Green synthesis of copper (II) oxidenanoparticles (CuONPS) using leaves extract of *Urtica dioica*, Characterisations and antimicrobial activity

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Abstract

Recently, the applications of green synthetic processes in field of biological, chemical and pharmacological sciences have achieved essential roles. One of the major applications of green chemistry is the synthesis of zero valent metal and metal oxide nanoparticles using natural and abundant biological materials. In the present study, we have carried out the successful synthesis of copper (II) oxide nanoparticles (CuONPS) by using the leaf extract of *Urtica dioica* and formation of nanoparticles confirmed by a visual color change of the solution and then UV-Visible spectroscopy. The synthetic method was found very low cost, effective, efficient and eco-friendly. The synthesised nanoparticles have been characterised by using different analytical methods such as FTIR, UV-Visible, powder XRD and FESEM. The antibacterial activity of CuONPS was checked by well diffusion method against *S. aureus*, *S. mutans* and *E. coli*. The maximum zone of inhibition was found for *S. mutans* after incubation period and at specific dosage of copper nanoparticles.

**Keywords:** Green synthesis, CuONPS, Characterisation, Antibacterial activity

1. Introduction

The practices of green synthetic processes in the field of nanotechnology have emerged as one of important field of research. Since last few years, researches have been focussed in the synthesis of metal and metal oxides nanoparticles using abundantly available biomaterials (Joshi and Chhabra, 2019). Conventional methods used in the synthesis of nanoparticles are suffering with high cost, low efficiency and needed harmful chemicals. A green synthetic approach used to synthesise metal and metal oxide nanoparticles is a good alternative over the conventional methods and characterised by low cost, high efficiency, environmentally friendly and effectiveness (Joshi and Prakash, 2019). The commonly biologically synthesised metal and metal oxide nanoparticles are silver (Ag), gold (Au), copper (Cu), platinum (Pt), manganese (Mn), iron (Fe), copper oxide (CuO), manganese oxide (MnO₂), calcium oxide (CaO), magnesium oxide (MgO), zinc oxide (ZnO), titanium oxide (TiO₂) and iron oxide (Fe₂O₃). These nanoparticles show many unique properties like toughness, high surface/volume ratio, ductility, specific magnetisations, optical properties, magnetisation and industrial applications (Singh et al., 2018).

CuONPS are generally used in the electronics, sensors, heat transfer systems, catalysts and antimicrobial agents (Din et al., 2017). They have excellent conductive properties and also used to make conductive pastes of thick film conductors, electrodes in printed circuit boards and in multilayer ceramic capacitor in electronic nduType equation here.stries (Joshi and Prakash, 2019). Biologically synthesised CuONPS are found valuable with eco-friendly nature, high biocompatibility, high antioxidant activity and high antimicrobial properties (Din et al., 2017). The plant *Urtica dioica* is a common weed and found abundantly in the hills of Kumaun and Garhwal (India). Locally, this plant is known as bicho ghas, scorpion grass, siyon and kandali and fully comprised with thorns that contain histamine, acetylcholine and uric acid and on touch it causes irritation and itching. Many peoples use this plant as vegetables and for pain relief; Scorpion grass or *Urtica dioica* is a perennial herb (Fig. 1a and b) belongs to an Angiospermc family Urticaceae. Medicinally, *Urtica dioica* is used in the treatment of malaria, gall blotting, arthritsis and sprain (Ahmed and Parasuraman 2014).
2. Material And Methods:
All the chemicals used in the synthetic method were of analytical grade and glassware before experiment washed with deionized water to remove dust and other water soluble impurities and dried in tray dryer. The collected leaves of *Urtica dioica* washed properly and were cut into small pieces. The cut leaves were grinded and 2 g of grinded leaves mixed with 100 ml of distilled water in 250 ml Erlenmeyer flask. Boiled this content for 10 minutes and then filtered by using Whatman no.1 filter paper. The filtrate was used as an extract and preserved at 4°C for further studies.

Copper sulphate (CuSO$_4$.5H$_2$O) was used as a precursor and dissolved in double distilled water to make 0.02M solution. Then, mixed 5 ml of leaf extract with 100 ml of copper sulphate solution and the pH of reaction mixture was maintained 12 by adding 0.1M sodium hydroxide (NaOH) solution. The formation of copper nanoparticles was confirmed by the color change of copper sulphate solution from blue to green (Fig. 1d). Further, the formation of CuNPs was confirmed by UV-Visible spectroscopy and this solution was kept at room temperature for 10 hours. After that, the solution centrifuged at a fixed rpm (10,000) for 15-20 minutes and CuNPs were settled down. The pellets of CuNPs have been washed 2-3 times with double distilled water and then dried for 2 hours in a hot air oven at 60°C under controlled conditions and then calcined. The freshly synthesised CuONPS preserved for characterisations and antibacterial activities. Characterisation of biologically synthesised CuNPs was done by using FTIR, UV-Visible, FESEM and powder XRD. The antimicrobial activity of CuONPS has been checked by using well diffusion method against *Escherichia coli*, *Streptococcus mutans* and *Staphylococcus aureus*. The petri dishes or plates containing Muller Hinton Agar (MHA) were prepared by poured liquid media on the plates and then solidified. The CuNPs have been loaded in the wells on agar plates and incubated for 24 hours at 37°C.

3. Results And Discussion:
3.1 Characterisations of CuONPs:
**FTIR and UV-Visible:**
Fourier transform infra-red spectroscopy is basically used to identify the type of bonds present on the surface of biologically synthesised nanomaterials over a frequency range 4500-500 cm$^{-1}$. This spectroscopy is very economical and applied to organic, polymeric as well as inorganic species. FTIR is highly sensitive, low cost and interference based non destructive technique with high percisions. The FTIR spectra of CuNPs is shown in figure 1 and the broad peaks are found at 3410 cm$^{-1}$, 1632 cm$^{-1}$, 1385 cm$^{-1}$, 1097 cm$^{-1}$ and 604 cm$^{-1}$. These peaks are indicating that the presence of O-H, C=O, C-O, C-C, Cu-C bonds etc on the surface of copper oxide nanopowder (Joshi and Prakash, 2019). The UV-Visible spectroscopy is primarily used to a quantitative analysis and concerned with the absorption of radiations in ultra violet and visible region. This radiation provides electronic transitions of electron from lower to higher energy levels in atoms or molecules. Under controlled conditions, the amount of radiation absorbed is related to the concentration of substances in solution. UV-Visible spectra of CuONPS are represented in figure 2; small peaks are obtained at 360 nm and 490 nm. These peaks indicate the formation of copper oxide nanoparticles by adding the leaf extract of *Urtica dioica* in precursor’s solution after 10 minutes.
3.2 FESEM and powder XRD:
Field Emission Scanning Electron Microscopy (FESEM) is used to explain a topographical study of nanoparticles. The morphological features of nanoparticles are studied by using the FESEM technique. The electrons are liberated from an emission source and focussed and deflected by electronic lenses inside high vacuum to produce a narrow scan beam. This narrow scan beam is used to scan the objects; the FESEM images of CuONPs (Fig. 3) indicate the polymorphic morphology and collective forms of CuONPs (Round et al., 1999). Powder X-ray diffraction (XRD) pattern of CuONPs is represented in figure 4, a diffraction pattern is obtained by the curve of intensity versus angle of diffraction (2θ). The characteristic peaks have been obtained at 18.6, 22.2, 42.4, 51.2 and 73.40. These peaks are corresponding to 011, 101, 111, 200 and 220 planes (JCPDS card no. 04-0836) for the standard spectrum of face centred cubic.
3.3 Antimicrobial activity:
Metal nanoparticles have been recognised for many applications in the areas of biotechnology, toxicology and medicine. Like gold (Au) and silver (Ag) nanoparticles, CuONPs are also excellent antibacterial agents and due to unique physical, chemical and small size, they can enter very easily into the cells of pathogenic bacterial species. Further, inhibition mechanisms take place inside the bacterial cells (Joshi and Chhabra, 2019). The dosage of biologically synthesised CuONPs were loaded in the wells on the agar plate and after incubation period a significant zone of inhibition obtained for *E. coli*, *S. mutans* and *S. aureus* (Table 1).

<table>
<thead>
<tr>
<th>Intensity</th>
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<tr>
<td>18000</td>
<td>14000</td>
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<td>16000</td>
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<tr>
<td>14000</td>
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<td>8000</td>
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Table 1 Antibacterial activity of CuONPs synthesised by using *Urtica dioica*’s leaf extract
<table>
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<tr>
<th>Bacterial species</th>
<th>Dosage of CuONPs (mg/ml)</th>
<th>Zones of inhibition (mm)</th>
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<tr>
<td><em>E. coli</em></td>
<td>10</td>
<td>36</td>
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<tr>
<td><em>S. mutans</em></td>
<td>10</td>
<td>44</td>
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<tr>
<td><em>S. aureus</em></td>
<td>10</td>
<td>22</td>
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**CONCLUSIONS:**
In the present work, we have carried out an efficient green synthesis of copper oxide nanoparticles (CuONPs) by using the leaf extract as a reducing agent of an abundantly found weed *Urtica dioica*. Synthesis process was found very economical and valuable in an eco-friendly manner. The synthesised nanoparticles were analysed by FTIR, UV-Visible, XRD and FESEM and tested for antibacterial activity against *E. coli*, *S. mutans* and *S. aureus*.

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**Conflict of interest:**
Authors declare that they have no conflict of interest.

**References:**