



## Synthesis, characterisation and its antimicrobial studies of silver and gold nanoparticles and their nanoalloys

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### ABSTRACT

This paper explains the antimicrobial studies of Ag-Au alloys. The silver nanoparticles were prepared by seeding method. In past few years, wide applicability of silver nanoparticles (AgNPs) in various field attract for an approach of rapid, cost effective and eco-friendly synthesis of AgNPs that is expanding research toward biological methods(1). Gold Nanoparticles was prepared by citrate reduction method. The citrate reduction method for the synthesis of gold nanoparticles (GNPs) has known advantages but usually provides the products with low nanoparticle concentration and limits its application. Herein, we report a facile method to synthesize GNPs from concentrated chloroauric acid (2.5 mM) via adding sodium hydroxide and controlling the temperature. It was found that adding a proper amount of sodium hydroxide can produce uniform concentrated GNPs with low size distribution; otherwise, the largely distributed nanoparticles or instable colloids were obtained(2). Their nanoalloys are prepared. Their antimicrobial activities are studied using against Gram- negative bacteria. Standard strain of *Escherichia coli* (ATCC 25922 strain), was used for evaluating antimicrobial property of AgNps-AuNps Nanoalloys.

**Keywords:** AgNps-AuNps Nanoalloys, chloroauric acid and *Escherichia coli*

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### INTRODUCTION

Over the past few decades, inorganic nanoparticles, whose structures exhibit significantly novel and improved physical, chemical, and biological properties, phenomena, and functionality due to their Nano scale size, have elicited much interest. Discoveries in the past decade have demonstrated that the electromagnetic, optical, and catalytic properties of noble-metal Nano crystals are strongly influenced by shape and size (3). Recent studies have demonstrated that specially formulated metal oxide nanoparticles have good antibacterial activity (4), and antimicrobial formulations comprising nanoparticles could be effective bactericidal material. However, in minute concentrations, silver is nontoxic to human cells. The epidemiological history of silver has established its nontoxicity in normal use. Microbes are unlikely to develop resistance against silver, as they do against conventional and narrow-target antibiotics, because the metal attacks a broad range of targets in the organisms, which means that they would have to develop a host of mutations simultaneously to protect themselves. Gold nanoparticles (GNPs), also named as gold colloids, have attracted increasing attention due to their unique properties in multidisciplinary research fields (5). Although GNPs are defined by tiny size, significant quantities of GNPs are likely required in many commercial and industrial applications. Remarkably, novel emerging applications bring a huge growth of the global demand of GNPs. For instance, biomolecule- and/or biopolymer-conjugated GNPs are largely used as biomarkers and biodelivery vehicles in the medicine/pharmacy, and in cosmetic products.

Bimetallic nanoparticles, also called nanoalloys, are at the heart of nanoscience because of their ability to tune together composition and size for specific purposes. By approaching both their physical and chemical properties.(6)

## EXPERIMENTAL SECTION

**2.Synthesis and Characterisation of Silver Nanoparticles:****2.1.Chemicals Required:**

Silver nitrate, ascorbic acid, sodium citrate tribasic dihydrate, and cetyltrimethyl ammonium bromide (CTAB) were of the highest purity available (Sigma). These reagents were used as received without further purification.

**2.2Synthesis of Silver Nanoparticles:**

The silver nanoparticles used in this work were synthesized by different methods. It is seeding method. First, silver seeds were prepared by rapidly injecting 0.5 ml of 10 mM NaBH<sub>4</sub> into an aqueous solution (with continuous stirring) containing 0.5 ml of 0.01 M AgNO<sub>3</sub> and 20 ml of 0.001 M sodium citrate. The resultant solution was stirred for ~5 min and aged for 1.5 h. In this method, the spherical silver hydrosols were prepared by reducing aqueous AgNO<sub>3</sub> with sodium citrate at near-boiling temperature(7). In a typical procedure, an aqueous solution of AgNO<sub>3</sub> (100 ml, 0.001 M) was brought to boiling, and then 3 ml of the silver seed solution and an aqueous solution of sodium citrate were added so that the final concentration of sodium citrate in the reaction mixture became 0.001 M. The solution was heated until the color was greenish yellow. The solution was cooled to room temperature. The silver nanoparticles were purified by centrifugation. To remove excess silver ions, the silver pellet was washed three times with deionized water. A dried powder of the nanosize silver was obtained by freeze-drying.

**2.3Characterisation of Silver Nanoparticles:****2.3.1.Uv-Visible Spectrometer:**

AgNPs were ascertained by distinctive peak observed at 413 nm  $\lambda$  in the pilot range of 300-900 nm  $\lambda$ . The specific silver surface plasmon resonance band were observed at 400-450 nm  $\lambda$ , gave indication of smaller AgNPs formation (<30 nm) (Figure 1).

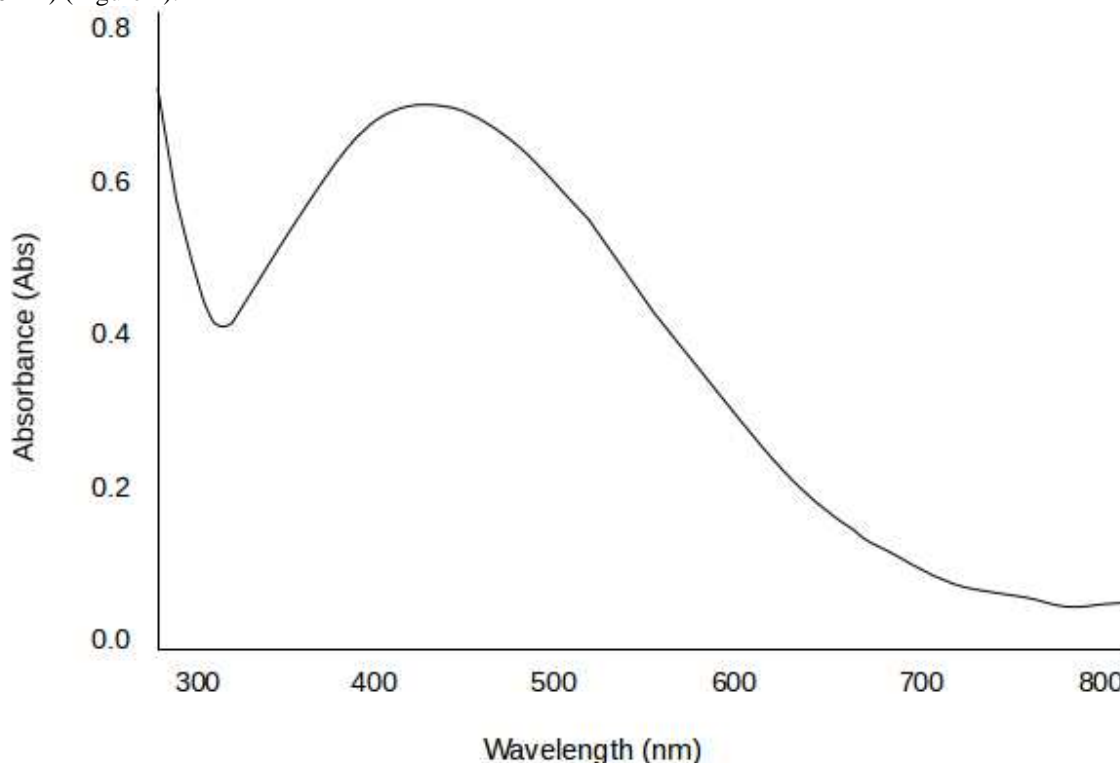


Fig.1.Uv-Visible Spectrometer of AgNps

**2.3.2 X-Ray Diffraction:**

X-ray diffraction is generally applied to certain chemical arrangement and crystal design of an objective; can be used for exposing the presence of AgNPs. The XRD patterns of resulted mixture gave signal that structure of AgNPs is face-centered cubic (fcc) and the AgNPs had a similar diffraction pattern. The X-ray Diffraction peaks observed in  $2\theta$  range of 30-80° and indexed to 111, 220 and 311 which confirms standard JCPDS file 04-0783 of silver are responsible for micrograph of the silver crystals (Figure 2).

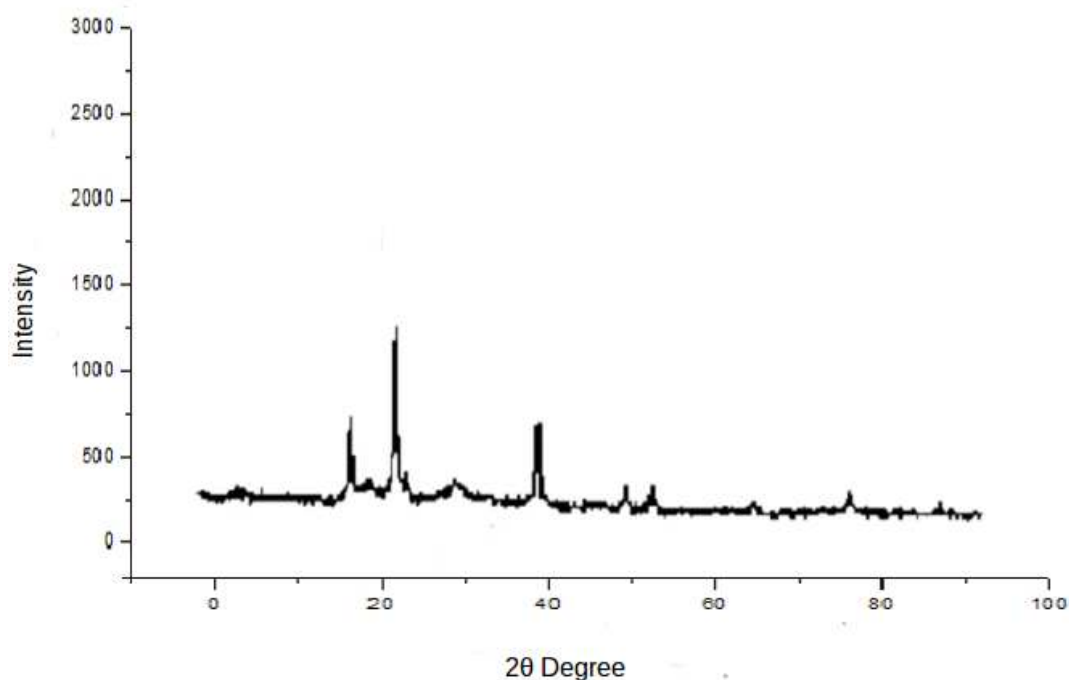


Fig.2.X-Ray Diffraction of AgNps

### 2.3.3.Scanning Electron Microscopy:

The structural characteristic of biosynthesised AgNPs were studied by scanning electron microscope, using an instrument of Hitachi S-4500(8). The average size of AgNPs observed from 10-30 nm with inter-particle distance and the shape was found to be spherical. The aggregation of the nanoparticles indicates that they were in the direct contact, but stabilized by a capping agent (Figure 3).

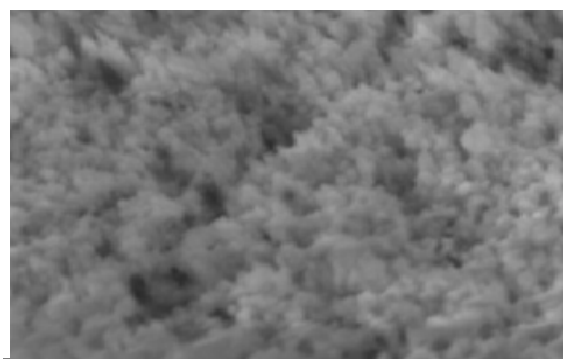


Fig.3.SEM Image of AgNps

### 2.3.4.Transmission Electron Microscopy:

The TEM images revealed Ag-nanocrystals were spherical in shape and in the range of 10-30 nm with considerable agglomeration (Figure 4).

## 3.Synthesis and characterization of AuNps :

### 3.1.Chemicals Required:

Hydrochloroauric acid trihydrate ( $\text{HAuCl}_4 \cdot 3\text{H}_2\text{O}$ , 99.9%), sodium citrate ( $\text{Na}_3\text{C}_6\text{H}_5\text{O}_7 \cdot 2\text{H}_2\text{O}$ , > 99%) and sodium hydroxide ( $\text{NaOH}$ , > 98%) were obtained from Sigma .All chemicals were used as received without any purification.

### 3.2.Synthesis of Au Nanoparticles:

GNPs were first synthesized from  $\text{HAuCl}_4$  solution with 50 ml deionized water in a round-bottom flask was added to 5mg chloroauric acid, respectively. After heating to boiling state, 0.3ml sodium citrate solution were rapidly introduced into the flask with drastic Stirring, respectively. The mixtures were continuously heated for a certain period till a ruby-red color appeared.

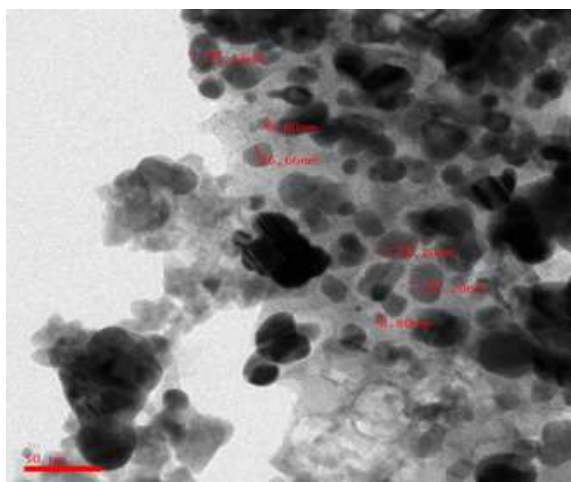


Fig.4.TEM image of AgNps

### 3.3.Characterisation of Au Nanoparticles:

#### 3.3.1.UV-VIS spectrophotometry:

Spectrophotometry is another important aspect for characterization of gold Nanoparticles. Generally, gold nanospheres display a single absorption peak in the visible range between 510-550 nm, because of surface Plasmon resonance and show heavy absorption of visible light at 520 nm. This gives brilliant red color to Gold Nanoparticle (GNP), which varies according to their size. The graphs the absorption of gold nanoparticle was measured in were plotted using single beam spectrophotometer and absorption diameter and the intensity of peak area as shown in the maxima was noted at different wavelength (390-630 nm Fig. 5).

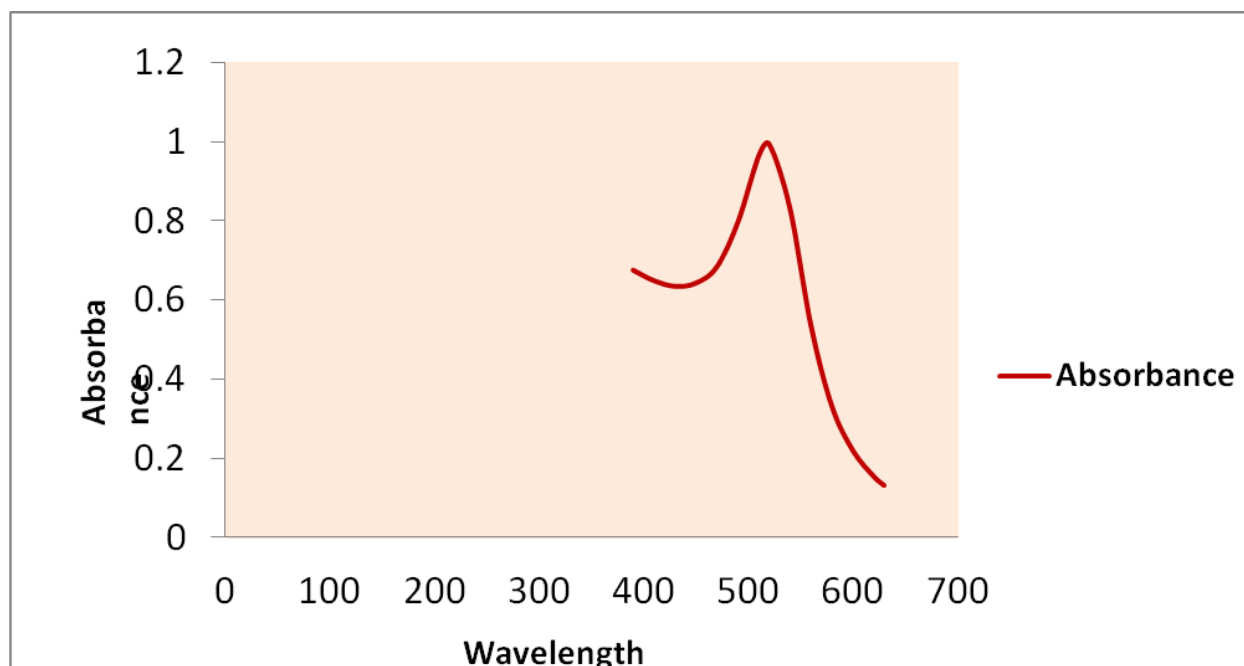


Fig.5 Uv-Visible Spectrometry for AuNps

#### 3.3.2.Transmission electron Microscopy:

The Transmission electron microscopy (TEM) images of the synthesized gold nanoparticles are reported in Fig.6 which shows highly stable and nearly spherical nanoparticles with average diameter in the range from 1–22nm

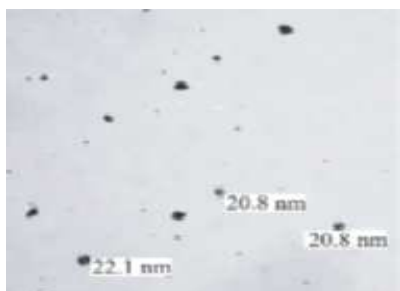


Fig.6. TEM micrographs of gold nanoparticles

### 3.3.3. Scanning Electron Microscopy:

The Scanning Electron Microscopy of Gold Nanoparticles are reported in Figure.7 which shows highly stable and nearly spherical nanoparticles with average diameter in the range from 10-11 nm.

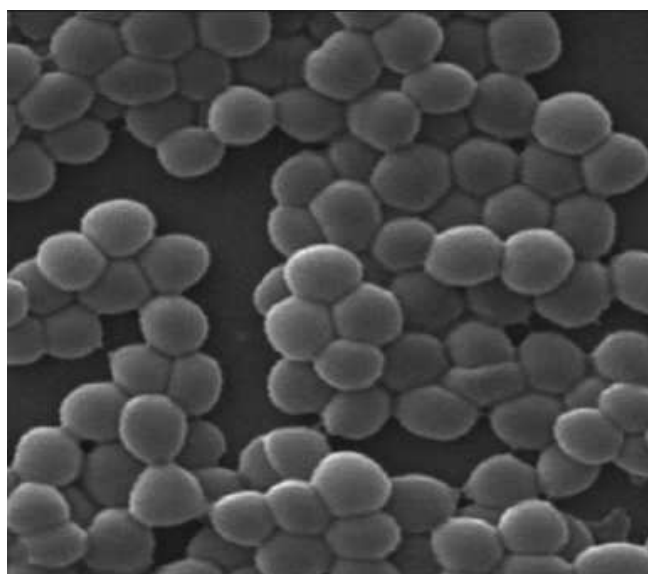
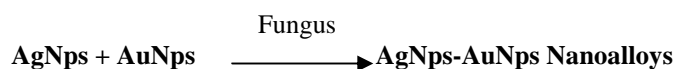


Fig.6. SEM image of gold nanoparticles

### 4. Synthesis of AgNps-AuNps Nanoalloys:

The exposure of a mixture of 1 mM AgNps and 1 mM AuNps solutions to different Percentage is Shown in the following table1. The results in the formation of highly stable Au–Ag alloy nanoparticles with dimensions of 8–14 nm depending on metal molar fraction. The amount of cofactor NADH released by the *Verticillium* fungus plays an important role in controlling the composition of the alloy nanoparticles.



#### 4.1. Composition AgNps and AuNps Nanoalloys:

The composition of AgNps - AuNps Nanoalloys given in Table.1

Table.1. Composition of AgNps-AuNps Nanoalloys

S.No	AgNps(%)	AuNps(%)
I	100	0
II	75	25
III	50	50
IV	25	75
V	0	100

#### 4.2. Antimicrobial Study of AgNps and AuNps Nanoalloys:

Anti microbial activity of AgNps-AuNps Nanoalloys were studied against *Escherichia coli* (ATCC 25922 strain), The bacterial strain used in test was grown on LB (Luria Bertani) Broth at 37°C overnight up to a turbidity of 0.5 Mac Farland standard (108 CFU per ml) (9). The agar well were filled with assorted concentration of of AgNps-AuNps nano alloys. The zone of inhibition is observed for AgNps-AuNps and their nano alloys.

### RESULTS AND DISCUSSION

The zone of inhibition observed at the surrounding area of AgNps-AuNps nano alloys filled with agar well". After incubation at 37°C for 24 hours. The test micro organism with AgNps-AuNps nano alloys. The result is indicated AgNps-AuNps nano alloys have strong-dose depend action against microbes. The experiment were done and mean values of ZOI were reported. The diameter of Inhibition in discussed in below table.2

Table.2.result discussion about zones

S.NO	Microbes	AgNps	zone of inhibition (mm)				Zone figure	
			Zone	AuNps	zone	Ag-Au nano alloys zone		
I	<i>E.coli</i>	100	22	0	-	100	22	A
II	<i>E.coli</i>	75	18.5	25	12	100	22.2	B
III	<i>E.coli</i>	50	15	50	13	100	22.5	-
IV	<i>E.coli</i>	25	14	75	17	100	22.1	C
V	<i>E.coli</i>	0	0	100	23	100	23	D



FIGURE A



FIGURE C



FIGURE B

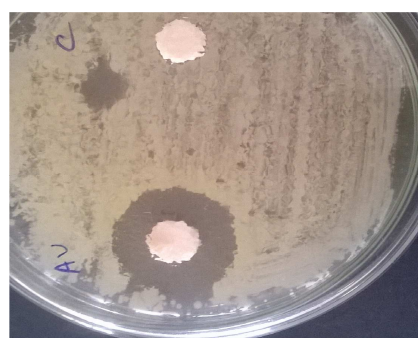


FIGURE D

### CONCLUSION

All these micro organism were found susceptible against nanoparticles "As growth of these microbes decreases with increase in concentration of of AuNps-AgNps Nanoalloys". So AgNps-AuNps Nanoalloys are Effective than AuNps and AgNps. The synthesis of AgNPs –AuNps Nanoalloys are without involvement of any toxic chemicals (10). It is acclaimed AgNPs –AuNps Nanoalloys have potent antimicrobial activity.

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