Silver Nanoparticles Synthesis Using Chitosan Extract by a Simple Chemical Method and Application in Bntibacterial Activity

Wisam J. Aziz, Hanan Khudair Mutlak*, Randa Kamel Hussain

Department of Physics, College of Science, Mustansiriayah University, Baghdad, Iraq

*Corresponding author, E-mail: **hanankhudair80@gmail.com**

Doi:10.29072/basjs.202126

Abstract

Ag nanoparticles (Ag NPs) were prepared via simple chemical method at 200 ^{o}C for 2 hours using chitosan extract and sliver (iii) nitrate (Ag_2NO_3) salt. The chitosan extract acts as a reducing, stabling and anti-caking agent to transfer Ag_2NO_3 salt to Ag NPs in short period of time. Some physical properties of prepared Ag NPs are investigated using X-ray Diffraction (XRD), Field Emission Scanning Electron Microscopy (FE-SEM), optical absorption which used to characterize the structural, morphology, and optical properties of prepared samples. The crystallite size of Ag NPs that prepared using chitosan extract was ranged from (20.8 to 64.4) nm with a cube shape . UV-Vis absorption spectrum of prepared Ag NPs showed that the energy gap is 3.6 eV. FESEM images confirmed that particles size of prepared Ag NPs wan ranged from 52 nm to 91 nm with spherical shape. Antibacterial activity of prepared Ag NPs was test against two type of bacteria, Gram-positive (Staphylococcus) and Gram-negative (Escherichia coils).

Article inf.

Received:
4/5/2021
Accepted
7/8/2021
Published
31/8/2021

keywords:

Ag NPs, chitosan extract, antibacterial, S-arureus, E. Coli

1. Introduction

Physical and chemical properties of materials can drastically improve or change when their particles size is reduce to the nanoscale scale [1-3]. Chitosan is a derivative of chitin which is the second most abundant naturally occurring polymer after cellulose. It has aroused a lot of interest since it may degrade in the body to form the building blocks of the body's tissues [4]. This is also crucial for the development of materials that interact positively with tissue, allowing for the regeneration of lost or wounded tissue. Other naturally occurring polymers have been used to depict a structural cellular environment and have been exploited for their desired characteristics. Chitosan is unique in that it has qualities that aren't present in petrochemicals, making it desirable. The fact that chitosan can be employed directly for biological purposes is due to these intrinsic qualities [5]. In recent years, NPs, especially silver nanoparticles (AgNPs), have increasing interest due to they exhibit extraordinary antibacterial activity [6, 7]. Physical, chemical, and biological processes can all be used to prepare sliver nanoparticles. However, preparation of nanoparticles from chitosan has been paid great attention because of their biocompatibility, biodegradability and hydrophilic properties, which endow them opportunities for different applications [8]. In the present work, Ag NPs are prepared using chitosan and their antibacterial activity against two types of bacteria is investigated.

2. Experimental part

2.1 The synthesized of chitosan extract.

silver nitrate (Ag_2NO_3)/copany certified ISO9001-2000 and chitosan are used in this study to prepare Ag NPs. A 10 g of chitosan were dissolved in 50 mL of distilled water and stirred for two hours at 70 °C using hot palte magnetic stirrer. The mixture was then left to cool to room temperature before being filtered with Whitman's filter paper befor It packing in glass tube for future use. Figure 1 depicts the chitosan extract after it has been produced.

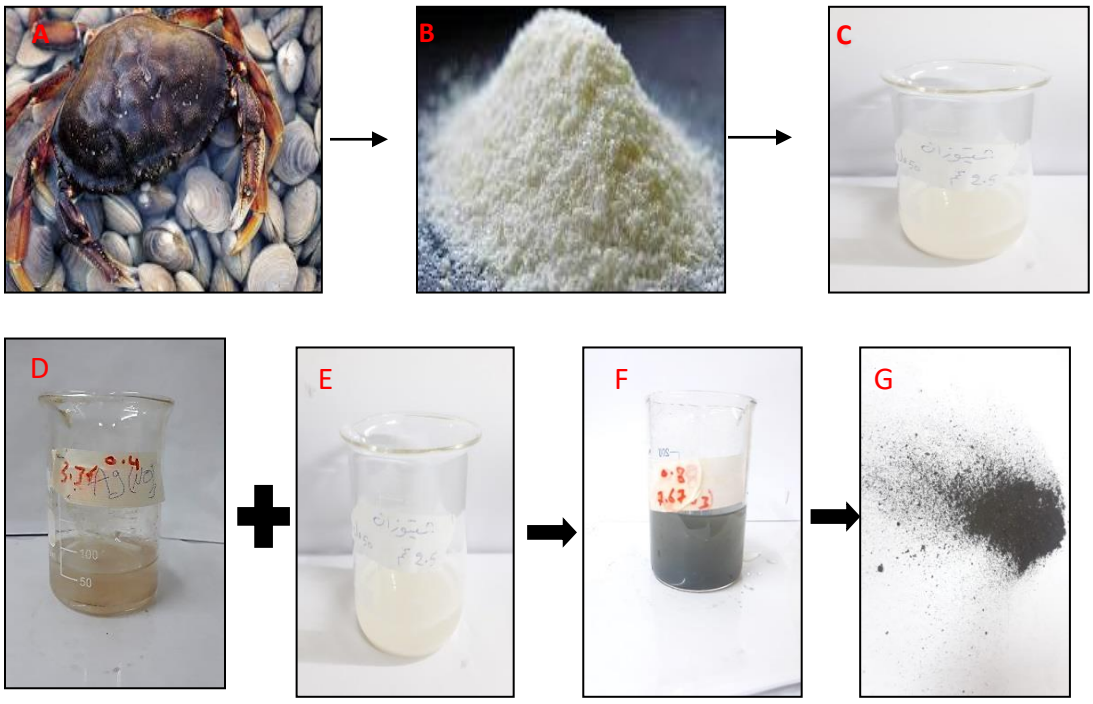

Figure 1: Steps of preparation Ag NPs using chitosan extract, (A) cambrian, (B) chitosan powder, (C) chitosan extract, (D) Ag2(NO3) salt, (E) chitasion extract, (F) Ag NPs, and (G) Ag NPs powder.

2.2 Preparation of Ag NPs using chitosan extract

A 50 mL (1 M) of silver nitrate (AgNO3) with concentration of 100mg/ml was combined with 50 mL of chitosan extract and kept at 80 °C for 30 minutes on the hot plate magnetic stirrer. The color of the solution was changed from white to dark gray indicating to form Ag NPs. Then, a 25 mL of prepared Ag NPs solution was baked for two hours at 200°C in a ceramic vessel to obtain Ag nanopowder in glass tube and kept for future diagnosis. Figure 1 demonstrates how Ag NPs were made from chitosan extract.

3. Results and discussion

3.1 Structural Properties

Figure 2 shows XRD pattern of Ag NPs that prepared using chitosan extract by a chemical method at 200 ^{o}C for two hours.The diffraction peaks that showed in the XRD pattern are corresponding to (012), (111), (200), (112), (220), and (311) planes of body center cubic (BCC) of Ag NPs. The crystal size was determined by Scherre requation [9]:

$$D\ (nm) = \frac{0.9*\lambda}{\beta\ cos\ (\theta)} \quad (1)$$

Where β is full width at half maximum , D is crystal size, K constant(0.94), and θ is diffraction angle The analyzed XRD results are listed in Table 1.

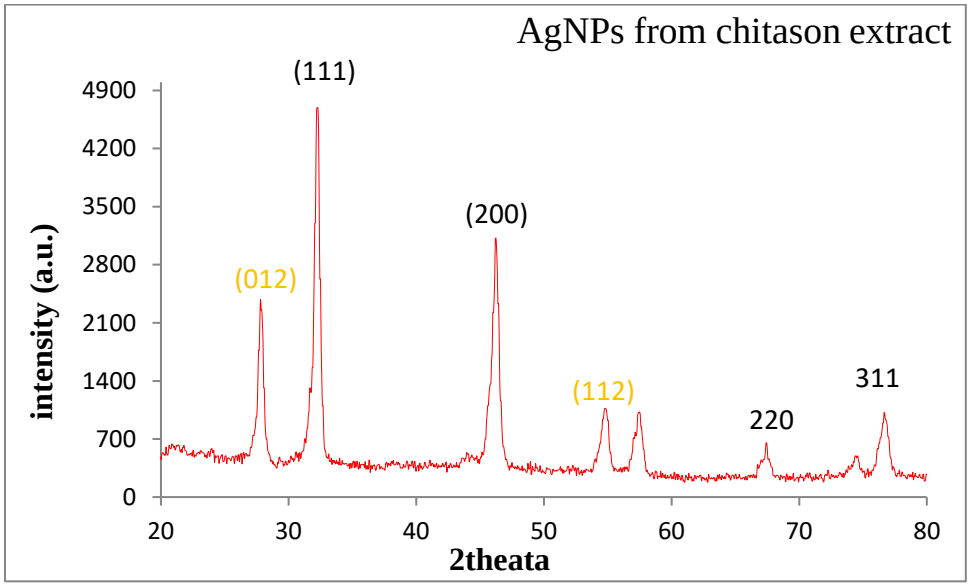

Figure 2 : XRD pattern of prepared Ag NPs.

Table 1: XRD analysis for prepared Ag NPs.

β(deg.)	2θ (deg.)	(hkl)	Crystallite size D (nm)
0.39	27.8	(012)	20.8
0.39	32.2	(111)	44.6
0.24	46.2	(200)	34.9
0.34	54.8	(112)	25.9
0.29	57.4	(220)	30.5
0.14	67.4	(311)	64.4

3.3 Surface Morphology

The surface morphology of prepared Ag NPs was investigated through FE-SEM images. Figure 3A & 3B shows the particles size of Ag NPs that prepared at 200 ^{o}C for 2 hours was ranged from 52nm to 91nm with cubic shape [10-11].

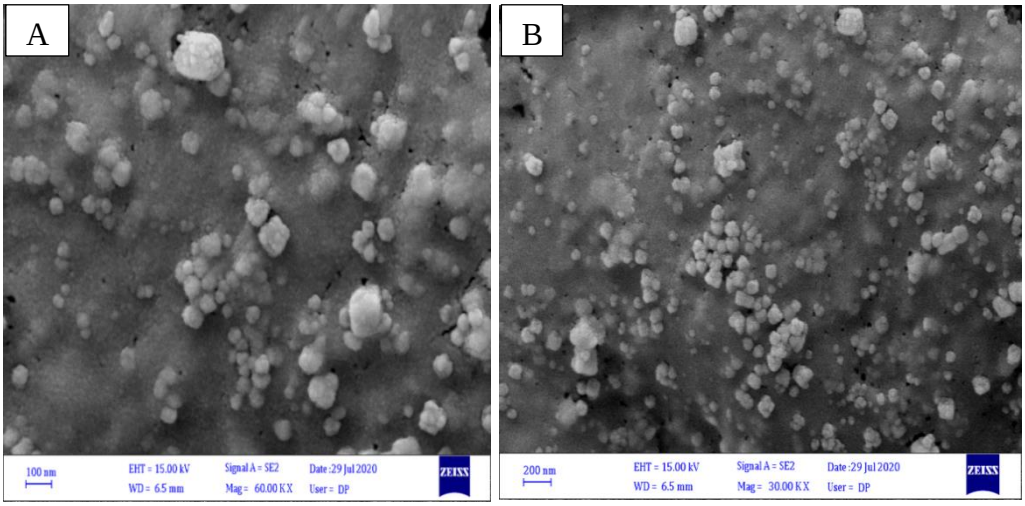

Figure 3: FE-SEM images ofAg NPs with magnification of, (A) 100 nm, and (B) 200 nm.

3.4 Optical properties

Optical properties of prepared Ag NPs was studied by optical transmission of light in the wavelength range of (200-900) nm using double-beam UV-VisS spectrophotometer type UV-1800, Shimadzu. Figure 4 shows the spectrum shows that the transmission of Ag NPs that synthesized from chitosan extract by a chemical method at 200 ^{o}C for two hours.. The value of the energy band gap of Ag NPs using chitosan extract is 3.8 eV as shown in Fig.4.

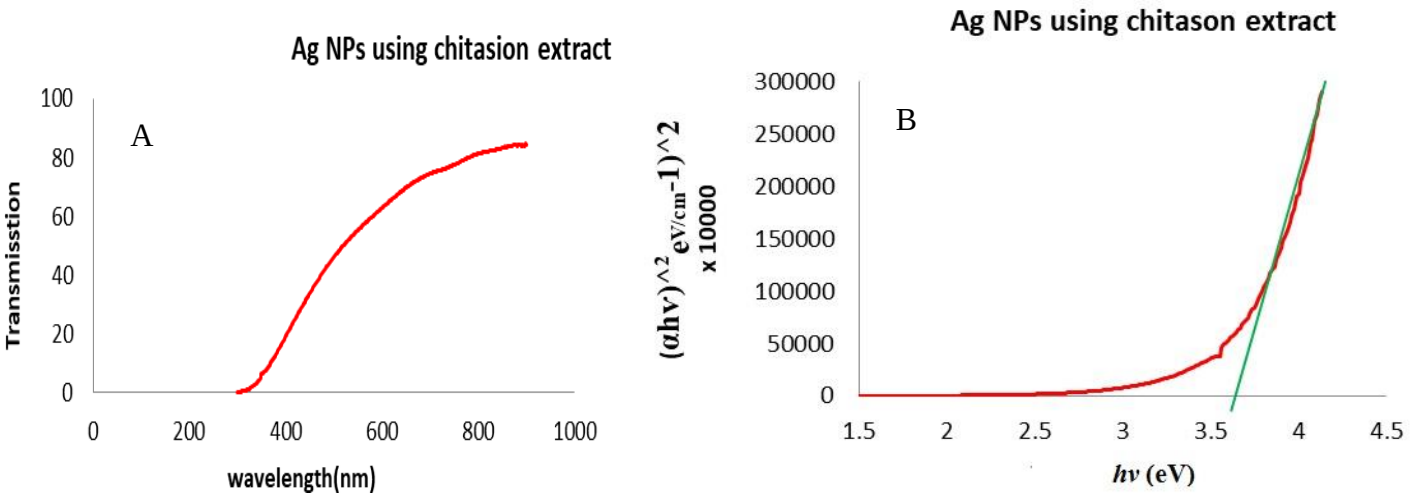

Figure 4: (A) Transmission of Ag NPs prepared from chitosan extract, (B) the plot of $(\alpha h\upsilon)^2$ versus photon energy of Ag NPs from chitosan extract.

3.5 Antibacterial Activity of Ag NPs

The antibacterial activity of the prepared AgNPs was investigated using disc diffusion method by measuring the inhibition zone gainst two isolates bacteria Gram-positive(Staphylococcus) and Gram-negative (Escherichia coils) [13-17]. Figure 5 shows the results of the inhibition zone and found that the AgNPs appeared inhibition zone against S. aureus of 19 mm while the zone became 11 mm for E-coli. W.S Abas et al. found that the AgNPs exhibited good reduction percentage against Gram-positive and Gram-negative bacteria at direct and 2 h post-exposure. Further, they conclude that the reason of killing the bacteria is DNA damage that confirmed by Real-time PCR results [18-19].

Table 1: Diameter of inhibition zone of Ag NPs against S-arureus and E. coli bacteria

Material	Plant extract	Gram-positive (+)	Gram-negative (-)	Percentage of inhibition zone (%)		Control
		S. aureus	E. coli	S. aureus	E. coli	
Ag NPs	Ghitason	19 mm	11mm	18	18	0

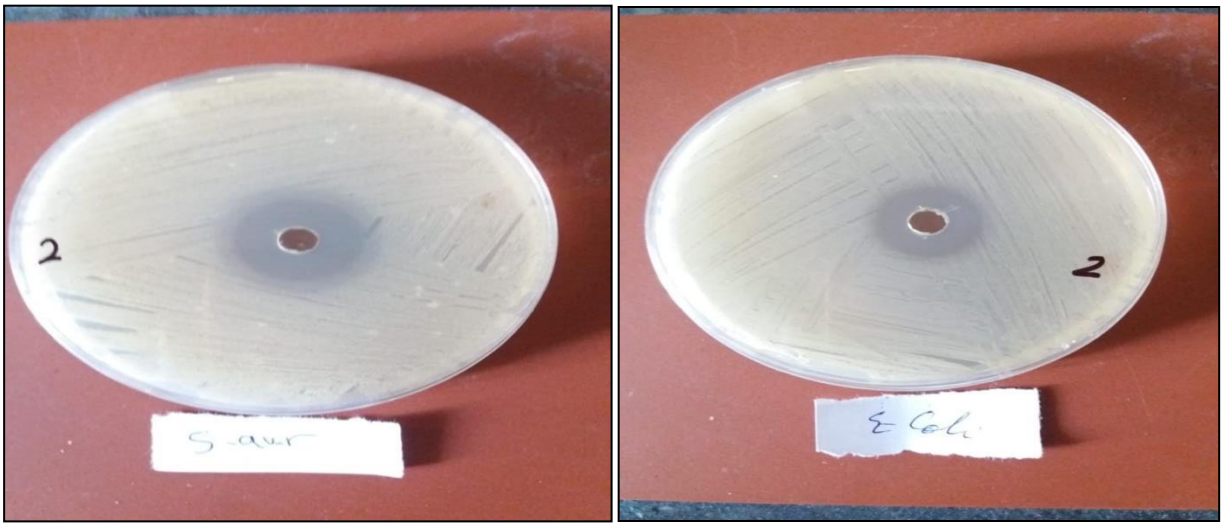

Figure 5: Photograph of the inhibition zone of Ag NPs against S- arureus and E. Coli bacteria

Conclusions

Ag nanoparticles (Ag NPs) were prepared via simple chemical method at 200 ^{o}C for 2 hours using chitosan extract with sliver (iii) nitrate (Ag_2NO_3) salt. The (*chitosan*) extract acts as a reducing, stabling and anti-caking agent to transform Ag_2NO_3 salt to Ag NPs in short period of time. It is observing that a changing in the color of the resulted solutions indicating to form Ag NPs. Different physical techniques such as X-ray Diffraction (XRD), Field Emission Scanning Electron Microscopy (FE-SEM), ultraviolet (UV-Vis) are used to characterize the structural, morphology, and optical properties of the prepared Ag NPs. The crystallite size of Ag NPs was ranged from 29 nm to 37 nm with a cube structure.. The energy gap of prepared Ag NPs is 3.6 eV while the particles size was ranged from 52 nm to 91 nm with cubic structure as confirmed by FESEM images The prepared AgNPs appeared antibacterial activity against S. aureus bacteria better than that appeared against E. coli bacteria.

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تخليق جزيئات الفضة النانوية باستخدام مستخلص الشيتوزان بطريقة كيميائية بسيطة وتطبيقها في النشاط المضاد للبكتيريا

وسام عزيز جعفر حنان خضير مطلك رندة كامل حسين

قسم الفيزياء-كلية العلوم-الجامعة المستنصرية-بغداد-العراق

تم تحضير التخليق الأخضر. لجسيمات الفضة النانوية (Ag NPs) المحضرة بالطريقة كيميائية بسيطة عند 200 درجة مئوية لمدة ساعتين باستخدام مستخلص الشيتوزان مع ملح نترات(Ag2NO3) الفضة. يعمل مستخلص (الشيتوزان) كعامل اختزال وتثبيت ومضاد للتكتل لتحويل ملح Ag2NO3 إلى Ag NPs في فترة زمنية قصيرة. يلاحظ أن التغير في لون الحلول الناتجة يشير إلى تكوين .Ag NPs من خلال تقنيات فيزيائية مختلفة مثل حيود الأشعة السينية (XRD) ، المجهر الإلكتروني لمسح الانبعاث الميداني (FE-SEM) ، الأشعة فوق البنفسجية (UV-Vis) التي تستخدم لتوصيف الخصائص الهيكلية والبصرية لـ .Ag NPs يتراوح الحجم البلوري لـ Ag NPs باستخدام مستخلص الشيتوزان من (29-37 نانومتر مع هيكل مكعب). أظهر طيف الأشعة المرئية وفوق البنفسجية أن فجوة الطاقة تبلغ 3.6 فولت من .Ag NPs تم تحديد مورفولوجيا السطح من Ag NPs باستخدام تحليل FESEM ، وحجم الجسيمات وشكل الحبوب من (52 إلى 91) نانومتر مع هيكل مكعب. جزيئات الفضة النانوية للتطبيق في النشاط المضاد للبكتيريا (موجبة الجرام) و (سالبة الجرام).