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Miscibility and Thermal Study of 4-Hydroxycoumarin Doped Chitosan Films

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ABSTRACT

In this study, new Chitosan/4-Hydropxycoumarin films were prepared and characterized. The influence of 4-Hydroxycoumarin on the surface morphology, thermal behavior of the chitosan films were studied. The experimental studies showed that surface morphology becomes uniform and surface roughness increases with increasing the concentration of 4-Hydroxycoumarin in the chitosan film. The appreciable intermolecular interaction among the chitosan and 4-Hydroxycoumarin is confirmed by the FTIR study. The thermal properties of the chitosan films slightly increased with the increasing concentrations of 4-Hydroxycoumarin. Also, the presence of single glass transition temperature in all Chitosan/4-Hydroxycoumarin films suggests the components present in the film were miscible. Due to the existence of 4HC into the chitosan film, the crystalline nature and water contact angle of CS decreases for the C4HC films. It can be expected that, the best properties of Chitosan/4-Hydroxycoumarin films were recorded in the study may play a vital role in food packaging and coating applications.

Keywords: Surface morphology, thermal behaviour, glass transition temperature, miscibility

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INTRODUCTION

In recent years, biodegradable polymers including cellulose, starch, chitosan and natural gums have been studied extensively to produce potential biodegradable packaging material with enhanced physicochemical properties. Nowadays there is an urgency to manufacture biodegradable and environmental friendly bio-based polymeric material [1] for active food packaging. Active food packaging is made to interact with product or its environment to extend shelf life of food. The basic raw materials for film forming and coatings can be obtained from the natural sources including starch, cellulose, proteins, polysaccharides, lipids and resins [2] which act as excellent barrier to oxygen, water vapour and oil [3].

Among natural biopolymers, chitosan (CS) is a natural cationic polysaccharide with active amino functional groups. CS is the deacetylated product of chitin which found most abundant polysaccharide in nature [4-5]. CS is a copolymer of N-acetyl-D-glucosamine and D-glucosamine. Besides its biodegradability and biocompatibility, CS has been widely reported that it has proven to have good antimicrobial property [6] against bacteria, yeasts and fungi [7-8]. Several approaches have been undertaken to get over the limitations of CS including polymerization and blending with other polymers which paid much attention to alter or tailoring the property of interest. CS, its own or blend component is used as a biomaterial [9-11] in water treatment, in food packaging and medicine [12-19].

4-Hydroxycoumarins (4HC) is one of the important precursors in the realm of organic synthesis. The interest towards 4HC has been amplified because, it is not only significant synthetic endpoints [20-21], but it contains structural nucleus of many natural products [22-24]. The derivatives of 4HC have shown a remarkably applications in pharmacological and physiological activities. The derivatives of 4HC are used as anticoagulant, antibacterial, antifungal, antitumor, antioxidant, anti-inflammatory agents [25-31]. Also, in recent years there are references to derivatives with HIV protease inhibitors [32]. In addition, 4HC is an important fungal metabolite and its production leads to further fermentative production of the natural anticoagulant dicoumarol. The dicoumarol is a fermentation product found in spoiled sweet clover silages and is considered a mycotoxin [33]. The study aims to prepare 4HC doped CS films. Also the present work intended to explore the influence of 4HC on CS films.

MATERIALS AND METHOD

Materials

The materials used in this study are chitosan from shrimp shells 75% (deacetylated) with viscosity min 200 cps was purchased from Loba Chemie Pvt. Ltd, Mumbai, India. 4-Hydroxycoumarin

(Sigma Aldrich), Acetic acid (Spectrochem Pvt. Ltd. Mumbai. India) and millipore water was used.

Preparation of the Chitosan Films

CS films were prepared by doping different concentration of 4HC onto the CS solution using solvent casting method and the composition for CS and CS/4HC films is shown Table 1. An exactly weighed (2 g) amount of CS was dissolved in 150 mL of 2% acetic acid. To the CS solution different concentrations of 4HC (0.02 g to 0.08 g dissolved in 5 mL 100% acetic acid) were mixed and the mixture of CS and 4HC (C4HC) was stirred for 3-4 h. Then, subsequently, definite volumes of homogeneous C4HC film solutions were poured onto the previously cleaned and dried petri dishes and left for solvent evaporation at normal room temperature for a couple of weeks. After ensuring evaporation of solvent, the films were peeled from petri dishes and stored in vacuum desiccators for further characterization.

Table 1: Composition table of CS and CS/4HC films.

Sample Code	Wt of Chitosan	Wt of 4HC
CS	2 g	00 g
C4HC-1	2 g	0.02 g
C4HC-2	2 g	0.04 g
C4HC-3	2 g	0.06 g
C4HC-4	2 g	0.08 g

CHARACTERIZATIONS

Atomic Force Microscopy (AFM)

The surface morphology of the composite film was recorded using Atomic force microscopy (AFM) using Nanosurf Easyscan2, (Switzerland) with the aluminum coated cantilever. All the topographic images of film samples were collected in contact angle mode using aluminum coated cantilever. The topographic images of film samples were taken and roughness of the films was analyzed.

Fourier Transform Infrared (FTIR) Spectroscopy

The Fourier transform infrared spectroscopy was used to probe the interaction among the components and films samples were screened for interaction using an ATR (attenuated total reflection) method of IR spectrometer (Perkin-Elmer Spectrum Version 10.5.4). All the film specimens were scanned between the 550 cm^{-1} to 4000 cm^{-1} at 4 cm^{-1} resolution.

Differential Scanning Calorimetry (DSC)

The DSC measurements were carried by using DSC Q20-V24.4 Build 122 system (TA Instruments, USA). The instrument has balance sensitivity and in the alumina pans samples were loaded and reference pan kept empty for film sample analyis. In addition, pan was heated under nitrogen atmosphere and a heating rate of the sample was kept 10^{o}C/min.

Thermogravimetric Analysis (TGA)

A thermogravimetric analysis technique (SDT Q600 V20.9 Build 20 –Universal V4.5A TA Instruments) was used to provide weight loss which is useful for the study of thermal stability of films. The film samples of masses 5 to 6 mg were used and heated in an inert nitrogen atmosphere (heating rate of 10°C/min) from ambient temperature to 600°C. The weight losses at different stages were analyzed from the curves of TGA.

The X-ray Diffraction (XRD)

XRD analysis of the films was carried out using a Rigaku SmartLab (Tokyo, Japan) X-ray diffractometer. A Cu K-beta radiation was used with working voltage 40 kV and current 30 mA. The scan was performed with continuous mode in the 2θ range from 5^{O} to 80^{O} and speed was 5^{O} min^{-1}.

Water Contact Angle (WCA) Measurements

The water contact angles of the films were measured by the drop method using a contact angle meter Model DMs-401 (Kyowa Interface Science Co. Ltd., Tokyo) to examine the hydrophilicity. A drop of millipore water was carefully dropped on the film surface, and the contact angles were measured. Each reported contact angle is the average value of three measurements.

RESULTS AND DISCUSSION

Atomic Force Microscopy

The surface morphology of the pure CS and C4HC films were analyzed by using atomic force microscopy and obtained topographic images with their 3D view were shown in Figure 1. For the topographic image of CS, 3.37 mV roughness was found. The results of AFM study indicates that, the roughness of C4HC films (0.02-0.08 g 4HC) was increase when compared to that of pure CS film, begins to alter roughness. This could be due to the distribution of 4HC in CS which influenced on viscosity of the CS film solution. After addition of 4HC, the area roughness slightly increased which confirmed that 4HC is less compatible with CS.

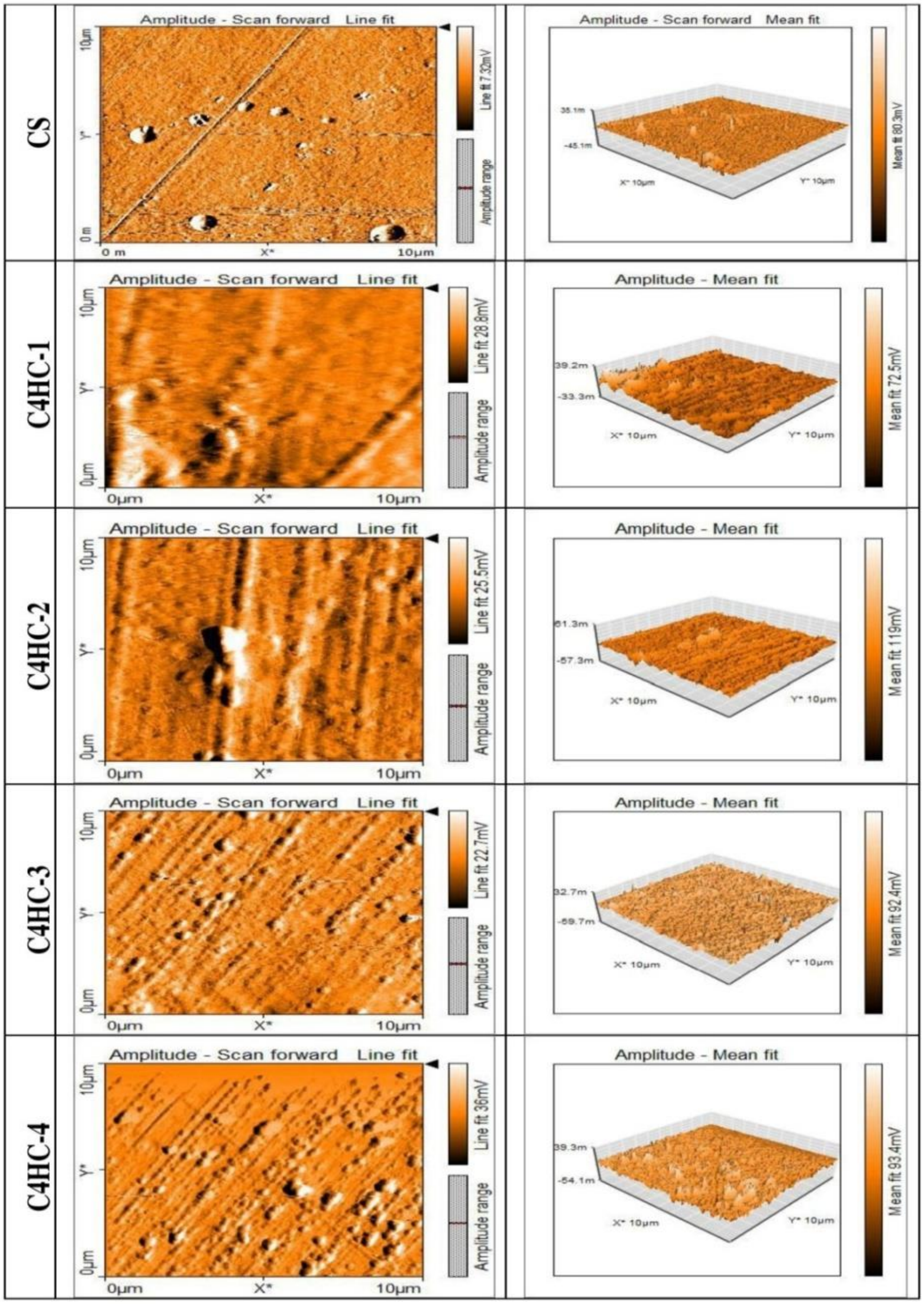

Figure 1: AFM topographic images and their 3D views.

Fourier Transform Infrared Spectroscopy

Fourier transform infrared spectroscopic analysis was carried to confirm the interaction among components. The FTIR spectra of CS and C4HC films are shown in Figure 2. The spectra of pure chitosan film shows a broad band at 3365 cm^{-1} which is due to the OH and NH hydrogen band stretching, & 2852 cm^{-1} CH stretching, 1642 cm^{-1} amide-I and 1029 cm^{-1} CO stretching vibration. The band at 1558 cm^{-1} is assigned for the NH bending (amide-II). The bands at 2921, 1412 and

1316 cm^{-1} are assigned to CH_2 bending due to pyranose ring. The FTIR spectra of 4HC shows peak at 3355 cm^{-1} which is due to the OH stretching and 1705–1599 cm^{-1} attributed to the –C=O vibrational stretching. The peak observed at 2981 attributed to the aromtatic –C-H stretching. The FTIR spectra of C4HC composite films showed changes in the peak value indicating that interaction among the CS and 4HC. The –OH observed in the CS is shifted to the lower value (3365 cm^{-1} to 3257 cm^{-1}) in C4HC films. This might be due to the hydrogen bonding in the films which leads to the intermolecular interaction. Also peak observed in the CS (2921 cm^{-1}) appeared at higher level and peak appeared at 1642 cm^{-1} shifted to the lower level (1636 cm^{-1}) in C4HC films. The shift in the peak value and peak intensity confirms that there is considerable interaction among the components.

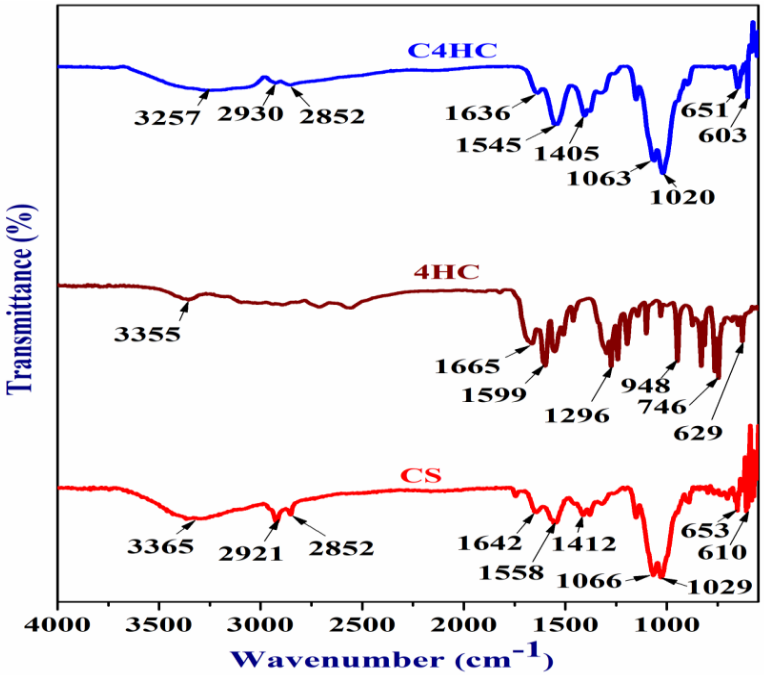

Figure 2: FTIR spectra of CS and C4HC films.

Thermogravimetric Analysis

The stability of the films were evaluated by using thermogravimetric analysis (TGA). Thermal distraction of chitosan presented two significant weight losses as shown in Figure 3. Initial weight loss observed at 41°C to 75°C due to loss of moisture and bound water (19.52 %). The second major weight loss of 49.57 % was observed at 259°C to 325°C, this could be attributed to the decomposition of saccharide structure present in the chitosan. Also, the incorporation of 4HC onto the CS presented two step degradation patterns. The increased thermal stability observed in the C4HC films. It is worth noting that, remarkable changes were noticed in C4HC series. The initial weight loss observed from 39°C to 156°C, this could be due to the evaporation of physically bonded water molecules. The maximum weight loss with 50 % to 54 % was observed between the temperature ranges 258^{o}C to 330^{o}C which is considerably higher than pure CS. This could be due to the fully destruction of films. The strong intermolecular interaction leads to the increased thermal stability of the film. This fact could indicate a good interaction between CS and 4HC. The results of TGA were good agreement with FTIR study.

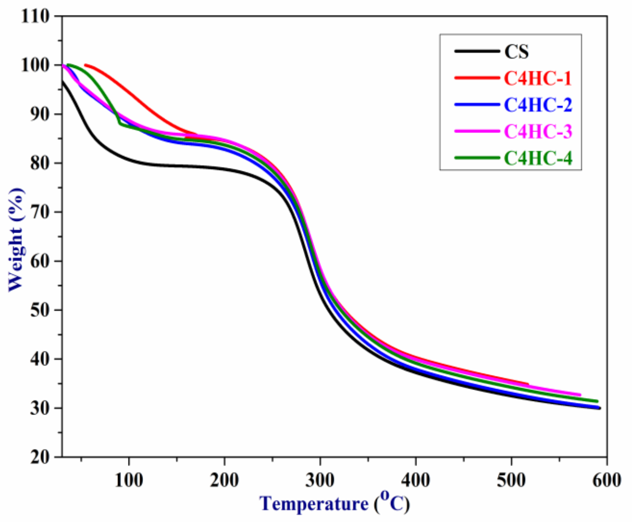

Figure 3: Thermogram of CS and C4HC films.

Differential Scanning Calorimetry

The graphs of DSC thermograms for CS and C4HC films were shown in the Figure 4. The importance of T_g can be realized in the study of miscibility of components. Moreover, the miscibility of the films depends upon the composition and the solvent used. In the C4HC films, T_g significantly decreased to lower value 45.39^{o}C, 48.79^{o}C, 49.99 ^{o}C and 53.39 ^{o}C for C4HC-1, C4HC-2, C4HC-3 and C4HC-4 respectively, this could be due to the considerable interaction among the CS and 4HC which indicates composite films were miscible.

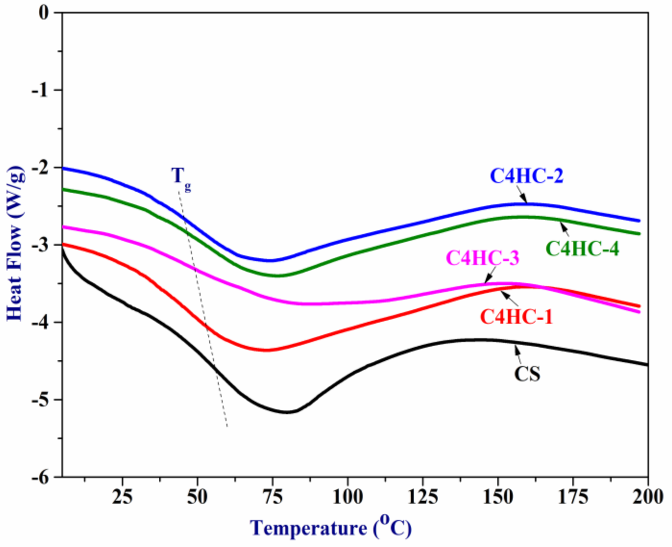

Figure 4: DSC Thermogram of CS and C4HC films.

X-ray Diffraction study

The X-ray diffraction patterns of CS and C4HC films are shown in Figure 5. Two crystalline peaks at 2θ = 11.59º and 28.9º were observed in CS film, these observations agreed with the results reported by others [34-35]. After the addition of 4HC in the chitosan film, the intensity of diffraction peak 11.59º of CS is diminished with increasing content of 4HC. It illustrates that the crystalline nature of CS decreases for the C4HC films due to the existence of 4HC. Also, the absence of any new diffraction peaks for C4HC films reveals a complete dissociation of 4HC on the CS matrix.

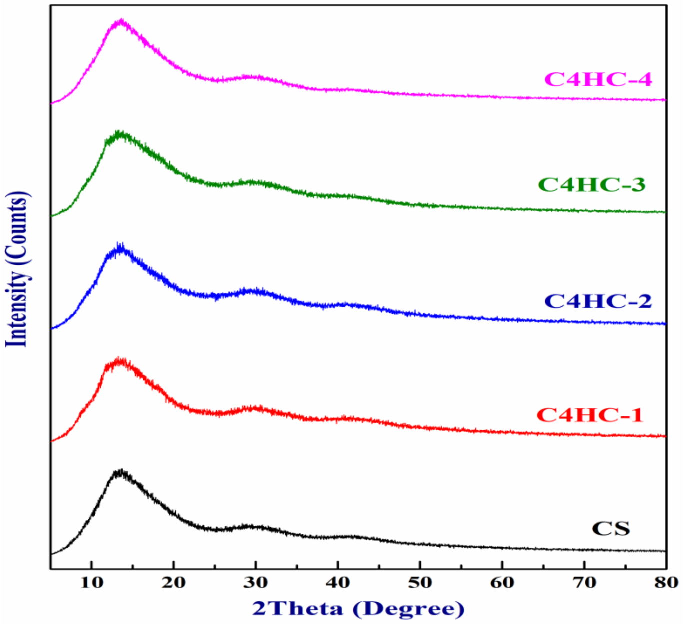

Figure 5: X-ray diffraction patterns of CS and C4HC films.

Water Contact Angle Measurement

Water contact angle measurements were carried out to understand the hydrophilic nature of the films. Below Figure 6 shows the images of water drops on the surface of films with contact angles. It is well known that, when contact angle values greater than 90° are obtained, there are hydrophilic interactions between the solid surface and the dissolution medium. The water contact angles 83.9O, 88.7O, 86.3O, 79.7O and 74.3O were found for CS, C4HC-1, C4HC-2, C4HC-3, and C4HC-4 respectively. It implies that all the films were hydrophilic. After addition of 4HC into the CS the decreased hydrophilicity was observed for C4HC-1 film, further increase in the concentration of 4HC the contact angles were decreased gradually. These results were correlated with the AFM results that the contact angles were decreased with increasing the roughness of the films. This behavior is likely to be associated with the hydrophobic backbone of the polymer chains. This effect may be due to the fact that the interaction between 4HC and CS.

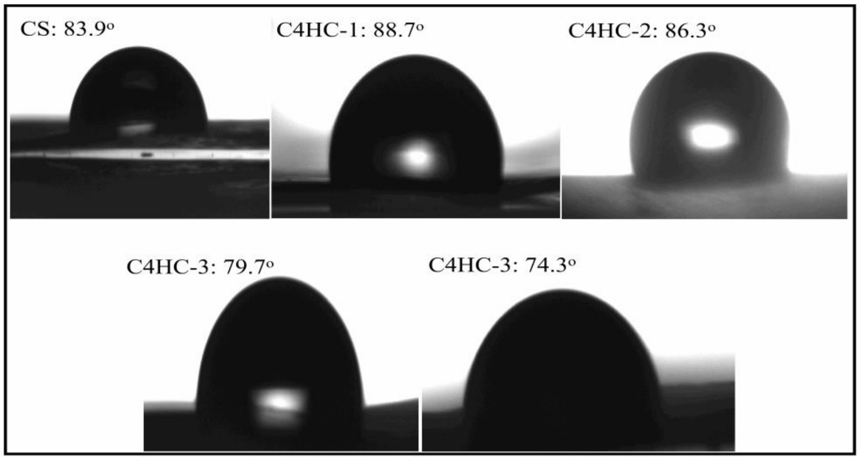

Figure 6: Water drop images on CS and C4HC films.

CONCLUSION

In the present study CS and C4HC films were successfully prepared and characterized. The results of AFM study showed the uniform surface morphology and surface roughness showed increasing order as the weight of the 4HC is increased in the CS film. The compatibility among components indicates the appreciable intermolecular interaction among the CS and 4HC which is confirmed by the FTIR study. The thermal properties of the CS films slightly increased with incorporation of different concentration of 4HC. In addition presence of single glass transition temperature in all C4HC composite films suggests the miscibility among the components. The XRD results illustrates that the crystalline nature of CS decreases for the C4HC films due to the existence of 4HC. The results of water contact angle study showed that contact angle decreased with addition of 4HC onto the CS indicating the films were hydrophilic in nature and affinity towards water increased in C4HC films when compared to pure CS. Further study can be extended to the application level by performing the different application oriented instrumental characterizations. It can be expected that, the best properties of C4HC composite films were recorded in the study may play a vital role in food packaging and biomedical applications.

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