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ORIGINAL RESEARCH PAPER

The preparation of chitosan-Ag nanocomposite for food packaging

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ABSTRACT

The present study was carried out to investigate the preparation of chitosan-Ag nanocomposite film for food packaging. Since chitosan is a suitable alternative to produce packaging films due to favorable factors such as biodegradability and abundance in the world, therefore in this study, it was prepared chitosan-Ag nanocomposite with antibacterial properties for food packaging by combining chitosan and Ag nanoparticles. The produced nanocomposite was characterized by XRD and FESEM. Antibacterial activity of the produced film was studied at different concentrations of silver nitrate against *Escherichia coli* and *Staphylococcus aureus*. The results showed high antibacterial activity in chitosan-Ag chitosan. It was also found that with an increase in the concentration of Ag nanoparticles in the nanocomposite to 0.03, the ant antibacterial effects increased and then remained constant.

Keywords: *Antimicrobial, Chitosan-Ag nanocomposite, Food packaging*

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INTRODUCTION

Today, the production of antibacterial and antifungal materials is a great challenge. These materials are used in important industries such as food industry, pharmaceutical industry and etc. Among antibacterial and antifungal products, chitosan is more important than others for many applications. Sadeghnejad et al. [1] reported that antibacterial the properties of silver nanoparticles increase with decreasing size of particle. Ghorbani et al. [2] have studied the antibacterial activity of polypropylene-Silver on E. coli and S. aureus. The polypropylene film surface was modified using the corona discharge method. Surface pre-treating with corona discharge increases the resin adhesion on the surface of the film for nanoparticle coating. In other work, polyethylene film was coated with copper nanoparticles and its antibacterial properties were studied. In addition, this investigate was carried out to determine the optimum copper concentration in coating solution for nanocomposite film preparation to increase antibacterial effects [3]. Kim et al. studied antibacterial effect of silver nanoparticles against E. coli, yeast and S. aureus [4]. The morphology and mechanical properties of polyamide / silver composite were studied by Ojeda et al. [5]. In another work, antibacterial activity of silver nanoparticles was studied by Phu et al. using various stabilizers. They found that silver-alginate nanoparticles have the highest antibacterial activity against the E. coli comparing to the other agents of AgNPs/PVP, AgNPs/PVA, AgNPs/sericin [6]. In 2015, the mechanical properties of epoxy nanocomposites were investigated. Moosa et al. used multi-wall carbon nanotubes (MWCNTs) as a filler for polymer matrix [7]. In another study, anti-corrosion property was studied by Fayomi et al. They added Zn and Fe_2O_3 nanoparticles to epoxy [8]. In 2018, the antifungal activity of polyurethane/ CuO film against penicillium was investigated.

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Their study showed that the optimum conditions were 2% solution, 10,000W of power and 5 min of time in corona discharge method [9].

In this work, it was prepared chitosan-Ag nanocomposite with antibacterial properties for food packaging and characterized by XRD and FESEM. Then, it was used from disc diffusion method to study the antibacterial activity of nanocomposite. Finally, it was determined the optimal conditions for antibacterial activity.

MATERIALS AND METHODS

In this study, the suspension of chitosan (100 ml, 0.5% wt., Sigma-Aldrich) was prepared by dissolving chitosan in acetic acid solution 0.1% wt. (pH = 3.53, Merck Company) by stirring for 90 min. It was added the silver nitrate solution (Merck Company) in various concentrations 0.06 M (sample 1) and 0.03 M (sample 2) and 0.015 M (sample 3) to chitosan suspension. Then, sodium hydride solution (Merck Company) was added to suspensions for the synthesis of nanoparticles. After adding this reducing agent, the solution was stirred for one hour [12]. Finally, the chitosan-Ag nanocomposite suspensions were prepared, centrifuged, washed and dried for four hours. It was prepared a film with a thickness of about 0.9 mm. They were characterized by XRD (PW 1830 X-ray Diffraction, Philips) and scanning electron microscopy (FESEM, Hitachi S4160, Cold Field Emission) to investigate the structure of chitosan-silver nanocomposite films. The X-ray diffraction (XRD) is a suitable method that provides comprehensive information on the chemical composition and crystalline structure of natural and industrial materials. Antibacterial activity of the films was performed using disc diffusion method [3]. In this method, two bacteria: one gram-negative of E. coli and one gram-positive S. aureus were used for antibacterial study. In addition, two antibiotics (vancomycin and penicillin) were used to compare with prepared films. All experiments were performed at room temperature.

RESULTS AND DISCUSSIONS

Results of X-ray diffraction (XRD)

XRD was used to prove the presence of silver nanoparticles in nanocomposites. The patterns were shown for three samples in Figs. 1 to 3. Fig. 1 shows the peaks recorded in the sample 1 in the values 21.0°, 38.5° and 77.8°. Also Fig. 2 and 3 shows the peaks recorded in the sample 2 and 3 in the values 21.9°, 39.0°, 78.6° and 21.0°, 38.9°, 78.8°, respectively.

In images 1 to 3, they were shown the presence of nanoparticles and the formation of chitosan-Ag nanocomposites with XRD pattern. XRD peaks confirmed the crystalline structure of the nanocomposite [2]. Fig. 4 shows the XRD pattern for silver. The average crystallite size was calculated based on the Debye–Scherrer equation [4]:

$$D = \frac{K\gamma}{\beta_{hkl} COS\ \theta} \quad (1)$$

Where β_{hkl} is the integral half width, K is a constant equal to 0.90, λ is the wavelength of the incident X-ray, D is the crystallite size, and θ is the Bragg angle. The size of the Ag nanoparticles

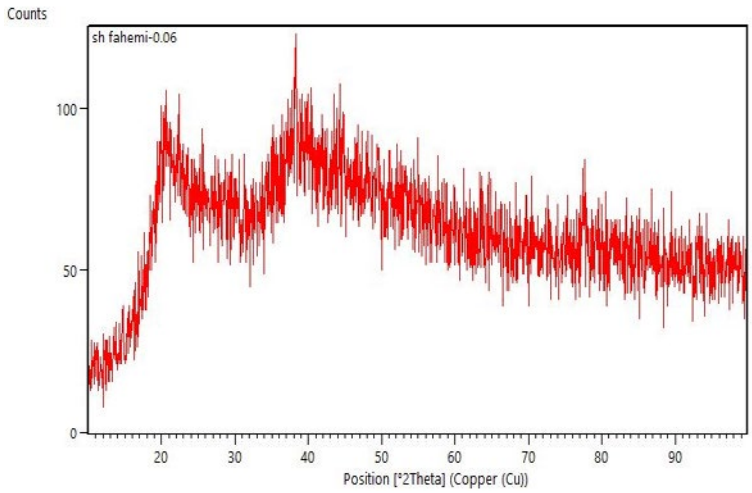

Fig. 1. XRD pattern of chitosan-Ag nanocomposite (sample 1)

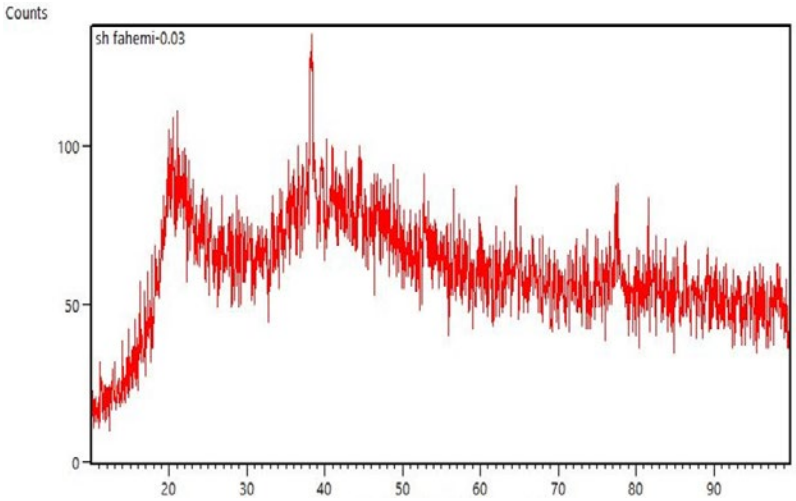

Fig. 2. XRD pattern of chitosan-Ag nanocomposite (sample 2)

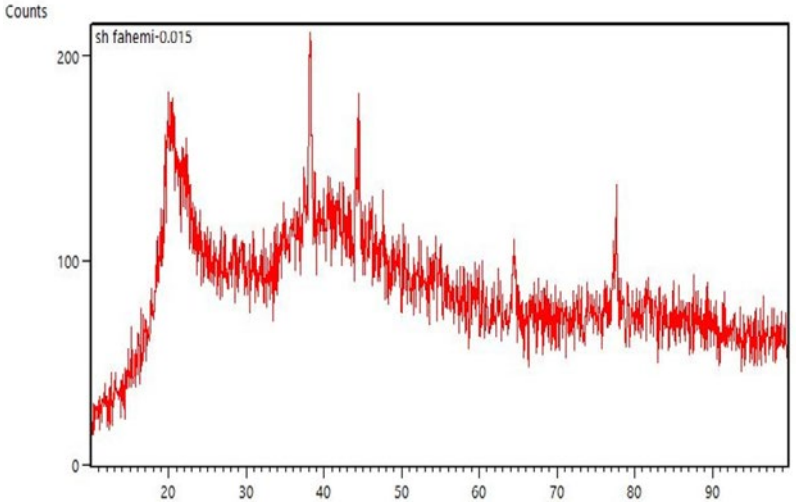

Fig. 3. XRD pattern of chitosan-Ag nanocomposite (sample 3)

Table 1. Comparison of nanoparticles size in samples

Concentrations	L1 (nm)	L2 (nm)	L3 (nm)	Mean size (nm)
0.06 M (sample1)	36.77	38.71	23.10	32.86
0.03 M (sample2)	26.76	21.79	30.82	26.46
0.015 M (sample3)	29.12	20.59	19.68	23.13

estimated from Debye-Scherrer formula was 23 nm.

The Results of field-emission scanning electron microscopy (FESEM)

Field-emission scanning electron microscopy (FESEM) was used to investigate the structure of the nanocomposite. As shown in Fig. 5, spherical Ag nanoparticles were well enclosed by chitosan. The obtained patterns showed that Ag nanoparticles were embedded suitably in a chitosan matrix. In Table 1, it is presented the nanoparticles size for samples. Table 1 indicates that the particles size increases with increasing samples concentration in prepared nanocomposite.

The antibacterial activity of chitosan-Ag nanocomposites

To study the antibacterial activity of chitosan-Ag nanocomposite, it was used from disc diffusion

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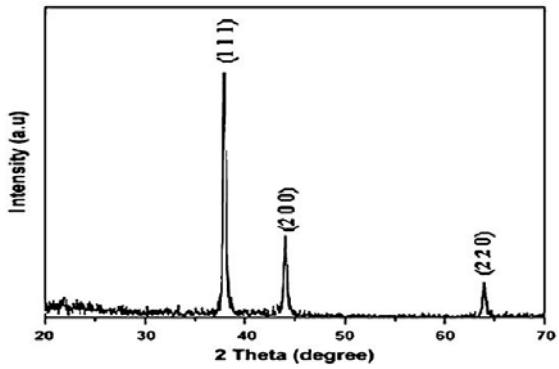

Fig. 4. XRD showing peak indices

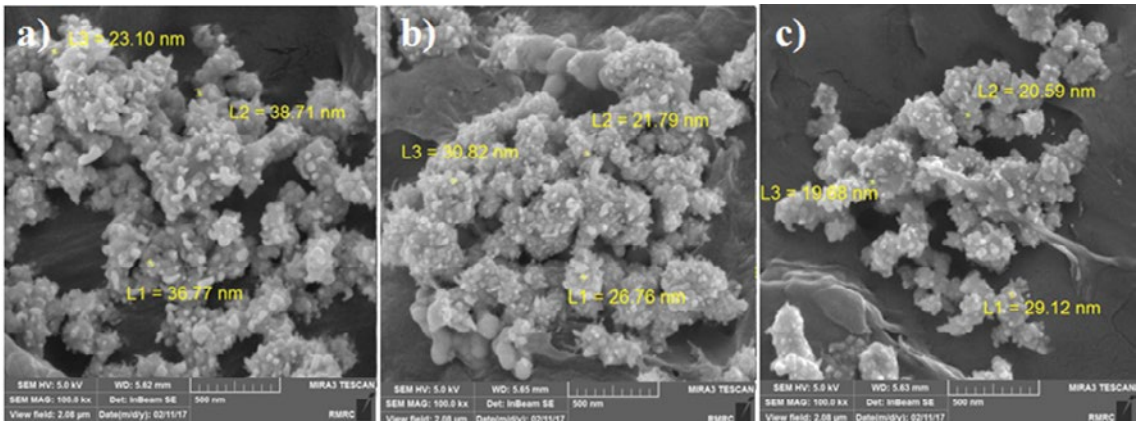

Fig. 5. SEM image of the chitosan-Ag nanocomposite, a) sample 1, b) sample 2, c) sample 3

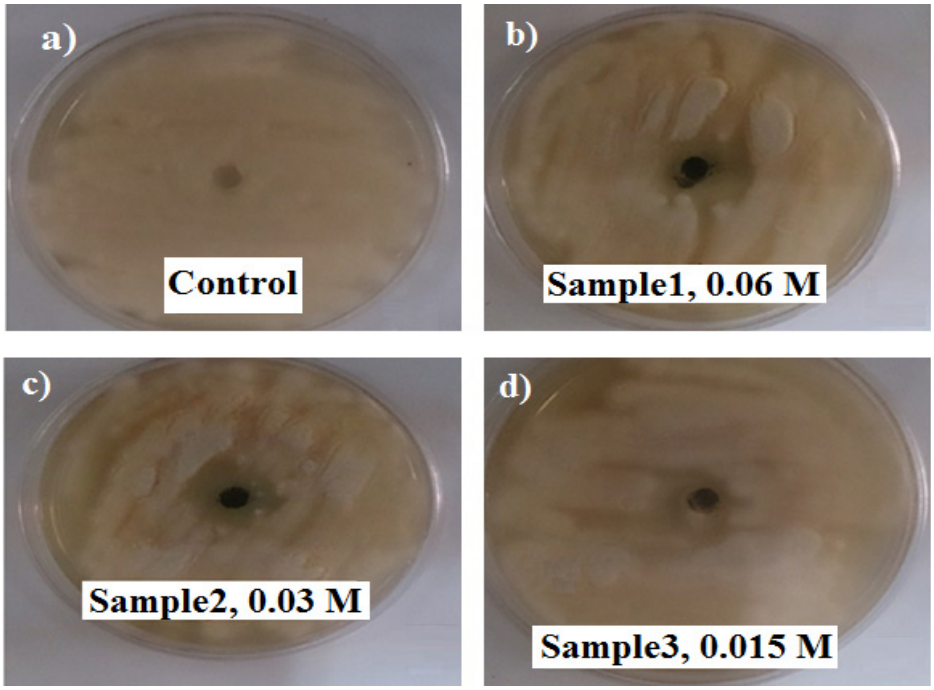

Fig. 6. Zone of inhibition: a) Control sample b) Chitosan-Ag nanocomposite, 0.06 M, c) Chitosan-Ag nanocomposite, 0.03 M, d) Chitosan-Ag nanocomposite, 0.015 M

Table 2. Antibacterial activity of chitosan-Ag nanocomposite on E. coli and S. aureus, zone of inhibition (mm)

Sample	Silver nitrate concentrate	zone of inhibition (mm)	
		E. coli	S. aureus
Control	-	0	0
Chitosan-Ag nanocomposite (sample 1)	0.06 M	7.8	6.4
Chitosan-Ag nanocomposite (sample 2)	0.03 M	7.4	6.1
Chitosan-Ag nanocomposite (sample 3)	0.015 M	4.9	4.3
Vancomycin	-	9.8	8.6
Penicillin	-	11.1	10.2

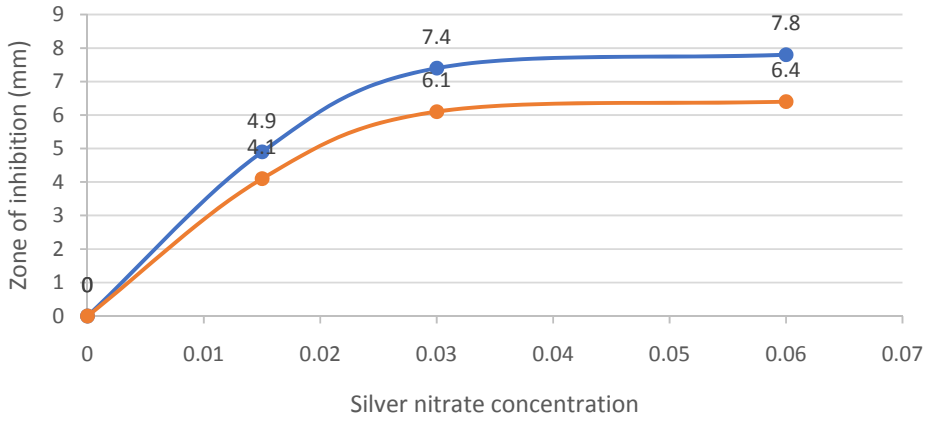

Fig. 7. The curve of inhibition zone of chitosan-Ag nanocomposite (Series 1: E. coli, Series 2: S. aureus)

method. The zone of inhibition was measured using a ruler. The results were presented in Fig. 5. It was observed that the zone of inhibition increased with increasing samples concentration in prepared nanocomposite. Table (2) shows the diameter of the inhibition zone. Considering the shape of the inhibition zone and their diameter, it was resulted that the antibacterial effect of the nanocomposite is related to the nanoparticles concentration.

It was revealed that the inhibition zone of bacteria growth increased by increasing the concentration of silver nitrate, but the zone of inhibition was the same at a concentration of 0.03 M and 0.06 M. In fact, the optimum concentration for the suitable antibacterial activity was concentration of 0.03 M. Fig. 7 shows the diameter curve of inhibition zone versus concentration of silver nitrate in the nanocomposite film.

CONCLUSIONS

In this study, it was prepared chitosan-Ag nanocomposite with antibacterial properties for food packaging by combining chitosan and Ag nanoparticles. It was characterized by XRD and FESEM. XRD peaks confirmed the crystalline structure of the nanocomposite. The average mean size of the Ag nanoparticles was 23 nm. FESEM showed spherical Ag nanoparticles were well enclosed by chitosan. In addition, it indicated that the particle size increased with increasing concentration of sample in prepared nanocomposite. Then, it was used from disc diffusion method to study the antibacterial activity of nanocomposite. The results showed that the optimum concentration for the suitable antibacterial activity was concentration of 0.03 M. Generally, it seems that this nanocomposite is suitable for food packaging with acceptable performance and great advantages.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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