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Green Synthesis and Characterization of Copper Nanoparticles Using Tulsi (*Ocimum Sanctum*) Leaf Extract

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ABSTRACT

The present study deals with green synthesis and characterization of copper nanoparticles using Tulsi *Ocimum sanctum* leaf extract. The leaf extract acts as both reducing and capping agent. The synthesized copper nanoparticles were confirmed by the change of colour after addition of leaf extract into the Copper Sulphate solution. The synthesized copper nanoparticles are characterized by UV–Vis Spectrometry analysis, Fourier transform Infrared analysis (FTIR), Scanning Electron microscopy (SEM) and Energy dispersive X-ray analysis (EDX). It was observed that the Ocimum sanctum leaf extract can reduce copper ions in to copper nanoparticles within 1 hr. Thus, this method can be used for rapid and ecofriendly biosynthesis of stable copper nanoparticles.

KEYWORDS Green synthesis, characterization, copper nanoparticles, *Ocimum sanctum*, leaf extract.

INTRODUCTION:

Nanotechnology plays a very important role in modern research and it is the most capable technology that can be applied almost all fields. It's growing interest in therapeutic filed for the ailments such as infection, cancer, allergy, diabetes and inflammation (Saranyaadevi *et al*., 2014). Among heavy metals, copper is most often leads to human poisoning and it is required by the body in small amounts, but toxic in larger doses (Akpor *et al*., 2010). The copper nanoparticles are potentially applied in the food package, water treatment, pharmaceutical, electronics, health care, food and feed, drug and gene delivery, energy science, cosmetics and environmental health. (Vasudeo Kulkarni *et al.,* 2015). Chemical synthesis is costly, toxic and potentially dangerous to the environment (Monalisa *et al*., 2013). But the biological approach of synthesis of nanoparticles is safe, cheap, low energy and time efficient (Javad Karimi *et al.,* 2013). Copper nanoparticles have been successfully synthesized by radiolysis, laser irradiation, thermal decomposition, vapour deposition, flame spray and chemical reduction. However, these methods suffer from drawbacks such as unsafe reaction condition, use of expensive chemicals, instruments and longer reaction time. To overcome these, green synthesis of copper nanoparticles are reported using plant leaf extracts (Ipsa Subhankari and Nayak 2013). The study related to the green synthesis and characterization of copper nanoparticles using Tulsi *Ocimum sanctum* is totally wanting. Hence the present study was carried out.

METERIALS AND METHODS:

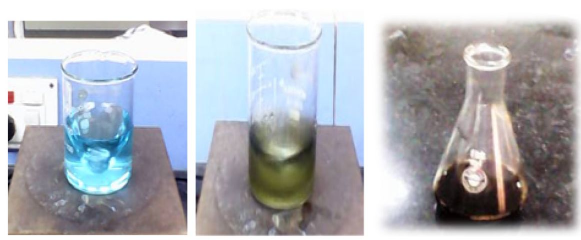
Collection and Preparation of Tulsi (*Ocimum sanctum)* leaf extract:

The leaf samples of Tulsi *Ocimum sanctum* were collected from Palani, Dindigul district, Tamil Nadu, India. Then it was cleaned and subsequently dried under shade to remove moisture completely. Then powdered and stored (Mahavinod *et al*., 2014)**.** 10 g of powdered leaves were taken in a beaker along with 100 ml of deionized water and it is allowed to boil at 60°C for 30 min under reflux condition, cooled down to room temperature (Heera *et al.,* 2015). The prepared solution was initially filtered through normal filter paper there by powdered leafy materials and filtered out. The filtrate was again filtered through What- man No.1 filter paper to get clear solution. The filtrate was stored at 4°C for future work (Saranyaadevi *et al*., 2014).

Synthesis and characterization of Copper nanoparticles

For the synthesis of copper nanoparticles (Cu NPs) 25 ml of *Ocimum sanctum* leaf extract was added to 100ml of 1mM aqueous $CuSO4.5H2O$ solution under continuous stirring. After complete addition of leaf extract, the mixture was kept for incubation for 24 hrs (Saranyaadevi *et al*., 2014). At particular time, the colour of the solution changes from light green to dark green. This indicates the formation of copper nanoparticles. Then the solution was centrifuged at 6000 rpm for 30 min followed by re-dispersion of the pellet in deionized water to remove any unwanted biological materials (Sreemanti Das *et al*., 2013). Biosynthesis of copper nanoparticles is given in figure 1 (A, B, C) and Table 1. The bio synthesised copper nanoparticles was characterized by using UV-Visible Spectroscopy (Lambda-35 Spectrophotometer) and Fourier transfer Infra Red Spectroscopy. The morphology of the prepared Cu product was examined by using SEM. The elemental composition of the reaction mixture was determined by EDX analysis (Saranyaadevi *et al*., 2014).

Figure: 1 Biosynthesis of copper nanoparticles. (A) Copper Sulphate, (B) Leaf extract (Ocimum sanctum), (C) Synthesized Copper Nanoparticles**.**



(A) **(B)** **(C)**

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Table 1: Colour change during biosynthesis of copper nanoparticles

S.No.	Solution	Before reduction	After reduction	Colour intensity	Time
1.	Ocimum sanctum leaf extract	Brown colour	Green colour	++	1 hr
2.	Copper sulphate ($CuSo_4$, $5H_2O$)	Blue colour	Green colour	++	1hr

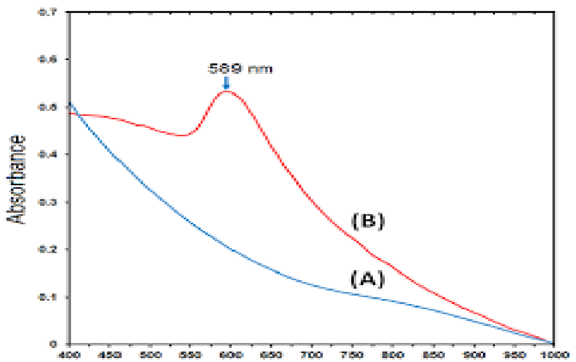
Colour intensity: Light - +, dark - ++ and high dark - +++

RESULTS:

I. UV- Visible Spectroscopy analysis:

It is generally recognized that UV- Visible spectroscopy could be used to examine the size and shape controlled nanoparticles in aqueous suspensions. Absorption spectra of copper nanoparticles formed in the reaction media has absorbance's peak at 589 nm is given in fig 2.

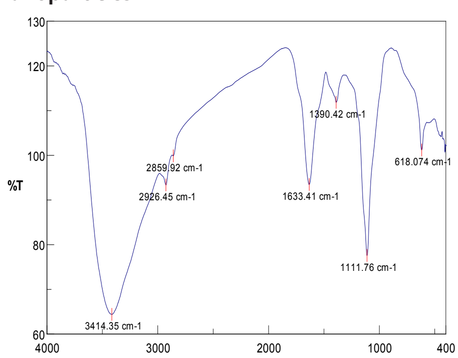
Figure: 2 UV-visible absorption spectroscopy analyses (A) leaf extract (B) copper Nanoparticles



II. Fourier transform infra red Spectroscopy (FTIR) analysis:

The FTIR analysis was used to identify the capping and stabilizing capacity of the leaf extract. FTIR measurement of plant extract and copper nanoparticles is given fig. 3(A) and (B) and Table 2 and 3. In Fig 3 (a) aqueous leaf extract showed the peaks at 3999.89, 2925.48, 1608.34, 1116.58, 617.109 cm^{-1}. The peak at 3999 cm^{-1} was due to the presence of O-H stretching of phenolic compound, C-H stretching of amines and N-H bending of amines as given in the table 2. In Fig 3 (b) for the copper nanoparticles, peak values at 3414.35cm^{-1}, 2926.45cm^{-1}, 59.92cm^{-1}, 1633.41 Cm^{-1}, 1390.42cm^{-1}, and 618.074cm^{-1} was observed. Peak at 618.074, 3999.89cm^{-1} corresponds to O-H stretching of phenolic compound, C-H bending of aldehydes compounds, C-H stretching of alkenes and N-H bending of amines is given in table 3. This functional group plays a very important role in the synthesis copper nanoparticles.

Figure 3: FT-IR analysis of (A) plant extract and (B) Copper nanoparticles



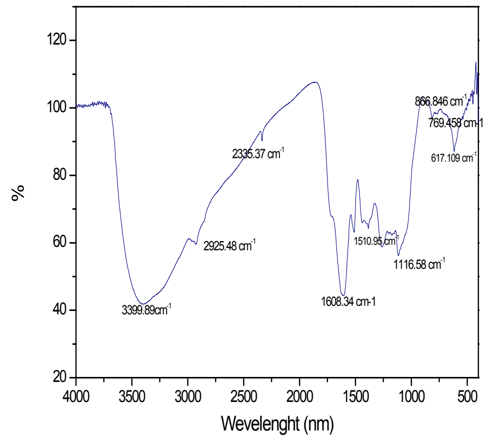


Table: 2. FT-IR analysis of leaf extract

1.	Frequency	Bond	Functional group
2.	3399.89	OH-Stretch	Alcohols and Phenols
3.	2925.48	CH- Stretch	Alcohols and Phenols
4.	2335.37	CH- Stretch	Alcohols and Phenols
5.	1608.34	NH-Bend	1^0 Amines
6.	1384.64	N=O bend	Nitro group
7.	1116.58	C-NH_2	Primary aliphatic amines
8.	617.109	C-H	Alkynes

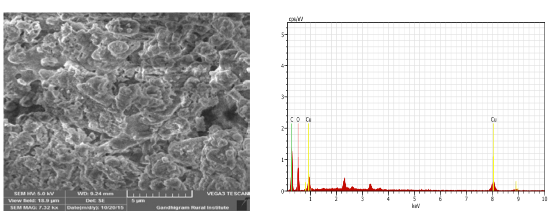
Table: 3 FT-IR analysis of copper nanoparticles

1.	Frequency	Bond	Functional group
2.	3414.35	OH-stretch	Alcohols and Phenols
3.	2926.45	CH stretch	Alkanes
4.	2859.92	CH stretch	Alkanes
5.	1633.41	C = O	β diketones
6.	1390.42	COO^-	Carboxylic acid
7.	1111.76	C-NH_2	Primary aliphatic amines
8.	618.074	- C ≡ C –H :C –H bend	Alkynes

3. SEM AND EDAX ANALYSIS:

The surface morphology and size of the nanoparticles were observed by Scanning Electron Microscopy (SEM) analysis. The typical SEM image shows that the product mainly consists of particles like copper nanoclusters with panoramic view and the size ranges from 150-200 nm. The spherical shape of the nanoparticles is shown in the SEM images (Fig.4 A). The quantitative and qualitative analysis of elements may be concern in the formation of copper nanoparticles and were identified by EDX analysis (Fig. 4 B). Due to the Surface Plasmon Resonance, the copper nanoparticles show the absorption peaks of higher counts.

Fig 4: (A) SEM Micrograph of CuNPs and (B) EDX analysis of CuNPs



(A) (B)

DISCUSSION:

In the present study, synthesis of copper nanoparticles using aqueous leaf extract *Ocimum sanctum* belonging to family *Lamiaceae*, without any harmful chemicals. The green synthesis of copper (Cu NPs) are subjected to UV-spectroscopy analysis and obtained a single peak but broad at 589 nm and are in conformity with the eco friendly biosynthesis of copper nanoparticles using flower extract of *Aloe Vera* (Javad Karimi *et al*., 2013). FTIR spectra revealed that three was thin film of proteins and some secondary metabolites that covered the metal nanoparticles. The studies are in conformity with the biosynthesis of copper nanoparticles using *Capparis zeylanica* (Saranyaadevi *et al*., 2014) and *Nerioum olender* leaf extract (Gopinath *et al.,* 2014). SEM image revealed that the particles appear to almost spherical in shape and are in conformity with the synthesis and characterization copper nanoparticles using papaya leaf extract (Suresh *et al.,*2014) and chemical synthesis and characterization of copper nanoparticles (Muhammad Sani Usman *et al*., 2013). To find out the purity of the metal particles synthesized EDX spectrum was obtained which showed along with copper, there were other elements. Thus the aqueous leaf extract of *Ocimum sanctum* is found to be a potential source of bio-reducant to reduce metal salts in to their nanoparticles (Jayalakshmi et al., (2014) .The study successfully demonstrates the convenient utilization of Ocimum *sanctum* leaf extract for the synthesis of copper nanoparticles

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