ISSN- 0975-1491 Vol 7, Issue 10, 2015

## **Short Communication**

# EICHHORNIA MEDIATED COPPER OXIDE NANOPARTICLES: IN VITRO ANALYSIS OF ANTIMICROBIAL ACTIVITY

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Received: 24 Jul 2015 Revised and Accepted: 22 Aug 2015

#### ABSTRACT

**Objective:** The present investigation determines the biological synthesis and characterization of Copper oxide nanoparticles from aqueous extract of *Eichhornia crassipes* and assessing its effects on antimicrobial activity against the pathogens.

**Methods:** In this method *Eichhornia* mediated copper oxide nanoparticles were synthesized and characterized by FT-IR, FESEM and EDX and also antimicrobial activity was determined using the well diffusion method.

**Results:** The antimicrobial activity of *Eichhornia* mediated copper oxide nanoparticles was tested against selective pathogens and maximum zone of inhibition was observed in *S. aureus* and *A. flavus* at  $100 \mu g/ml$  concentration.

**Conclusion:** The green synthesized copper oxide nanoparticles have antimicrobial activity against selective microorganisms and it can be effectively used as a good antimicrobial agent.

**Keywords:** Copper oxide, *Eichhornia*, EDX (Energy-dispersive X-ray spectroscopy), FESEM (Field emission scanning electron microscope), Fourier transform infrared spectroscopy (FT-IR), Nanoparticles and Antimicrobial activity.

Copper oxide nanoparticles achieved more significant interest due to their unique physical and chemical properties. They are widely used as gas sensors [1-3], superconductors [4-7], catalysts [8], solar energy exchange tools [9], etc. Copper oxide nanoparticles play a vital function in medicinal field as antioxidants [10] and antibacterial [11] agents. Chemical methods have adverse effects in medicinal field due to the inclusion of toxic compounds and to overcome this, green chemistry method is used for the synthesis of nanoparticles. In green methods, extract of living organisms such as plants, microorganisms and enzymes etc., are used [12]. Tabernaemontana mediated synthesized copper oxide nanoparticles are monodispersed with an average size of 48±4 nm and its antimicrobial activity was seen against UTI pathogens [13]. Acalypha indica mediated copper oxide nanoparticles were biosynthesized with an average size between 26-30 nm and its effect was seen against antimicrobial and anticancer agent [14]. Eichhornia crassipes (Family: Pontederiaceae) is one of the aquatic noxious weeds of the world. It is resistant to all eradication methods (mechanical, chemical, biological or hybrid means). Therefore, nano biotechnology approaches have been utilized to minimize the difficulty of aquatic weed exclusion and management [15].

In this study, the green synthesized copper oxide nanoparticles were tested for antimicrobial activity using well diffusion method.

E. crassipes were collected from Ukkadam Lake, Coimbatore, Tamil Nadu, India (11 °31'N; 76 °39' E). All the chemicals used in this analysis were bought from Sigma-Aldrich chemicals, India and double distilled water has been used for synthesis of copper oxide nanoparticles. Bacterial and fungal strains such as Escherichia coli, Pseudomonas aeruginosa, Pseudomonas fluorescens, Proteus vulgaris, Staphylococcus aureus, Candida albicans, Aspergillus flavus , Aspergillus niger and Fusarium oxysporium were obtained from the Department of Microbiology, School of Life Sciences, Karpagam University, Coimbatore, India. The culture samples were maintained on nutrient broth and potato dextrose broth.

*Eichhornia* mediated copper oxide nano particles were synthesized according to Sivaraj *et al.* 2014. The functional groups, elemental analysis and shape of the green synthesized copper oxide nanoparticles were analysed by Fourier transform infrared spectroscopy (Perkin-Elmer 1725X), Field emission scanning electron microscopy (Model

JSM6390LV, JOEL, USA) and Energy dispersive X-ray spectrometer (RONTEC's EDX system, Model Quan Tax 200, Germany.

Antimicrobial activity of <code>Eichhornia</code> mediated copper oxide nanoparticles were determined using well diffusion method [16]. The bacteria and fungi were cultured in nutrient and potato dextrose broth at room temperature and kept in orbital shaking incubator (Remi, India) at 200 rpm for 2-3 d. The muller hinton agar plates (bacterial culture) and potato dextrose agar plates (fungal culture) were prepared and microbial strains were swabbed. After 5 min the well (5 mm size) was made by using gel puncher and different concentrations (25  $\mu g$ ,  $50~\mu g$ ,  $75~\mu g$  and  $100~\mu g/ml$ ) of the copper oxide nanoparticles was added in the well. The positive control (10  $\mu g/ml$ ) (tetracycline) and (amphotricin B) was prepared and poured into wells. The plates were incubated at 37 °C for 24h (bacteria) and room temperature for 48h (fungi). After incubation, the antimicrobial activity was assessed. Each screening test was performed with three replicates and the mean values are recorded.

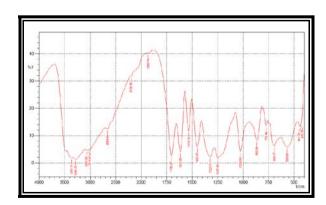


Fig. 1: FT-IR spectra showing the functional groups present on the surface of synthesized *Eichhornia* mediated copper oxide nanoparticles

FT-IR spectra analysis was performed to find the functional groups present in the surface of green synthesized copper oxide

nanoparticles (fig. 1). The spectrum showed bands at 428 and 497 cm<sup>-1</sup> corresponding to metal-oxygen (M-O). The band at 819 and 912 cm<sup>-1</sup>shows the presence of C-C stretching of alkanes [17, 18]. The bands at 1354 and 1624 cm<sup>-1</sup> refer to N-H bending mode. The intense bands observed at 1217 cm<sup>-1</sup> corresponding to C-O-C stretch was also obtained [17].

Field emission scanning electron microscope (FESEM) showed that the copper oxide nanoparticles (fig. 2) are spherical in shape. This is similar to the earlier literature [19].

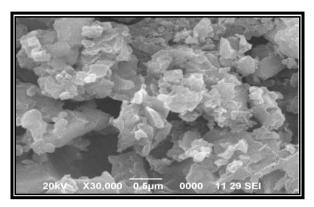


Fig. 2: FESEM showing the shape of the synthesized *Eichhornia* mediated

### Copper oxide nanoparticles

It is shown that 79.18% of copper and 20.82% of oxygen were present in green synthesized *Eichhornia* mediated copper oxide nanoparticles by Energy-dispersive X-ray spectroscopy (EDX) (fig. 3) which was very similar to the reports, Rajeshwari *et al.* [14], Rajiv *et al.* [20] and Vanathi *et al.* [15].

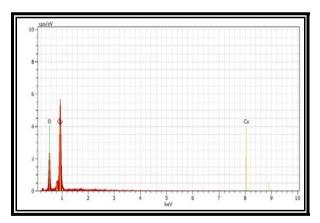


Fig. 3: EDX spectra showing the percentage of copper and oxygen in Eichhornia mediated copper oxide nanoparticles

Fig. 4 and fig. 5 shows the results of the antibacterial activity of the *Eichhornia* mediated copper oxide nanoparticles. The zone of inhibition was maximum at the concentration (100  $\mu$ g/ml) of copper oxide nanoparticles. The highest zone of inhibition was observed in *S. aureus* (25.33±1 mm) and *A. flavus* (21.66±1 mm) at a concentration of 100  $\mu$ g/ml. Similarly, the lowest zone of inhibition was observed in *Proteus sp.*, (15.33±1 mm) and *Fusarium culmorum* (9.21±1 mm) at a concentration of 25  $\mu$ g/ml which are similar to the previous studies [21]. Rajiv *et al.* reported the antifungal activity of *Parthenium* mediated zinc oxide nanoparticles [20]. Rajeshwari *et al.* reported the antibacterial activity of copper oxide nanoparticles against UTI pathogens [13]. Azam *et al.* found the antibacterial

activity of copper oxide nanoparticles against both gram-positive and negative bacterial strains [11]. The results confirm that green synthesized copper oxide nanoparticles shows excellent antimicrobial activity.

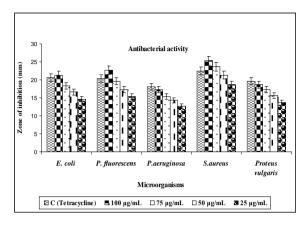


Fig. 4: Antibacterial activity of *Eichhornia* mediated copper oxide nanoparticles

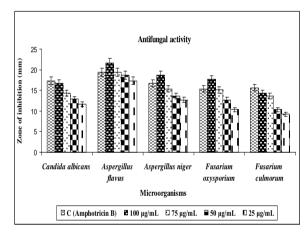


Fig. 5: Antifungal activity of *Eichhornia* mediated copper oxide nanoparticles

The study demonstrates the green synthesis of copper oxide nanoparticles from *E. crassipes* extract by simple, inexpensive and eco friendly method. It has been confirmed from the results that the *Eichhornia* mediated copper oxide nanoparticles have effective antimicrobial activity against *A. hydrophila, S. pyogenes, S. aureus, E. coli, A. flavus, F. culmorum* and *P. aeruginosa*.

#### **ACKNOWLEDGEMENT**

We thank to Management of Karpagam University, Coimbatore, Tamil Nadu, India for providing necessary facilities to carryout this

#### **CONFLICT OF INTERESTS**

**Declared None** 

## REFERENCES

- Bednorz JG, Muller KA. Possible high Tc superconductivity in the Ba-La-Cu-O system. Z Phys B 1986;64:189–93.
- Berry AD, Gaskill KD, Holm RT, Cukauskas EJ, Kaplan R, Henry RL. Formation of high T superconducting films by organometallic chemical vapor deposition. Appl Phys Lett 1988;52:1743–5.
- Malandrino G, Condorelli GG, Lanza G, Fragala IL. Growth of epitaxial TlBaCaCuO a-axis oriented films on LaAlO3 buffer

- layers grown on SrTiO3 substrates. J Alloys Compd 1997;251:314–7.
- Malandrino G, Condorelli GG, Lanza G, Fragala IL, Uccio US, Valentino M. Effect of Ba---Ca---Cu precursor matrix on the formation and properties of superconducting Tl2Ba2Can-1CunOx films a combined metalorganic chemical vapour deposition and thallium vapour diffusion approach. J Alloys Compd 1997;251:332-6.
- 5. Ishihara T, Higuchi M, Takagi T, Ito M, Nishiguchi H, Takita T. Preparation of CuO thin films on porous  $BaTiO_3$  by self-assembled multibilayer film formation and application as a  $CO_2$  sensor. J Mater Chem 1998;8:2037–42.
- Ishihara T, Kometani K, Hashida M, Takita Y. Application of mixed oxide capacitor to the selective carbon di oxide sensor. J Electrochem Soc 199;138:173–6.
- Tamaki J, Shimanoe K, Yamada Y, Yamamoto Y, Miura N, Yamazoe N. Dilute hydrogen sulfide sensing properties of CuO-SnO<sub>2</sub> thin film prepared by low-pressure evaporation method. Sens Actuators B 1998;49:121–5.
- Zhou K, Wang R, Xu B, Li Y. Synthesis, characterization and catalytic properties of CuO nanocrystals with various shapes. Nanotechnol 2006;17:3939-40.
- 9. Ko J, Kim S, Hong J, Ryu J, Kang K, Park C. Synthesis of graphene-wrapped CuO hybrid materials by  $CO_2$  mineralization. Green Chem 2012;14:2391–3.
- Das D, Nath B, Phukon P, Dolui S. Synthesis and evaluation of antioxidant and antibacterial behavior of CuO nanoparticles. Colloids Surf B 2012;101:430–3.
- 11. Azam A, Ahmed AS, Oves M, Khan MS, Memic A. Antimicrobial activity of metal oxide nanoparticles against Gram-positive and Gram-negative bacteria: a comparative study. Int J Nanomed 2012;7:3527–35.
- 12. Parashar V, Parashar R, Sharma AC, Pandey AC. Parthenium leaf extract mediated synthesis of silver nanoparticles: a novel approach towards weed utilization. Digest J Nanomater Biostruc 2009;4:45–50.

- Rajeshwari Sivaraj, Pattanathu Rahman KSM, Rajiv P, Hasna Abdul Salam, Venckatesh R. Biogenic copper oxide nanoparticles synthesis using *Tabernaemontana divaricate* leaf extract and its antibacterial activity against urinary tract pathogen. Spectrochim Acta Part A 2014;133:178–81.
- Rajeshwari Sivaraj, Pattanathu KSM Rahman, Rajiv P, Narendhran S, Venckatesh R. Biosynthesis and characterization of *Acalypha indica* mediated copper oxide nanoparticles and evaluation of its antimicrobial and anticancer activity. Spectrochim Acta Part A 2014:129:255–8.
- 15. Vanathi P, Rajiv P, Narendhran S, Sivaraj Rajeshwari, Pattanathu KSM Rahman, Rajendran Venckatesh. Biosynthesis and characterization of phyto mediated zinc oxide nanoparticles: a green chemistry approach. Mater Lett 2014;134:13–5.
- Bauer AW, Kirby WM, Sherris JC, Turck M. Antibiotic susceptibility testing by a standardized single disk method. Am J Clin Pathol 1966;45:493–6.
- 17. Huang J, Li Q, Sun D, Lu Y, Su Y, Yang X, *et al.* Biosynthesis of silver and gold nanoparticles by novel sundried *Cinnamomum camphora* leaf. Nanotechnol 2007;18:105104–15.
- Tas AC, Majewski PJ, Aldinger F. Chemical preparation of pure and strontiumand/or magnesium-doped lanthanum gallate powders. J Am Ceram Soc 2000;83:2954–60.
- Asharf Shah M, Al-Ghamdi MS. Preparation of copper (Cu) and copper oxide (Cu<sub>2</sub>O) nanoparticles under supercritical conditions. Mater Sci Appl 2011;2:977-80.
- Rajiv P, Rajeshwari S, Venckatesh R. Bio-Fabrication of zinc oxide nanoparticles using leaf extract of *Parthenium hysterophorus* L. and its size-dependent antifungal activity against plant fungal pathogens. Spectrochim Acta Part A 2013:112:384-7.
- Jayaseelan C, Abdul Rahuman A, Vishnu Kirthi A, Marimuthu S, T Santhoshkumar, Bagavan A, et al. Novel microbial route to synthesize ZnO nanoparticles using Aeromonas hydrophila and their activity against pathogenic bacteria and fungi. Spectrochim Acta Part A 2012;90:78–84.