



Fabrication of antibacterial blend film from poly (vinyl alcohol) and quaternized chitosan for packaging



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ABSTRACT

Blend films from poly (vinyl alcohol) (PVA) containing *N*-(2-hydroxy) propyl-3-trimethyl ammonium chloride chitosan (HTCC) were prepared via a simple mixing and casting method. The films were characterized by Fourier transform infrared spectroscopy (FTIR), X-ray diffraction measurements (XRD), scanning electron microscopy and ultraviolet-visible measurements (UV-vis). The effects of HTCC amount on mechanical properties, oxygen permeability, water vapor permeation, and antibacterial properties against Gram-negative (*Escherichia coli*) and Gram-positive (*Staphylococcus aureus*) of the films were investigated. FTIR and XRD analysis show that HTCC and PVA in the blend films interacted by hydrogen bonding. SEM and UV-vis analysis reveal the good compatibility between HTCC and PVA. Compared with pure PVA film, the blend films had greater elongation at break, lower water permeability, and higher antibacterial activity. The HTCC addition decreased the tensile strength and the light transmittance. The results suggest that HTCC/PVA blend films have a potential as packaging materials.

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1. Introduction

Many natural biopolymers such as cellulose, chitosan, starch, soybean protein isolate, alginate, pectin and others have been extensively developed due to the growth of environmental concerns on non-biodegradable materials in the field of food packaging [1–5]. Compared with conventional plastic materials, the natural polymers have several advantages including non-toxicity, biodegradability, wide availability, and biocompatibility [6]. However, some of their properties must be improved to be competitive with petroleum-based products, especially their poor mechanical properties, low thermostability, and high water-sensitive properties. Over the years, researchers have adopted many chemical and physical methods to modify biopolymer materials for promoting the development of films, such as crosslinking, graft copolymerization, chemical modification or blending [7,8]. Among them, polymer blending is simple and effective method to obtain new material with desired properties [9]. poly (vinyl alcohol) (PVA), a non-toxic and biodegradable synthetic polymer, has excellent miscibility and film forming properties [10]. It has been widely utilized for the preparation of blends with other renewable biopolymers [11].

It is highly important for packaging materials with proper mechanical properties, water and oxygen barrier properties, biodegradation, and higher thermostability, antibacterial activity [12]. Chitosan, a cationic polysaccharide that is an abundant natural biopolymer, is composed of β (1 → 4) glucosidic bonds [13]. Chitosan and its derivatives have the potential to be used as packaging materials due to its antibacterial activity, non-toxicity and good film-forming property [14]. However, chitosan of high molecular weight is soluble only in acid media. Moreover, it starts to lose its antibacterial activity at pH value higher than 6.5. In contrast, its quaternary ammonium derivatives have higher activity against a variety of bacteria and water-soluble over a wide range of pH values than chitosan itself [15,16]. Among them, *N*-(2-hydroxy) propyl-3-trimethyl ammonium chloride chitosan (HTCC) is a promising candidate for antibacterial packaging because of the low cost, ease of processing and high antibacterial activity. Nevertheless, HTCC film materials exhibited disadvantages of high sensitivity to water and poor mechanical properties, which makes it unsatisfactory for application in food packaging. A study has been carried out to prepare crosslinked blend film of HTCC and PVA by crosslinking with glutaraldehyde for potential applications in wound-dressing [17]. As known that glutaraldehyde is toxic and glutaraldehyde-crosslinked film is not suitable for food packaging. Furthermore, there is no specific report on the blend film HTCC and PVA by simple mixing and casting method. The effects of HTCC addition on their mechanical properties, light transmittance, oxygen permeability and water vapor permeation

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haven't been researched in detail aiming to use in food packaging fields.

The aim of the present work was to study the structural, physical and antibacterial properties of HTCC/PVA blend films. The films were characterized by Fourier transform infrared (FTIR) spectroscopy, X-ray diffraction (XRD), and Scanning electron microscopy. Mechanical properties, oxygen permeability (OP), water vapor permeability (WVP), as well as light transmission were also investigated. The antibacterial activity against Gram-negative (*Escherichia Coli*) and Gram-positive (*Staphylococcus aureus*) bacteria was evaluated.

2. Experimental

2.1. Materials

HTCC (>91% degree of substitution) with an average molecular weight of 50,000 was purchased from Tianhua Bio-Assistants Co., Ltd., (Shandong, China). Poly(vinyl alcohol) (PVA; average Mw = 84000–89000 g/mol; degree of polymerization (DP) = 1700–1800; 88% alcoholysis) was purchased from Sinopec Shanghai Petrochemical Co., Ltd., (Shanghai, China). All the commercial products were used without further purification. Other chemicals were of analytical grade and used as received. *E. Coli* and *S. aureus* were obtained from Qingdao Hope Bio-Technology Co., Ltd., (Qingdao, China). All other biochemical reagents were purchased from AoBoXing Bio-tech Co., Ltd., (Beijing, China).

2.2. Preparation of HTCC/PVA films

A HTCC solution (5.0 wt%) was prepared by dissolving HTCC in distilled water at room temperature with mechanical stirring at 500 rpm for 30 min. PVA was dissolved in distilled water under constant stirring at 80 °C for 2 h to achieve a clear solution of 5.0 wt%. HTCC and PVA solutions were mixed together with different mass ratios, and the resulting solution was stirred vigorously for 30 min. After the solution was degassed in a vacuum oven, it was transferred into a Teflon pane (30 cm × 30 cm × 3.5 cm). The film-forming solution in the Teflon pane was dried at 50 °C to obtain films with uniform thickness. The HTCC/PVA films were denoted as HP-5, HP-10, HP-20 and HP-30 in a weight proportion of 5:95; 10:90; 20:80 and 30:70, respectively. The control of pure PVA film was obtained by the process as above.

2.3. Characterization

FTIR spectra were recorded on a Nicolette 6700 spectrometer (Thermo Fisher Scientific Co., Ltd., MA, USA) with attenuated total reflection mode at a resolution of 4 cm⁻¹. XRD patterns were carried out by using a D/max-2200 diffractometer (Rigaku, Japan) at a voltage of 40 kV and a current of 30 mA using Cu-K α radiation with a scanning rate of 5/min. The scanning scope of 2 θ was ranged from 5 to 40 at ambient temperature. The morphology of the films was examined under a Quanta 200 scanning electron microscope (Philips-FEI Co., AMS, The Netherlands) with an accelerating voltage of 5 kV. Samples were frozen in liquid nitrogen and snapped immediately, and coated with a gold layer prior to observation and then observed at different magnifications. Films thicknesses were measured with an ID-C112XBS micrometer (Mitutoyo Corp., Tokyo, Japan) at ten random positions on the film. Light transmission through the films (4 × 3 cm²) was recorded in the range of 200–800 nm by using an ultraviolet-visible (UV-vis) spectrophotometer (UV-2600, Shimadzu, Kyoto, Japan). Tensile tests were measured by using an auto tensile tester (XLW-PC,

PARAM, Jinan, China) equipped with a 500 N load cell. Measurements were performed at a strain rate of 300 mm m⁻¹ in. The OP through films were measured by using a Perme OX2/230 (Labthink, Jinan, China) according to standard method ASTM D3985-05 (2002) at 23 °C and 0% relative humidity (RH). The WVP through films were determined at 23 °C and 60% RH by using a Mocon Permatran-W 3/61 (MOCON, MN, USA) according to GB/T 26253 (GB/T, 2010).

2.4. Antimicrobial activity

The disk inhibition zone assay was used to evaluate the antimicrobial activities of the films against the tested bacteria according to the previous method [18]. Film samples were aseptically cut into disc 0.8 cm in diameter and then placed on NB plates, which had been previously smeared with 200 μ l of NA culture containing approximately 10⁸ CFU/ml of bacteria. The plates were incubated at 37 °C for 12 h before examination for inhibition zones around test film disc. The diameter of the inhibition zone was measured using a caliper. Three replicate tests were carried out under the same conditions for each sample.

2.5. Statistical analysis

Multiple samples were tested, and the results were reported as the mean \pm standard deviation. The data were analyzed using a one-way analysis of variant (ANOVA) by means of the SPSS software and differences among mean values were processed by Duncan's multiple-range tests. Significance was defined at P < 0.05.

3. Results and discussion

3.1. FTIR analysis

FTIR spectra of HTCC, PVA and HTCC/PVA blend films are shown in Fig. 1. In the spectrum of HTCC, the bands at around 3366 cm⁻¹ and 3218 cm⁻¹ are due to —OH and —NH stretching vibration. Band at 3037 cm⁻¹ is assigned to the N⁺—H stretching vibration. The weak band at wavenumber ranging from 2989 to 2865 cm⁻¹ corresponds to the C—H stretching vibration of the —CH₂— and —CH— groups. The absorption bands at wavenumber at 1646 and 1357 cm⁻¹ correspond to the referring to the C=O and C—O stretching of amide group, respectively [19]. A strong peak at 1480 cm⁻¹ is attributed to C—H bending of the methyl substituents of quaternary ammonium groups [20]. The peaks matched with saccharide backbone are easily viewed at 1166 cm⁻¹ (anti-symmetric stretching of the C—O—C), and around 1022 cm⁻¹ (skeletal vibrations involving the C—O stretching), and 965 cm⁻¹, 924 cm⁻¹, 884 cm⁻¹ (skeletal vibrations involving the C—C stretching) [21,22]. Bands for PVA appear at around 3283 cm⁻¹ (—OH stretching vibration), and at around 2937 and 1325 cm⁻¹ (C—H stretching and wagging vibrations) [23]. The C=O and C—O stretching vibrations of the unhydrolyzed acetate group in PVA are observed at approximately 1732 and 1086 cm⁻¹, respectively. The bands are easily viewed at 1422 cm⁻¹ (the second O—H bending stretching), 1023 cm⁻¹ (C—C—C stretching vibration), 945 and 834 cm⁻¹ (skeletal vibration) [24]. As for HP blend films, characteristic bands are similar to those of PVA. The band at 1646 cm⁻¹ could be attributed to HTCC in the HP blend films. Moreover, the peak intensity increased with the increase of HTCC content. While, the —OH stretching vibration bands at around 3283 cm⁻¹ broadened, suggesting hydrogen bonding between HTCC and PVA molecules in the blend films.

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