

Silver Nanoparticles Synthesized by Using *Matricaria chamomilla* Extract and Effect on Bacteria Isolated from Dairy Products

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Abstract

This study was conducted for the synthesis of small sized silver nanoparticles using the plant extract of *Matricaria chamomilla*. *Matricaria chamomilla* is rapid, non-toxic, eco-friendly, cost effective for the synthesis of nanoparticles. The results showed that the color formed by combining the extract with the 1mM silver nitrate solution changed to deep brown. The synthesis and characterization of silver nanoparticles was confirmed by ultraviolet-visible spectrophotometry and Fourier Transform Infrared Spectrometry techniques. UV-VIS was also used of showed peak absorption measurements in the wavelength 430 nm. FTIR spectrum indicates for detection of functional groups responsible for the formation on the surface of the nanoparticles. When evaluating effectiveness of silver nitrate with the plant extract on *Staphylococcus aureus*, *Escherichia coli* and *Brucella melitensis*. The result showed the synergistic action of AgNO₃ and the plant extract leading to enhance antibacterial activity.

Keywords: Silver nanoparticles, Antimicrobial agent, *Matricaria chamomilla*, UV-Vis, FTIR.

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تخليق جزيئات الفضة النانوية باستخدام مستخلص نبات البابونج *Matricaria chamomilla* وتأثيره على بكتيريا المعزولة من منتجات الألبان.

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الخلاصة

أجريت هذه الدراسة لتخليق جسيمات الفضة النانوية الصغيرة الحجم باستخدام مستخلص نبات البابونج *Matricaria chamomilla* هو عامل مختزل ومناسب للبيئة لإنتاج الجسيمات النانوية. وأظهرت النتائج أن تغير لون المزيج المتكون من المستخلص و 1 mM نترات الفضة إلى اللون البني الداكن. ووصفت الجسيمات الفضة النانوية المصنعة باستخدام جهاز طيف الأشعة فوق البنفسجية وتقنيات التحليل الطيفي الأشعة تحت الحمراء FTIR. أظهر الطيف المرئي للأشعة فوق البنفسجية للمحلول المائي الذي يحتوي على الجسيمات الفضة النانوية عند طول الموجي 430 نانومتر. يشير FTIR للكشف عن مجموعات وظيفية مسؤولة عن تشكيل على سطح الجسيمات النانوية. عند تقييم فعالية نترات الفضة مع المستخلص النباتي على المكورات العنقودية الذهبية، الإشريكية القولونية والبروسيللا ميليتنسيس. وأظهرت النتيجة العمل التآزري من $AgNO_3$ والمستخلص النبات مما يؤدي إلى تعزيز النشاط المضاد للبكتيريا.

الكلمات المفتاحية: جسيمات الفضة النانوية، عامل مضاد للميكروبات، *Matricaria chamomilla* ، UV-Vis ، FTIR .

Introduction

Nanoparticles can be simply defined as particles that have diameter lesser than 100 nm. As a result of their small size, NPs have large surface area to volume ratio, which results in higher reactivity and unique surface properties [1]. The antimicrobial efficacy of these nanoparticles is due to their nanoparticles size, providing them with unique chemical and physical properties as well as increasing the surface ratio to the size and high interactivity [2].

That silver and its compounds have wide-spectrum effects for their effectiveness antimicrobial of bacteria and fungus in addition to viruses since ancient times [3, 4] Compared with other minerals, being highly toxic to microorganisms and less toxic to mammalian [5].

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Microorganisms are used for the synthesis of nanoparticles in biological method [6], enzyme [7] and plant or plant extract by chemical and physical method [8, 9]. The biological methods are more efficient and less toxic than physical and chemical methods [10]. Because the use of chemicals causes the accumulation of some toxic substances on the surface of nanoparticles [11]. Plant extracts are cost effective, environment friendly and easily scaled up process for large scale synthesis [12, 13].

Plant extracts Used for nanoparticles synthesis advantageous over other biological processes by removing the detailed process of preserve cell cultures [14, 15]. There are many studies that use plant extracts for synthesis of silver nanoparticles, particularly *Ziziphora tenuior* [16, 17], *Olea europaea* [18], *Justicia adhatoda* [19] *Catharanthus roseus* [20], *Eucalyptus hybrida* [21], lemon [22], *Garlic* [23].

Several studies indicate that silver ions interact with amino acids containing a group SH [24] and play a role in bacterial disabling [25] Disconnection between the electron carrier within the respiratory chain, which inhibits the enzymes of the respiratory chain or interferes with membrane permeability of protons and phosphates[24]. This study aimed to synthesize and characterize silver nanoparticles and to investigate the activity of silver nanoparticles prepared by using *Matricaria chamomilla* extract (Figure 1) against different pathogenic microorganisms.



Figure 1: *Matricaria* plant

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Materials and Methods

Isolation and diagnosis:

Isolate and identification *Staphylococcus aureus*, *Escherichia coli* and *Brucella melitensis* from dairy products (milk, yogurt and cheese) were carried out according to the morphology and biochemical characteristics on and diagnosed according to the standard diagnostic methods [26,27].

Preparation of the aqueous extraction:

25 grams of dried *Matricaria chamomilla* dissolve in 100 ml of distilled water and leave to boil for ten minutes. The aqueous extract was filtered using Whatman Filter paper No.1 [28].

Synthesis of silver nanoparticles (agnps):

1mM Silver nitrate (AgNO_3) was prepared by weighing 17mg (AgNO_3) and dissolved in 100 ml of distilled water. It stored in brown bottle at 4°C away from light [29]. Add 1ml of aqueous plant extract to 9ml of 1mM silver nitrate solution. The brown color formation indicates that AgNPs were synthesized from the plant extracts [30].

Uv-visible spectrophotometer:

The Synthesis of silver nanoparticles was characterized by UV-Visible spectrophotometer (Thermo Scientific-NanoDrop1000 labtech international) from scientific reaches center/Salahaddin University device at wavelengths of 400-800 nm [29, 31].

FTIR Analysis:

The solution consisting of silver nitrate with the plant extract from which the silver nanoparticles was formed was dried at 75°C. The powders were characterized by using FTIR spectrometer (Shimadzu) in a wavelength between 4000-400 cm^{-1} [32].

Agar well-diffusion method:

To evaluate the antibacterial effects of silver nanoparticles was determined by standard agar well diffusion method [33, 34]. The bacteria cultures were prepared in 10 ml of nutrient broth followed by overnight shaking at 37°C and were swabbed onto the plate a Muller– Hinton agar

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by using sterilized cotton swabs [35]. Four wells by sterile boring cork (5 mm diameter). The 1mM AgNO_3 was added to the well No.1 and the Plant extract was added to the well No.2. A 100 microliter (include Mix Plant extract & AgNO_3) were added into well No. 3. The distal water was added to the well No.4 to make it as a negative control and incubated at 37°C for 24 hrs. The results were analyzed in terms of the zones of inhibition formed around each well [36].

Results and Discussion

The color changed rapidly to dark brown when adding plant extract solution to [1mM] AgNO_3 solution within 10 min shown in figure (2), the color change indicates the formation of silver nanoparticles during that is referred with the work done by [37, 38].

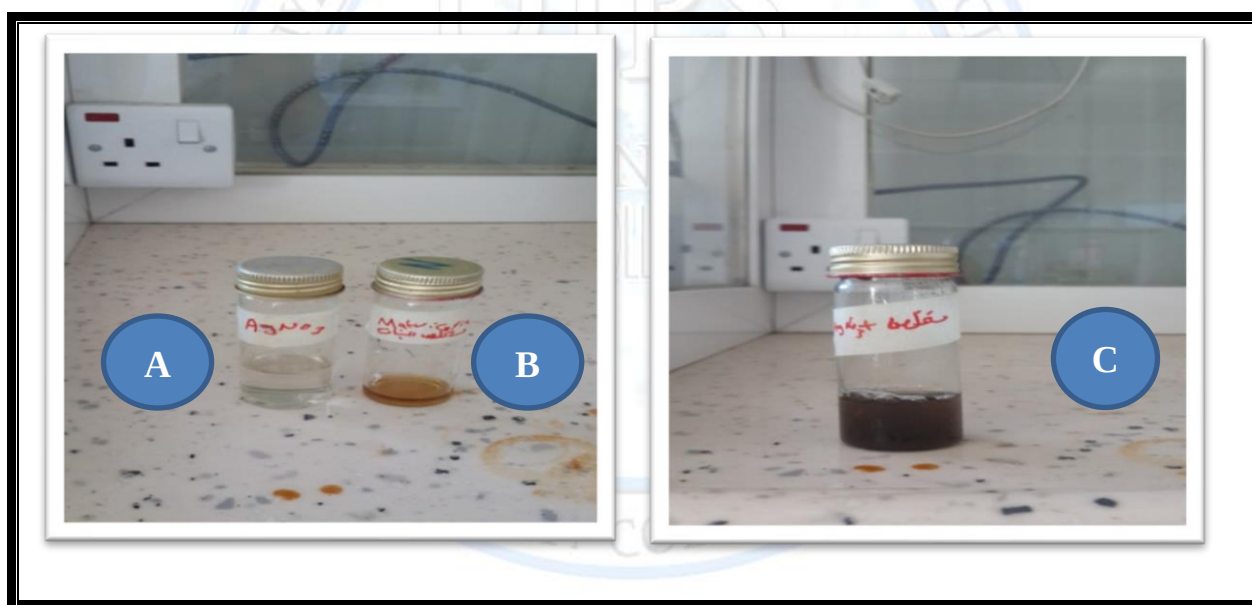


Figure 2: Photographs showing reaction solutions (A) 1mM AgNO_3 without *Matricaria chamomilla* extract (B) Aqueous solution for *Matricaria chamomilla* extract (C) color change from Yellow to dark brown after 10 min.

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The characterization of silver nanoparticles synthesized by aqueous solution AgNO_3 mixed with *Matricaria chamomilla* extracted may be confirm by UV-Vis spectroscopy shown in figure (3). The spectrum showed maximum absorption band at 430 nm indicating that the nanoparticles were scattered in the aqueous solution and preventing the AgNPs from agglomeration in UV-Vis Figure (3). This spectroscopy is consistent with the findings of other researches in which silver nanoparticles were synthesized using plant [39].

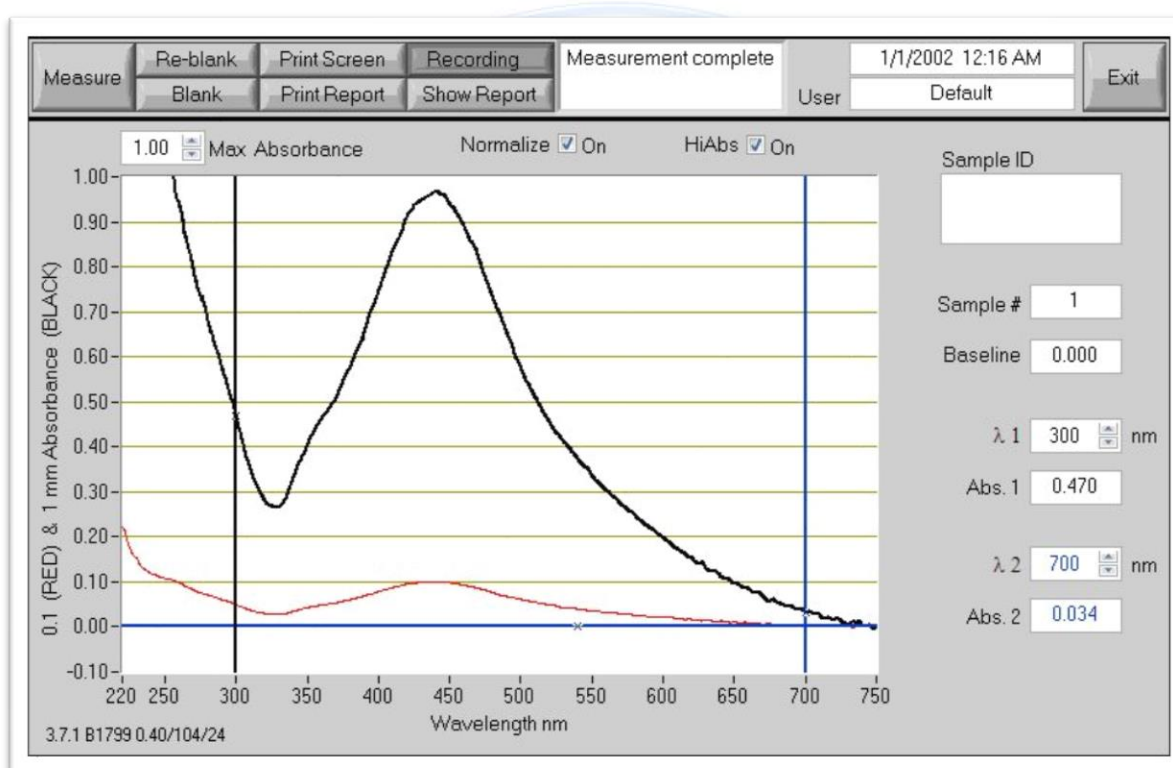


Figure 3: Absorption spectrum of the Silver Nanoparticles

The existence of broad band at 3363 cm^{-1} assigned to the absorption vibration of O–H groups, while O–H stretch vibration of carboxylic acid occurred at 2733 cm^{-1} , the band at 2395 cm^{-1} attributed to the C–H stretch in alkane part. The absorption vibration at 1398 cm^{-1} due to the stretching C–N of amide. The out of plane bending (coop) of C–H appears at 800 cm^{-1} [40]. Such interactions between metal nano particles and plant constituents have been reported in previous study [41]. the bone data are seen in the FTIR spectrum of silver Nanoparticles prepared from plant extract Figure (4). The proteins present me be acts as capping agent.

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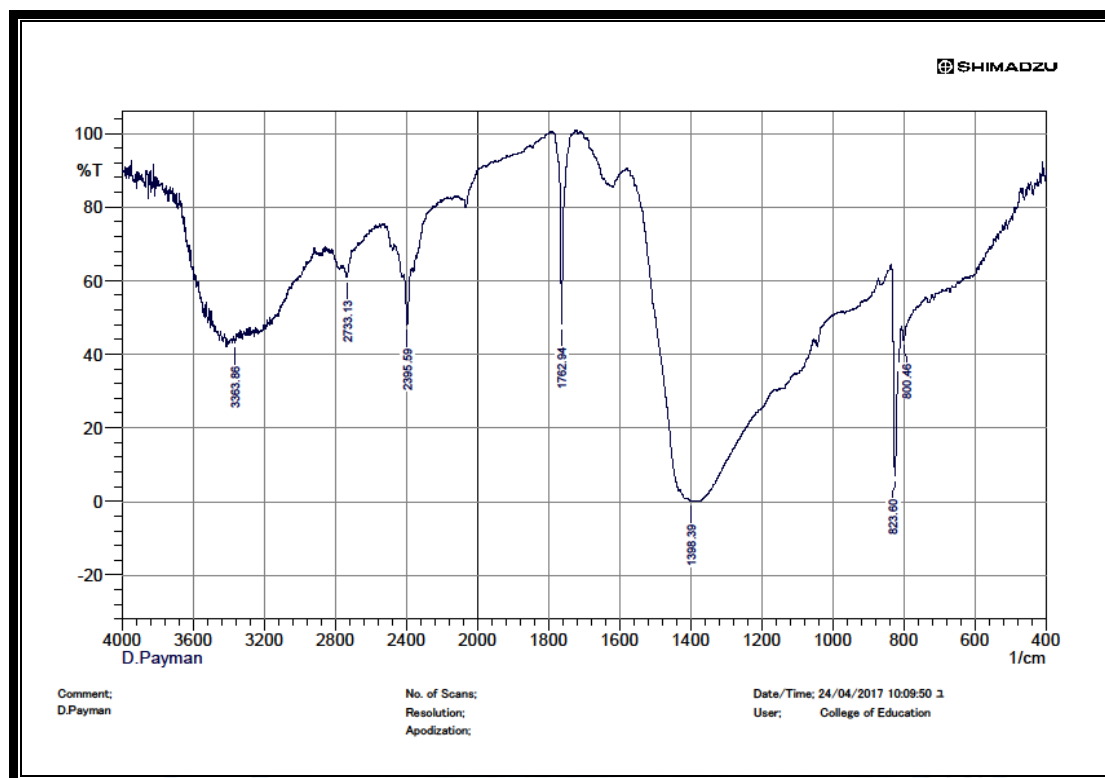


Figure 4: The FTIR spectrum of silver Nano powder

In this study also, we make a combination between *Matricaria chamomilla* extract with silver nanoparticles Table (1) and study their effect on microbial growth and the results showed that the effect will increase with assistant of silver nanoparticles.

Table 1: Effect of *Matricaria chamomilla* extracts with AgNO_3 on microbial activity (inhibition zone in mm).

No.	Zone of inhibition (in mm)		
	AgNO_3	Plant extract only	Mix Plant extract& AgNO_3
<i>Escherichia coli</i>	17	16	20
<i>Staphylococcus aureus</i>	18	12	23
<i>Brucella melitensis</i>	16	14	18

The mechanism in which the nanoparticles interaction with the bacterial cells is that the nanoparticles launches ions that react with group (-SH) for proteins transporting materials food that stand out from the bacterial cell membrane there by means of increase in permeability of

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the cell membrane, that enable plasma protein flowing out of the cell throw the interrupted cell membrane [42]. When silver molecules enter to the bacterial cell, the bacteria block to protect DNA from them, where nanoparticles act to attack the respiratory chain, leading to cell division and death [43]. When analyzing the protein of *E. coli* exposed to small amounts of nanoparticle there are changes in the membrane proteins, which can cause penetration of the membrane, a defect in the membranes and the loss of a large amount of potassium, in addition to low levels of ATP [4].

Conclusion

In the present study we found that *Matricaria chamomilla* extract may be good source for the synthesis of Ag-NPs and these synthesized nanoparticles had considerable antibacterial activity against (*Staphylococcus aureus*, *Escherichia coli* and *Brucella melitensis*) isolated from dairy products. It was economical, non-toxic, and environmentally benign and has exhibit broad-spectrum biocide activity against different bacterial isolates.

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