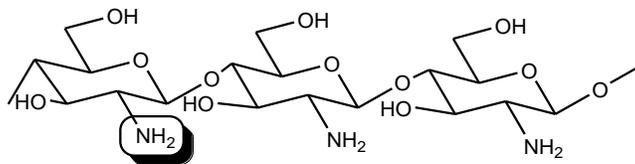


Figure-3 Molecular structure of chitosan

Most applications discussed in the following sections are still in the experimental stage, with only a few commercial products available in the marketplace. However, the future of chitin and its derivatives appears to be very promising as both have several interesting characteristic features, which make them suitable for numerous applications in industry. It is the following materials and technologic: They are biodegradable, breaking down slowly to harmless carbohydrates, carbon dioxide and water. They are non-toxic biopolymers. They are biologically reproducible since they represent a major component of biomass. They are remarkably stable in concentrated alkaline solutions even at high temperatures. They have amino and hydroxyl groups which are chemically modifiable. They are highly biocompatible, not only in animal but also in plant tissues. They can be processed into gels, beads, powder, fibers, colloids and films. In some cases, by synergistic effects, the blend provides better properties than the pure components [3,4]. Since chitosan has many functional groups, it can be modified by blending with other polymeric materials which is expected to be useful for some applications. The formation of polymeric blends constitutes a perspective way of making materials with new properties, especially from natural polymers that are of special importance. Numerous investigations have been reported on the studies of films made from chitosan [5-9] and chitosan blends with natural polymers [10,11-14] or synthetic polymers [15,16].

MATERIALS AND METHODS

CHEMICALS AND SAMPLES:

Original chitosan was purchased from central institute of Fisheries Technology (CIFT) Cochin, Kerala with viscosity average molecular weight about 3,00,000 k. Lactic acid, tween 20 and tridaxprocumbens extraction were obtained from Islamiah College (Autonomous), Vaniyambadi.

INSTRUMENTS:

The following instruments were used in analysis of collected sample and all the spectra were taken at room temperature;

FT-IR:

FT-IR spectra analyses of CS1, CS2 and CS3 in 4000-400 cm^{-1} wave length range, using ATR method. Where st'peter's university Avadi Chennai.

Powder X-Ray Diffraction:

Powder X-ray diffraction (XRD) analysis were recorded at Technology Business Incubator-VIT, Vellore.

Antimicrobial activity:-

Antimicrobial analysis was performed using standard agar well diffusion method to study the antibacterial and antifungal activities of the compounds. Each bacterial and fungal isolated was suspended in Brain Heart infusion (BHI) broth and diluted to approximately 10^5 colony forming units (CFU) per ml. They were flood-inoculated onto the surface of BHI agar and then dried. Five millimeter diameter wells were cut from the agar using a sterile cork-borer and 30 μ L of the sample solution were poured into the wells. The plates were incubated for 18 h at 37 $^{\circ}$ C for bacteria and at room temperature for fungi. Antimicrobial activity was evaluated by measuring the zone of inhibition in mm against the test microorganisms. DMG was used as solvent control.

Chitosan solution preparation/tridaxprocumbens extraction blend

Chitosan solution (1% w/v) was prepared using 1% lactic acid after complete mixing and stirring in 10 min. Extraction of *Tridax procumbens* from plant collected and extraction was filtered with Whatman number 1 paper and used for further studies. Different percentages of *Tridax procumbens* extraction (0%, 1%, 2%, 3%) and chitosan solution mixed and was homogenized using a sonicator for 10 minutes. The film solution was cast over the evenly leveled plate and vacuum dried for three days at room temperature.

Results and Discussions

Fourier Transform Infrared spectral analysis

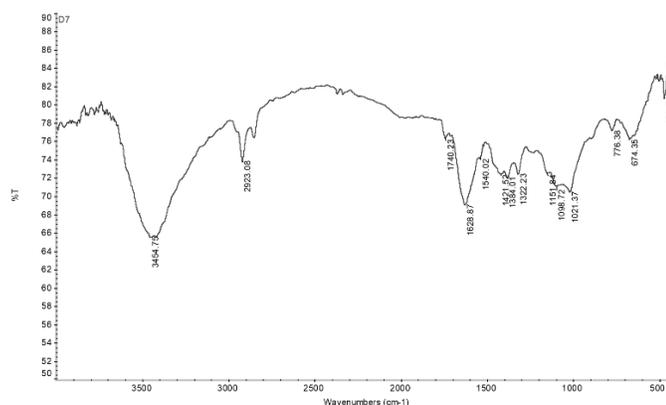


Figure-1 : FTIR spectra of Pure Chitosan

The FTIR spectra of pure chitosan (Figure-1) showed a strong absorption band at 3454 cm^{-1} due to OH and amine N-H symmetrical stretching vibrations. A peak at 2923 cm^{-1} was due to symmetric $-\text{CH}_2$ stretching vibration attributed to pyranose ring. A peak at 1156 cm^{-1} was assigned to the structure of saccharide. The sharp peak at 1384 cm^{-1} was assigned to CH_3 in amide group. The broad peak at 1021 and 1098 cm^{-1} indicated the C-O stretching vibration in chitosan and peaks at 1628 and 1540 cm^{-1} were due to C=O stretching (amide I) and NH stretching (amide II). The absorption bands at 1151 cm^{-1} was assigned to the anti-symmetric stretching of C-O-C bridge, and 1098 and 1021 cm^{-1} were assigned to the skeletal vibrations involving the C-O stretching.

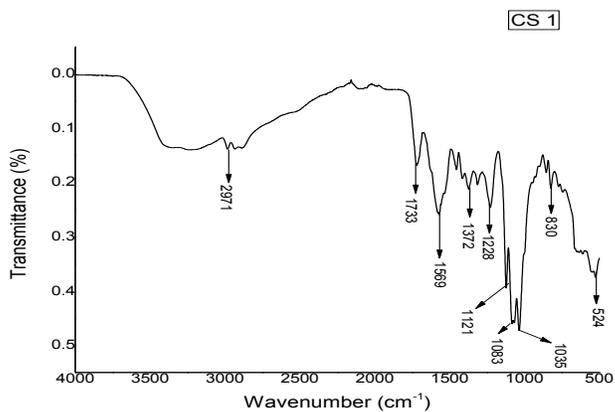


Figure -2 : FTIR spectra of CS1 (1%)

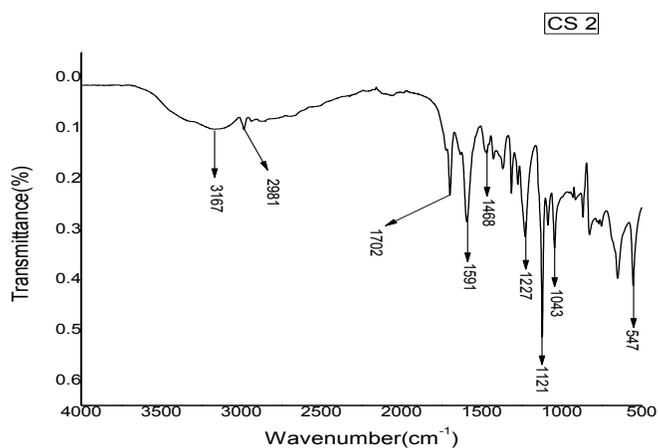


Figure-3 : FTIR spectra of CS2 (2%)

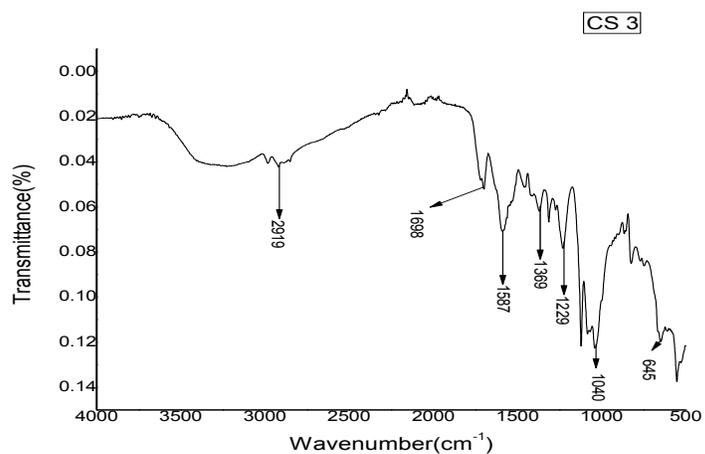


Figure -4 : FTIR spectra of CS3 (3%)

X-Ray Diffraction Studies

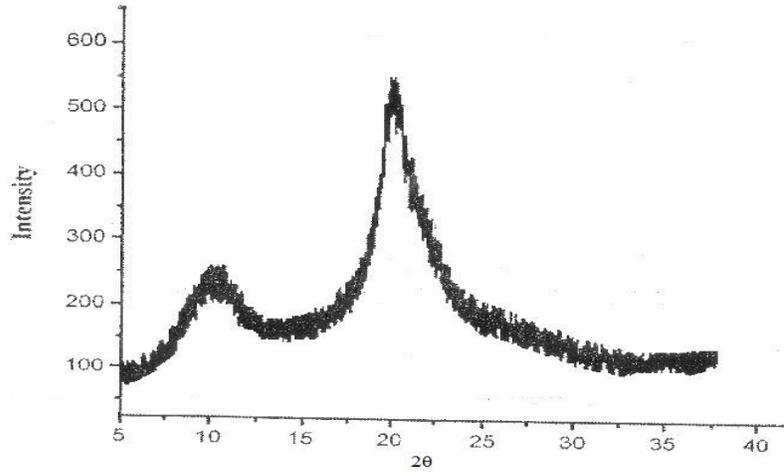


Figure-5 : X-ray diffraction pattern of pure chitosan

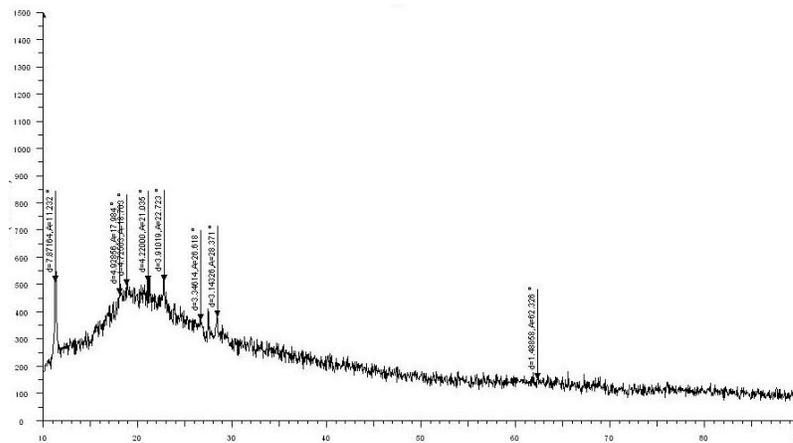


Figure-6 : X-ray diffraction pattern of CS1

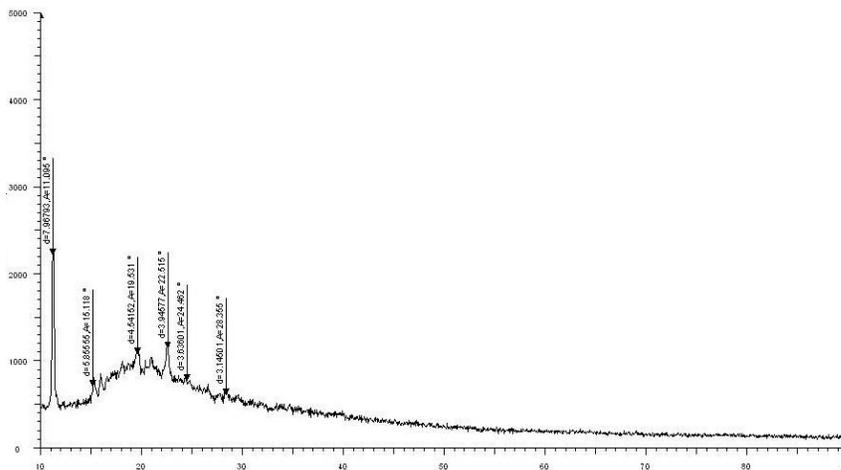


Figure-7 : X-ray diffraction pattern of CS2

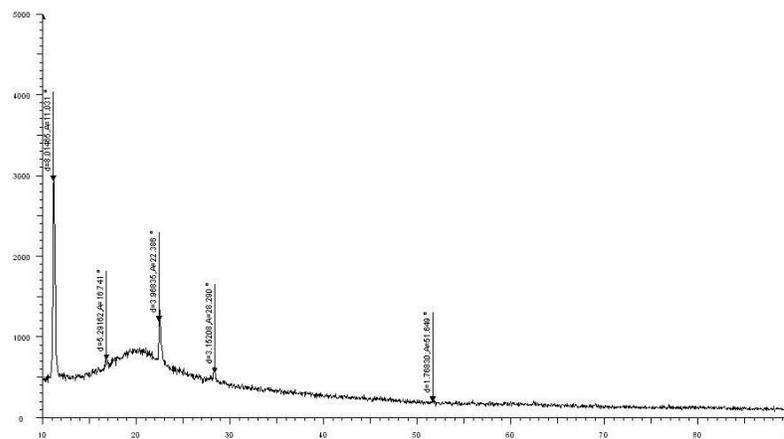


Figure-8 : X-ray diffraction pattern of CS3

Figure-5 shows the X-ray diffraction pattern of pure chitosan which showed distinct crystalline peaks at around 2θ values 100 and 200. This is because of presence of plenty of –OH and –NH₂ groups in the chitosan structure, which could form stronger inter and intramolecular hydrogen bonds and the chitosan structure has certain regularity, so that the molecules form crystalline regions easily which was similar to the results reported by Duan and his co-workers.

Figure-6 shows the X-ray diffraction pattern of CS1. The 2θ values at 100 is remain same as chitosan. But the nitrogen peak of 2θ value 200 is decreased and broad peak was obtained. It indicates that crystalline nature of chitosan was decreased. Whereas the amorphous nature was increased. This result indicate that there is an interaction between the extract and the chitosan. Which make the disappearance of inter and intramolecular hydrogen bonds in chitosan. Due to that the chitosan cost it's crystallinity and becomes amorphous. The same peak is present in CS2 and CS3.

Antimicrobial Activity

The antimicrobial activity of the chitosan derivatives against some microorganisms was studied. The bacteria selected for the study were *Escherichia* and *Staphylococcus aureus* and Kp. The results are shown in Table: 1. In the case of bacterial and fungus control, the synthesized drugs showed considerable activity but less than the standard drug. Among the tested microbes, chitosan blend showed higher activity against *S.aureus* (gram positive) bacteria. Generally, the modified chitosan were found to be more active towards fungal and bacterial species. Between the CS1, CS2, and CS3, later one showed slightly better antimicrobial activity than the former one. This may be due to the in surface modification of the blend during the blending process

| ORGANISM | Zone of inhibition (mm) | | | | |
|-----------------------|-------------------------|----------|-----|-----|-----|
| | Control | Chitosan | CS1 | CS2 | CS3 |
| E.coli | 0 | 15 | 17 | 19 | 21 |
| Staphylococcus aureus | 0 | 13 | 15 | 18 | 19 |
| Kp | 0 | 18 | 20 | 21 | 22 |



Figure-9 Antimicrobial Activity of E.coli
(1.Chitosan, 2.CS1, 3. CS2, 4. CS3)



Figure-10 Antimicrobial Activity of Staphylococcus aureus
(1.Chitosan, 2.CS1, 3. CS2, 4. CS3)

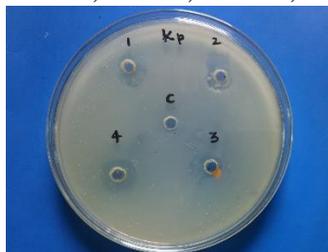


Figure-11 Antimicrobial Activity of Kp
(1.Chitosan, 2.CS1, 3. CS2, 4. CS3)

CONCLUSION

Tridax-procumbens extraction impregnated chitosan films were successfully developed by varying the concentration of the extract. Further the film were characterised by FT-IR and XRD carried out to study the interaction between the chitosan and tridax-procumbens extraction films. It was observed that the increasing concentration of extraction. The prepared blend chitosan film was characterized by FT-IR . The crystally nature of the chitosan was studied by XRD. The XRD data revealed that the crystal nature of the derivatives were decreased by the increasing the concentration of the extract. The antimicrobial activity of the chitosan derivatives were investigated against some bacteria and fungi. It was found that the derivatives have a significant effect against all the microorganisms. Among the prepared film CS3 should higher antimicrobial activity that the pure chitosan, CS1 and CS2. This may be due to the presence of antimicrobial in the extract. This would encourage the use of these biopolymeric chitosan derivatives as effective antimicrobial agents. The work clearly indicated that chitisan derivative and its blend could be used as antimicrobial agents in food packging materials after further studies.

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