Synthesis and Characterization of Silver Nanoparticles by Chemical Reduction Method and their Antimicrobial Activities

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Abstract: Silver nanoparticles can be synthesized using a simple solvent free, economic and eco-friendly chemical reduction method. The production of silver nanoparticles from silver nitrate and subsequently reducing with glycerol. The structural characterization of synthesized nano particles was carried out using XRD and SEM. The optical characterization was carried out using UV and FTIR. The XRD result shows that the nano particles are of spherical shape and the average crystal size of the silver nano particle is in the range of 5nm and 10nm. The SEM analysis shows that the shape of the nano particles is nano spherical. The quality and purity of the silver nano particles are confirmed using XRD spectral analysis. The nanoparticles of silver showed high antimicrobial activity against gram positive bacteria such as Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa which is a highly methicillin resistant strain.

Keywords: Silver nanoparticles, XRD analysis, Optical properties, Antimicrobial activity.

1. Introduction

Nanotechnology is an emerging field of science which involves synthesis and development of various nanomaterials. Particles in the nano-range display unique physical and chemical properties and represent useful materials in biological applications. The integration of nanoparticles with biological molecules has lead to the development of diagnostic devices, contrast agents, and important tools in cancer therapy. Nanoparticles are now being developed for various biological applications such as medicines, antimicrobial agents[1,2,3], wound dressing[1,4], drug targeting and deliveries [1,5,6], transfection vectors [1,7], bioimaging and labelling agents [1,8]etc.

Silver nanoparticles can be synthesized using various methods: Biological [9], hydrothermal [10], electrochemical deposition [11], photochemical [12], pulsed laser deposition [13], etc. The most popular preparation of Ag colloids is chemical reduction of silver salts by sodium borohydride or sodium citrate. This preparation is simple, but the great care must be exercised to make stable and reproducible colloid. However, Solution temperature, concentrations of the metal salt, reducing agent and reaction time influences the particle size.

In the present work, the synthesis of silver nanoparticles from aqueous solution of silver nitrate using glycerol as a reductant is carried out. Further silver nanoparticles were characterized using UV-VIS spectrometer, scanning electron microscopy (SEM) and antimicrobial activity.
2. Materials and methods

2.1 Materials

The following analytical grade materials were used without further purification: silver nitrate (AgNO₃), A.C.S. reagent (Sigma – Aldrich, 99% purity by wt) and the glycerol (anhydrous) were obtained from Merck (99%).

2.2 Synthesis of Silver Nanoparticles

About 0.1g of the silver nitrate was weighed and made into a paste with 2 drops of glycerol in a 50ml silica crucible. The amount of glycerol used was optimized after several trials. The excess glycerol leads to charring of substances. The initial temperature was set to 50 °C and the temperature was slowly raised to 100 °C in the muffle furnace. The substance is maintained at 100 °C. The silvery white crystalline silver nanopowder obtained was characterized.

2.3 Characterization of Silver Nanoparticles

The surface morphology of the synthesized nanoparticle was characterized using SEM analysis. The scanning electron microscope used for this purpose is a Jeol-JSM-3.5 CF-Japan. The powder X-ray diffraction was performed using Scifert X-ray diffractometer with a CuKα radiation. The diffracted intensities were recorded from 10 to 70° angle. The absorption spectrum was recorded using PerkinElmer LS 45 spectrophotometer. For the spectroscopic analysis, Silver sample was dispersed in UV-VIS methanol with the help of the sonicator. The silver nano particles are sonicated in methanol for 10 min. The spectrum was recorded under room temperature.

2.4 Antimicrobial Test

The antimicrobial activity of silver nanoparticles is investigated on three types of pathogenic bacteria such as Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa that were cultured on agar plates added with same concentration of silver nanoparticles by disc diffusion method.

3. Results and Discussion

3.1 Structural Characterization and Morphology

The structural characterization was carried out using powder XRD. Fig. 1 shows typical XRD pattern of the as-obtained silver nano particles. All the diffraction peaks can be well indexed to the BCC phase of silver reported in JCPDS card. No. (JCPDS-89-1397).
The analysis of powder XRD pattern at room temperature shows that the sample formed is single phase with the BCC symmetry. The absence of extra peak claims the purity of the substance and also the complete conversion of silver nitrate.

From the above results obtained, it is clear that the synthesis of silver nanoparticles becomes chemical reduction method. The analyzed material is finely ground, and average bulk composition is determined. The particles or grain size of the particles on the silver nanoparticles was determined using Debye Sherrer’s equation.

\[ D = \frac{0.94 \lambda}{B \cos \theta} \]

![Fig.1. XRD spectrum for the silver nano particles.](image)

The surface morphology of the resulting powder was examined using scanning electron microscope. The SEM micrographs of the silver powder shown in Fig. 2 represent the formation of silver nano particle in single phase and the constituents are nano sphere. The nano spherical grown even up to the length of 5 nm and 10 nm.

![Fig.2. A&B - SEM micrograph of the silver nano particles.](image)

### 3.2 Optical Characterization of Silver Nanoparticles

The silvery white crystalline silver nano powder was insoluble in water and almost in all organic solvents. Hence a UV–Visible spectrum was recorded for the silver nano dispersed in methanol solution and is represented in Fig. 3. The absorption band observed at 207.15 nm is the characteristic peak of Silver nano material.
UV – VIS absorption results confirmed the formation of silver nanoparticles prepared in liquid by chemical reduction method (silver nitrate AgNO$_3$ is reduced by glycerol C$_3$H$_8$O$_3$).

![Graph showing UV-VIS spectrum of silver nanoparticles using methanol as solvent.](image)

**Fig.3. UV-VIS spectrum of silver nano particles using methanol as solvent.**

The FTIR spectra of pure silver nitrate and reduced silver nitrate show considerable variation in the peaks of spectra Fig.4. Regarding the silver nitrate is different types of peaks were found where as in the purified silver nanoparticles 6 peaks like 2390, 1760.7, 1630, 1405.2, 1274.5, 825cm$^{-1}$.And the peak centred at 1405.2cm$^{-1}$ which is present in the spectrum of Ag - nanoparticles. The reduction of certain peaks is the clear indication of the loss of certain groups.

![Graph showing FT-IR spectrum for the silver nanoparticles.](image)

**Fig.4. FT-IR spectrum for the silver nanoparticles.**

### 3.3 Antimicrobial Activity of Silver Nanoparticles

Silver nanoparticles are synthesized by the chemical reduction methods which have been found highly toxic against pathogenic bacteria. In this place, the silver nanoparticles are displayed in antimicrobial activity.

Synthesized silver nanoparticles are synthesized by chemical reduction method have been found highly toxic against pathogenic bacteria. Fig: 5 shows silver nanoparticles is exhibited in antimicrobial activity against both gram negative and gram positive pathogenic bacteria such as Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa as it shown clear inhibition zone.
The maximum ZOI values is observed as 6mm in Escherichia coli, 8mm in Staphylococcus aureus, and 4mm in Pseudomonas aeruginosa bacteria for 50µl concentration of silver Nanoparticles as shown in the Table: 1 the zone of inhibition (ZOI) for different pathogenic bacteria of silver Nanoparticles.

![Image](image1)

<table>
<thead>
<tr>
<th>S.N</th>
<th>Species</th>
<th>50µl</th>
</tr>
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<tbody>
<tr>
<td>1.</td>
<td>Escherichia coli</td>
<td>6±1mm</td>
</tr>
<tr>
<td>2.</td>
<td>Staphylococcus aureus</td>
<td>8±1mm</td>
</tr>
<tr>
<td>3.</td>
<td>Pseudomonas aeruginosa</td>
<td>4±1mm</td>
</tr>
</tbody>
</table>

**Conclusion**

In conclusion, the synthesis of very fine silver powder by the reaction of silver nitrate and glycerol were achieved. The SEM analysis tells us about the morphology of the particles which are of nano sphere. Silver nanoparticles the range is 5nm and 10nm were synthesized using glycerol as a reducing agent. The XRD analysis revels that the silver nanoparticles formed BCC close packing. The XRD spectrum revels the purity of the compound. The characteristic peaks in the absorption spectrum confirm the formation of silver nano particles. The silver nanoparticles are showing the good performance of antimicrobial activity against clinical pathogenic bacteria like Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa.
References