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Green Synthesis of AgNPs and CuNPs using *Tambala* (Pera) Stem Extract

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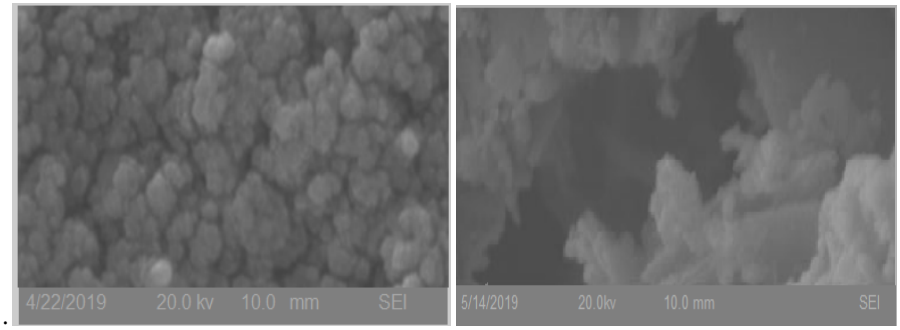
ABSTRACT

Eco-Friendly and cost-effective green synthesis method for silver and copper nanoparticles using Tambala (Pera) stem aqueous extract. The prepared silver (AgNPs) and copper Nanoparticles (CuNPs) was confirmed by using some techniques like SEM, X-RD, UV-Visible and EDX. AgNPs and CuNPs has different size having 18-22 nm and 43-58 nm. The average size of AgNPs and CuNPs were found to be 21.8 and 52.7 nm calculated by using sheerer equation. The prepared Silver and copper nanoparticles show microbial activities. The UV-Visible spectra for the AgNPs are obtained at 434 nm and for CuNPs it is found at 522nm. The capping molecules of NPs were negatively charged and were passably stable as revealed by zeta potential measurements and the XRD patterns illustrated their crystallinity. The synthesized AgNPs could efficiently inhibit various pathogenic organisms, including bacteria and fungi. The current research opens a new avenue for the green synthesis of nano-materials.

Graphical Abstract



SEM of AgNPs **SEM of CuNPs**

Keywords: AgNPs, CuNPs, *Tambala* (Pera) Stem, Eco-friendly synthesis, UV-Visible and SEM.

INTRODUCTION

Green synthesis of herbal extract is more important in chemical and physical system, these are cost effective, eco-friendly, easily scaled up for large-scale preparations. It is not required to use high

energy, temperature, pressure and hazardous chemicals. Using plants for nanoparticle preparation are superior over than additional biological developments as they are simply accessible, safe to handle, it eliminates the elaborate process of preserving cell cultures and have a broad changeability of metabolites that entertainment as reducing agents. Silver and copper has revealed a powerful inhibitory and a comprehensive spectrum of anti-microbial activity, which was handed down from the past to check and nurse various infections or diseases [**1**]. In previous few years, groups of bacterial infection in industrial sectors including environmental, food, particular care products, artificial textiles, packaging, healthcare, clinics, and public health hazard due to development of fight to antibiotics with their additional uses in bacteria have established universal consideration of scientific public to advance new age anti-microbial agents, formulations and methods [**2-4**]. Moreover, since of their large surface area these nanoparticles are used for purification of waste water treatment [**5**] and in the deletion of froths like arsenic, Lead, mercury and Sulphur from water or waste waters [**6**]. To overwhelmed the limitations of these conventional methods, green preparations has developed as an ecofriendly alternative for low-cost development of nanoparticles, which are extremely effective and biocompatible and can be used in a change of applications [**7, 8**].

In this research paper, we are prepared ecofriendly silver and copper nanoparticles of *Tambala* (Pera) stem extract. The prepared nanoparticles were characterized by UV-Visible spectroscopy (UV-Vis), Scanning electron microscopy (SEM) and X-ray diffraction (XRD) techniques. Moreover, the important factors for controlling the reaction and the anti-microbial activity of AgNPs and CuNPs were evaluated. This work provided a probable for the production of AgNPs and CuNPs without the participation of deadly chemicals.

MATERIALS AND METHODS

Materials: Silver nitrate ($AgNO_3$) and Copper Nitrate ($Cu(NO_3)_2$) was purchased from Sigma–Aldrich, Mumbai. The glass wares were washed with dilute nitric acid and water, then dried in hot air oven. *Tambala* (Pera) stem dry powder obtained from local market.

Preparation of *Tambala* (Pera) Stem extract: Stem Dry powder collected from local market from local area, Mumbai. The dry powder was taken in a beaker and 500 cm^3 of water was added. The resultant mixture was boiled for 3h with frequent stirring in order to get concentrated extract. Then extract was filtered by using Whatman filter paper and filtrate was collected in a beaker. This aqueous extract was kept in close container for father use.

Figure 1. *Tambala* (Pera) plant

Preparation of Silver nanoparticles (AgNPs): An aqueous solution 0.005M of $AgNO_3$ was prepared. he AgNPs were prepared by chemical reduction of an aqueous solution of 0.005m of 25 cm^3 silver solution and 50 cm^3 of extract. This solution firstly boiled to reduce the volume of solution one and

half than centrifugated at 4000 rpm for three hours during this process the colour of solution changes to brown. This solution kept overnight to get dark colour. From this solution UV-Vis was recorded.

Preparation of Copper nanoparticles (CuNPs): CuNPs were Eco-friendly prepared from 50 cm^3 of extract was taken in a 250 cm^3 beaker. To this, 25 cm^3 of 0.002M of copper solution was slowly added. The colour of the solution readily changed when Copper solution was added and colloidal suspension was formed. The suspension was then stirred for five hours at 500 rpm using magnetic stirrer bar. The stirring was stopped and precipitate was allowed to settle down. The solution was then centrifuged several times at 4000 rpm to get CuNPs. The solution dried at 60°C for four days and was stored in a tight sealed bottle and kept in dark place.

Method: Remi (1 MLH) magnetic stirrer with magnetic was bar used for synthesis of AgNPs and CuNPs. UV-Visible spectra were recorded on UV-Visible spectrophotometer (UV-2450), Shimadzu having Version 2.32 software. Scanning electron microscopy (SEM) images were obtained from S-4800 field emission SEM system (FEI Quanta 200) operating at 20.0 kV equipped to perform elemental chemical analysis by energy dispersive X-ray spectroscopy (EDX). XRD studies were carried out using Maxima 7000S XRD (Shimadzu).

RESULTS AND DISCUSSION

UV-Visible Analysis: UV-Visible spectra were recorded for AgNPs and CuNPs in the range of 250-800 nm. For this, minimum amount of CuNP were added in 5 cm^3 water and suspension was sonicated for 20 min to have better dispersivity of nanoparticles. As it can be seen from figure 2 band at 434 nm was observed which is a prominent characteristic of silver nanoparticles is. There was a little self-aggregation and AgNPs were found to be stable in aqueous medium. The UV-Visible spectra of CuNPs prepared from stem extract are shown in figure 3. The absorption band of CuNPs occurs at 522.0 nm. The CuNPs exhibit a yellowish-brown color in aqueous solution due to the excitation in UV-visible spectrum depending upon the particle size. The absorption bands for CuNPs have been reported to be in the range of 300-800 nm. The intensity of peak increases as a function of times increases.

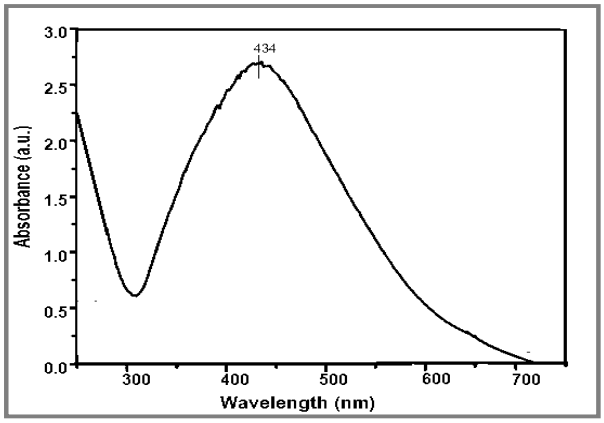


Figure 2. UV-Visible spectra of AgNPs.

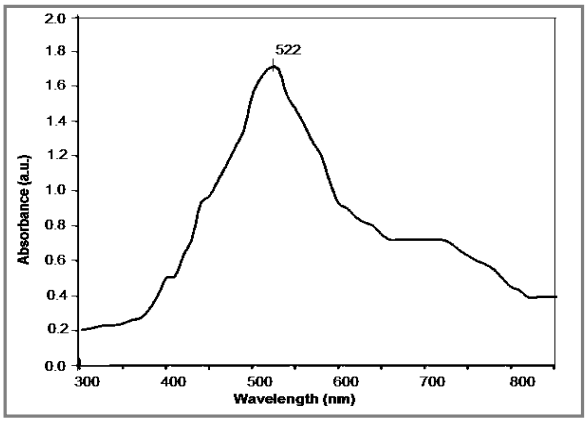


Figure 3. UV-Visible spectra of CuNPs.

X-Ray Diffraction (XRD) Analysis: The X-Ray of the silver and copper metal nanoparticles was recorded between 2θ values 20° and 90° degree and exhibits crystalline nature of the NPs with previous reports showing possible peaks of silver and copper metal in figure 4 and 5 [**9-10**].The formation of nanoparticles and its composition was analyzed using XRD technique by spreading the AgNPs on glass substrate with wide range of Bragg angles 2θ at a scanning rate of 2 θ min^{-1}. The five distinct diffraction peaks were obtained at 2θ = 38.5°, 44.5°, 64.7°, 77.5° and 81.7 (Figure 4). These peaks were accredited for (111), (200), (220), (311) and (211) planes of a faced center cubic (fcc) lattice. XRD analysis is also used for calculation of particle size by making use of Debye Scherrer equation [**11**] given below:

$$t = 0.89 \times \lambda / \beta \cos \theta. \quad \ldots(1)$$

Where t is particle size of nanomaterials, λ is X-ray wavelength, β is Line broadening half maximum intensity and θ = Braggs angle of diffraction. By incorporating all these values in equation (I), the particle size of AuNPs using *Tambala* (Pera) Stem extract was found to be 21.8 nm.

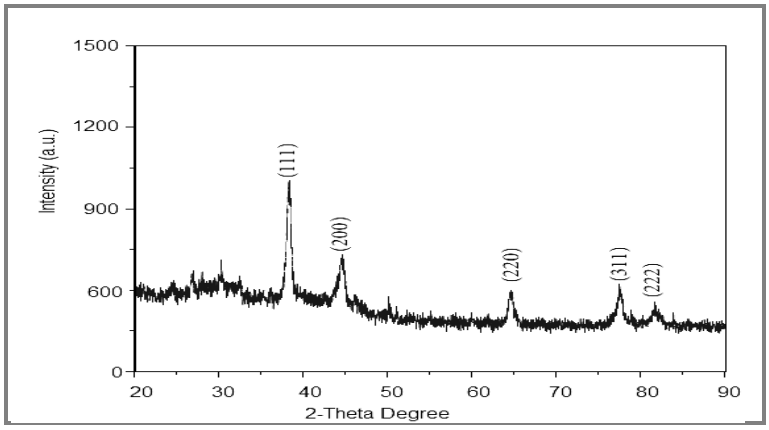


Figure 4. X-ray Diffraction AgNPs.

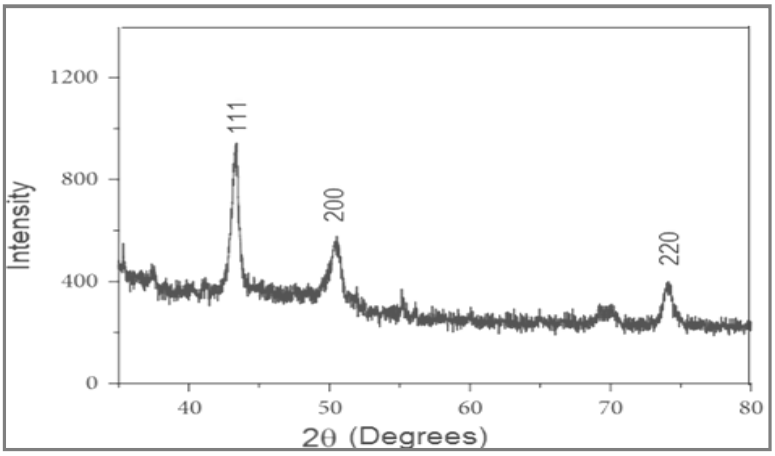


Figure 5. X-ray Diffraction AgNPs.

The X-Ray Diffraction of CuNPs was recorded between 2θ values 30° to 80° exhibits crystalline nature showing possible peaks of copper metal in figure -5. Bragg's diffraction peaks for copper nanoparticles are observed at 43.7°, 50.9°, 74.7° corresponding to 111, 200 and 220 respectively, representing face centered cubic structure of copper. The average crystallite size of CuNPs was calculated to be about 52.7 nm respectively, using Scherer formula, where λ is incident X-ray wavelength (Cu Kα = 1.542 Å), β is full width half maximum in radians of the prominent line that is, (111), and θ is position of that line in the pattern. The miller indices ($h\ k\ l$) were calculated by using d-spacing values and each peak of the diffraction pattern is assigned [12]. The data are listed in table 1.

Table 1. Peak Intensity of d-spacing Value for and CuNPs

S. No.	AgNPs				CuNPs			
	2θ	d-spacing	$1000/d^2$	hkl	2θ	d-pacing	$1000/d^2$	hkl
1	38.5	2.29	190.83	111	43.7	1.93	268.09	111
2	44.5	1.95	263.15	200	50.9	1.75	326.8	200
3	64.7	1.49	450.45	220	74.7	1.19	709.21	220
4	77.5	1.27	617.28	311	-	-	-	-
5	81.7	1.15	757.57	211	-	-	-	-

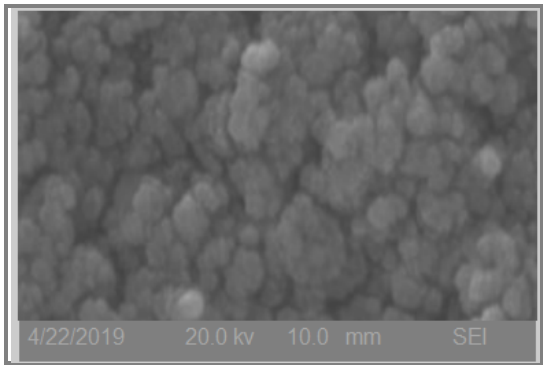


Figure 6. SEM of AgNPs.

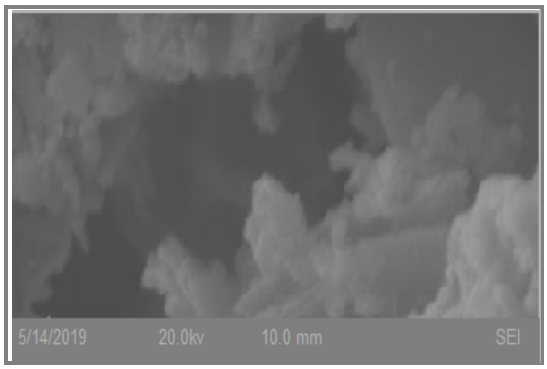


Figure 7. SEM of CuNPs.

APPLICATION

Applications of Silver and Copper Nanoparticles for determining their catalytic activity: The AgNPs synthesized by green method were employed to investigate catalytic activity, using reduction of 4-nitrophenol to 4-aminophenol by sodium borohydride as a standard/model reaction. To a 1 cm^3 cuvette, freshly prepared sodium borohydride (0.5 cm^3, 0.5 M) along with 4-nitrophenol (0.7 cm^3, 0.5 mM) were added. The UV-Visible absorption spectra were recorded for the reduction of 4-nitrophenol catalyzed by AgNPs. 4-aminophenol in aqueous form shows λ_{max} at 292 nm when scanned in the range of 200-800 nm, whereas 4-nitrophenol shows its characteristic spectra having absorption maxima at 318 nm. After addition of sodium borohydride, the peak for 4-nitrophenol (318 nm) shifts to 400 nm which is attributed to the formation of 4-nitrophenolate ion as observed in the literature. After addition of AgNPs to the previous solution, the cuvette was shaken vigorously for some time and was then placed in the spectrophotometer. The disappearance of peak at 400 nm was observed and new peak was formed at 292 nm which was because of the absorption of 4-aminophenol.

Antimicrobial activity: The antimicrobial potential of test compounds was determined on the basis of mean diameter of zone of inhibition around the disc in millimeters. The zones of inhibition of the tested microorganisms by the extracts were measured using a millimeter scale. Using Disc diffusion method an important reduction in the bacterial growth exhibiting a dose dependent consequence growth in concentration increased the zone of inhibition. The prepared Silver and copper nanoparticles showed significant antibacterial activity against gram positive and gram- negative bacteria. Nutrient Agar plates with specific cultures were incubated 24 h. The filter paper discs which were coated with silver and copper nano particles 25 mg dm^{3-1} it was placed on to the surface of agar plates [**13**]. The zone of inhibition after 24 h incubation at 37°C was recorded. The disc without nano particle was used as negative control. The Results shows that, AgNPs and CuNPs synthesized from *Tambala* (Pera) extract were more effective against bacteria. The AgNPs and CuNPs show the anti-bacterial activity against both gram positive and gram-negative bacteria. Comparing the zone of inhibitions, it can be concluded that the silver nano particles have greatest anti-bacterial against *E. coli* and *K. Pneumonia*.

Table 2. Microbial Activity of AgNPs and CuNPs

Concentration	*K. Pneumonia*		*E. Coli*	
	AgNPs(mm)	CuNPs(mm)	AgNPs(mm)	CuNPs(mm)
1.0M	5.4	4.4	8.7	7.1
0.5M	7.9	7.2	10.2	9.2
0.1M	8.2	8.1	11.7	10.4
0.05M	11.0	10.8	12.4	11.5
0.01M	12.8	12.0	13.2	13.1
0.005M	18.4	17.8	19.4	18.5
0.001M	20.3	19.4	22.1	21.8

CONCLUSION

Eco-friendly Green synthesis of AgNPs and CuNPs of *Tambala* (Pera) stem extract was achieved. The synthesis of these NPs was simple, safe, eco-friendly, which did not produce any harmful bi-products. The phytochemicals present in the plant acted as reducing agent for formation of AgNPs and CuNPs. The Synthesized NPs are stable and are characterized by means of UV-visible spectroscopy, X-Ray Diffraction and SEM. AgNPs and CuNPs has different size having 18-22 nm and 43-58 nm. The average size of AgNPs and CuNPs were found to be 21.8 and 52.7 nm calculated by using sheerer equation. The prepared Silver and copper nanoparticles show microbial activities. The UV-Visible spectra for the AgNPs are obtained at 434 nm and for CuNPs it is found at 522 nm. Synthesis of low-cost route of AgNPs and CuNPs has been found economically viable. These AgNPs and CuNPs proved to be an excellent catalyst for facile reduction of 4-nitrophenol to 4-aminophenol. Green synthesized MNPs have various relevance's in biochemical-pharmacological investigations such as anti-microbial activities. In addition, this green synthetic practice would be a better alternative to the accessible methods.

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