



COMPARISON AND BIOSYNTHESIS OF COPPER NANOPARTICLES USING AEGLE MARMELLOS AND MORINGA OLEIFERA LEAF EXTRACT

V.Sumathi ^{1*}, A.Padmapriya², T.Umarani³, K.Vinothini⁴

¹Assistant Professor, Department of PG Physics, SRNM College, Sattur, Tamilnadu. India.

^{2,3,4} II M.SC., Physics, Department of PG Physics, SRNM College, Sattur, Tamilnadu. India.

Abstract:

The present study involves the green & eco-friendly synthesis of CuNp's using the leaves extract. The Biosynthesis was done by using the aqueous solution of leaves extract and copper sulphate solution. A fixed ratio of plant extract to metal ion was prepared and the colour change was observed which provided the formation of Cu nps. The synthesized CuNp's were characterized by XRD, UV, SEM, FTIR & Antibacterial activity. The CuNp's formed were confirmed by the characteristics surface Plasmon Resonance (SPR) peak found at 436nm in UV-visible spectra. The morphological study from SEM images, it is confirmed that spherical shape Cu nano particles settled by the leaves extract residue. FTIR spectrum clearly illustrates the green synthesis of Cu nanoparticles mediated by the leaves extract and also specified some functional group responsible for the reduction of copper sulphate to form copper nanoparticles and the capping agent present in the leaves extract. From XRD result, we conclude that the particle size was calculated at 3nm and 60.36 nm for two samples respectively. The antibacterial activities were taken as to gram positive as well as gram negative bacteria's.

Keywords: Bio synthesis, XRD, FTIR, UV, SEM, EDX and Anti bacterial activity.

1. Introduction:

Nanotechnology is one of the most active areas of research in modern material science and technology. To synthesize nanoparticles there are many ways such as sol-gel method, chemical reaction and co-precipitation. Compared to those methods bio synthesis method is one of the best method for the production of nanoparticles in recent years. Plant mediated synthesis of nano particles is a green chemistry approach that connects nano technology with plants. The root word 'bio' means life. This bio synthesis method have several advantages over other methods namely cost effectiveness, simplicity, use of less temperature, the usage of less toxic materials, moreover it is compatible for medical and food applications [1-3]. In recent years, Synthesis of metal Nanoparticles using plant leaf extract has attracted attention of many researchers because of availability of



materials, inexpensive and processes easy to carry out in any laboratory, use of non-toxic reagent. In recent years, Cu nano particles have attracted much attention of researchers due to its application in industrial and medical applications. The biological property shown by Cu nanoparticles are wound dressings and biological properties [4-5], antibacterial, industrial use such as gas sensors, catalytic process, high temperature superconductors and solar cells [6-8].

Aegle Marmelos is commonly known as Bael or Bilva, which is a historic plant of India. Aegle Marmelos belongs to Rutaceae family and grown as a temple garden plant and leaves are used to pray Lord Shiva. Aegle Marmelos is an important medicinal plant with several medicinal applications in traditional and folk medicine systems. Traditionally, Aegle Marmelos is used in the treatment of diarrhea [2]. Moringa oleifera has an impressive range of medicinal uses with high nutritional value and medicinal benefits. The various parts of Moringa oleifera contains Seed pods, Flowers, Root, Bark are used in various medicinal preparations [3].

In this work, we report the biosynthesis of Cu nanoparticles from the leaves of Aegle Marmelos and moringa oleifera.

2. Experimental Methods:

2.1. Plant name:



Aegle marmelos



Moringa Oleifera

2.2. Preparation of the leaf extract:

Leaves were collected and used to prepare the aqueous extract. The required amount of leaves were collected and thoroughly washed 3 and 4 times using normal water and distilled water to remove impurity. The cleaned leaves were dried under sunshade to remove moisture completely. Then the dried leaves were gushed by adding drop of distilled water. After crushing, the leaf extract was filtered out. Then leaf extract were taken into a measuring jar of about 10ml. This extract was used to further work.

2.3. Preparation of Cu np's:

Copper sulphate and distilled water were purchased and used with further purification. In this work, a 0.1 M of copper sulphate with 100 ml distilled water was kept under constant stirring using magnetic stirrer to completely dissolve the copper sulphate for two hours. After complete dissolution of copper sulphate, 10 ml



leaves extract solution was added under high speed constant stirring, drop by drop touching the walls of the vessel. The reaction was allowed to proceed for 2 hour after complete addition of leaf extract. The beaker was sealed at this condition for 4 hours. After the completion of the reaction, the solution was allowed to settle for overnight and further the supernatant solution was separated carefully. Then dried in room temperature the copper nano particle was formed. The collected nano particle were synthesized by XRD, FTIR, SEM , EDX ,UV Analysis and antimicrobial activity.



3. Characterization techniques:

XRD patterns of the samples were investigated using X ray diffractometer (X PERT PRO measurement system) to study the crystalline nature . Cu K α radiation of wavelength $\lambda = 1.5406 \text{ \AA}$ was used as a diffraction source and with the step size of the of 0.05° from the diffraction range $10 - 80^\circ$. The morphology studies of samples were studied by Scanning electron microscope (SEM) with an operating voltage 20 kV with the magnification range 7500- 30,000. The FTIR spectra of the prepared samples were recorded in the range $400-4000 \text{ cm}^{-1}$ using a Shimadzu FTIR spectrometer. The antibacterial study of green synthesized copper nano particles were established against both gram negative and gram positive pathogenic bacteria such as serratia marcescens (gram negative), Escherichia coli (gram negative), pseudomonas aeruginosa (gram negative) , methicillin resistant staphylococcus aureus (MRSA) (gram positive) using agarwell method.

3.1 XRD Studies:

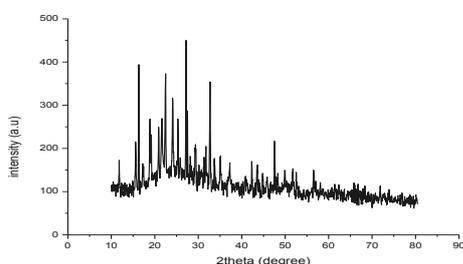


Fig: 1

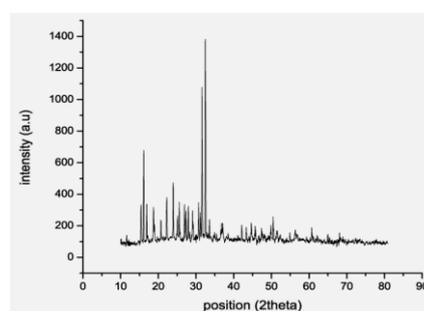


Fig: 2

XRD spectra of Green copper nanoparticles



Fig1 shows the XRD pattern of copper nanoparticles. The crystalline nature copper nanoparticle synthesized from aegle marmelos has been studied by XRD analysis. XRD spectrum of copper nanoparticles using Aegle marmelos leaf extract shows distinct diffraction peaks around 32.5 degree which are indexed by the (100) of the cubic face centered copper. There sharp Bragg peaks might have resulted due to crystalline nature of copper nanoparticles. Using Debye scherrer's formula the grain size of the synthesized copper nanoparticles is calculated and its value is 3 nm.

Fig 2 shows the XRD pattern of copper nanoparticles using Moringa Oleifera leaf extract shows distinct diffraction peaks around 27.1961 degree which are indexed by the (100) of the cubic face centered copper. There sharp Bragg peaks might have resulted due to crystalline nature of copper nanoparticles and its grain size of the green synthesized copper nanoparticle has the value of 60.36 nm.

3.2 FTIR analysis:

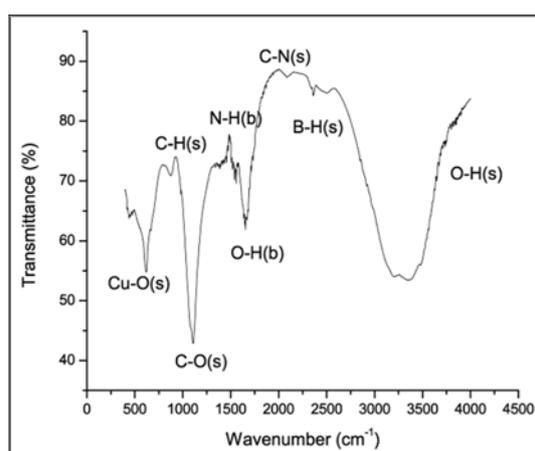


Fig: 3

FTIR spectra of Green copper nanoparticles

Fig3 shows that the FTIR spectrum revealed that the absorption bands at above 3819.06cm⁻¹ are may be responsible for the presence of O-H stretching vibrations. A band at 2360.87cm⁻¹ is due to B-H stretching vibrations. The vibration band at 2085.05cm⁻¹ is due to C-N stretching vibrations. The band at 1651.07cm⁻¹ is due to O-H stretching vibrations. The vibration band at 1519.91cm⁻¹ is due to N-H stretching vibrations. The vibration band at 1107.14cm⁻¹ is due to C-O stretching vibration. The vibration band at 873.75cm⁻¹ is due to C- H vibrations.



3.3 UV-Visible spectroscopy analysis:

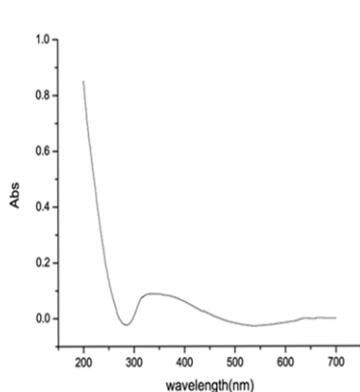
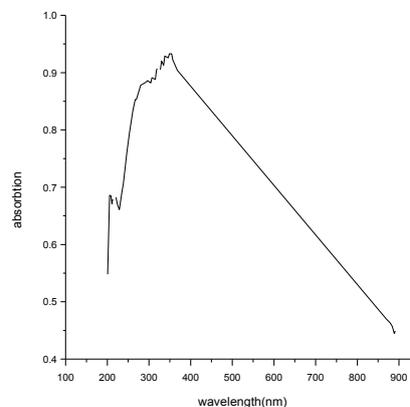


Fig: 4



Figs: 5

UV spectra of Green copper nanoparticles

Fig 4 and 5 shows the absorption spectrum of green copper nanoparticles. During exposure to plant extracts was observed as a result of the blue colour to dark green colour change. The colour change is due to the surface plasma resonance phenomenon. The metal nanoparticles have free electrons which gives the SPR absorption band due to the combination vibration of electrons of metal nanoparticles in resonance with light wave. The sharp bands of copper nanoparticles were observed around 439 nm and band gap of this sample is 2.849eV in case of Aegel marmelos. The Cu nanoparticles using Moringa leaf extract formation was confirmed from the peak at 352nm .The peak value was found to be gradually decreased with increase in wavelength. And the calculated band gap of prepared sample is 3.5601 eV in case of Moringa Oleifera. That is very higher than reported value of 2.41eV to 2.61eV in copper bulk material. The increase in the bandgap of the copper nanoparticles with decrease in particle size may be due to the quantum confinement effect.

3.4 SEM Analysis:

Fig 5 and 6 shows the SEM image of green copper nanoparticles. It exhibits that almost all the nanoparticles are of spherical shape.

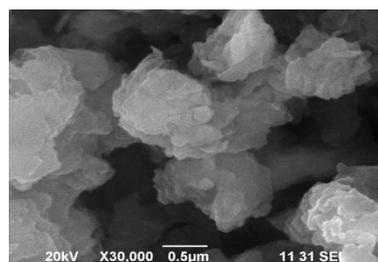
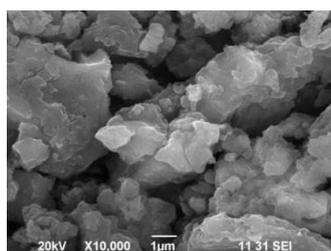




Fig: 5

Fig: 6

SEM image of Green copper nanoparticles

3.5 EDX-Analysis:

The quantitative and qualitative analysis of element may be concerned in the formation of the Cu nanoparticle. The presence of Copper nanoparticle was confirmed by EDX analysis. EDX can be used to determine which chemical elements are present in a sample and can be used to estimate their relative abundance. The accuracy of this quantitative analysis of small composition is affected by various factors. Many elements will have overlapping X-ray emission peaks (eg.O,K,Si,S). The accuracy of the measured composition is also affected by the nature of the sample. But, the Cu peak must be very high. Fig 7 shows that the Energy dispersive spectrum (EDX-image) of bio synthesized copper nanoparticle.

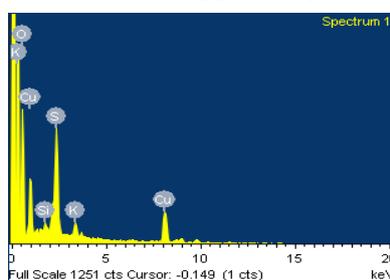


Fig: 7 EDX images of green copper nanoparticles

3.6 Antimicrobial study on green copper nanoparticles:

The antibacterial study on bio synthesized copper nano particles were established against both gram negative and gram positive pathogenic bacteria using agar well method as shown in Fig 8. The zone of inhibition was observed to be more in gram negative bacteria when compared to gram positive bacteria. The maximum ZOI values is observed as 15mm in serratia marcescens bacteria for 100 µl concentration of bilva leaf extract copper nanoparticles shown in table. The zone of inhibition values observed by pseudomonus aeruginosa were found to be 8mm for Moringa leaf extract nano particles. . Bilva leaf extract nanoparticles has small in size compared to Moringa extract nanoparticles. so the smaller particle size contributes to more effective interaction with microbes.

Microbes	Zone of Inhibition(mm) for	
	Bilva leaf extract nps	moringa leaf extract nps
	100 µl	100 µl
MRSA	11 mm	7mm
E-Coli	14 mm	8mm



<i>pseudomonus aeruginosa</i>	13 mm	8mm
<i>serratia marcescens</i>	15 mm	8mm

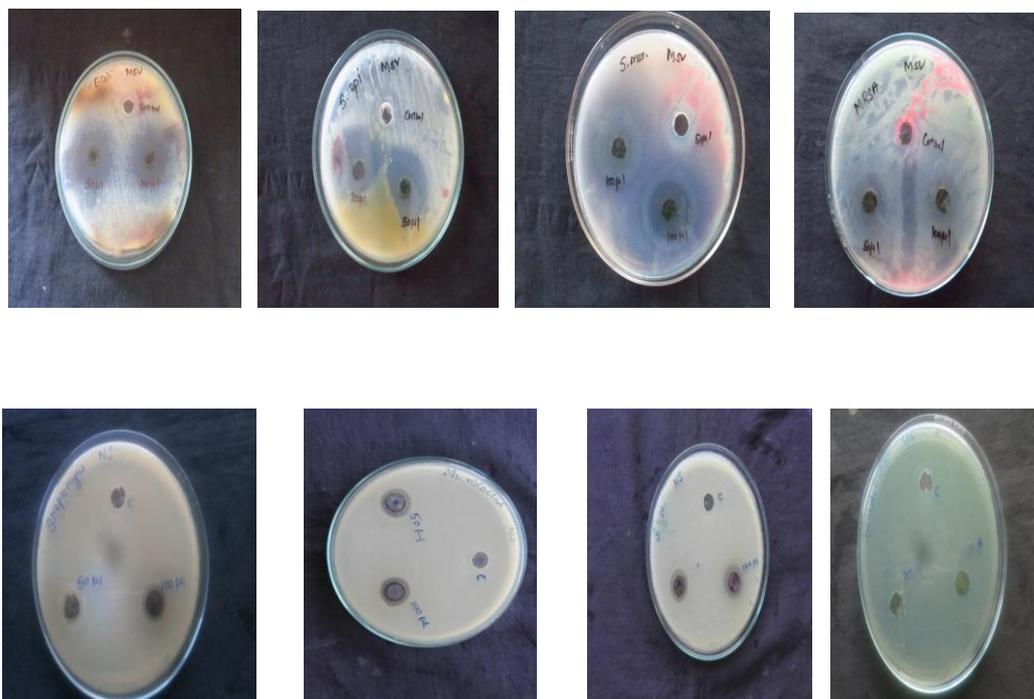


Fig: 8 Zone of Inhibition (ZOI)

4. Conclusion:

Copper nanoparticle was successfully synthesized by biological method using *Aegle marmelos* and *Moringa olifera* leaves extract. The surface Plasmon resonance property of synthesized nanoparticle was studied by UV Spectroscopy and the red shift of the spectra was found to be at 439nm and 362 nm. The morphological study of Cu np using SEM suggests that the nps are spherical in shape. EDX spectrum was confirmed the presence of copper nanoparticles. FTIR spectrum concludes some functional group present in the prepared samples. XRD conclude that the nps peaks around in the angle of diffraction $2\theta=32.51^\circ$ and 27.19° . The particle size was calculated at nanometer range. The antibacterial activity of cu nps concludes that the cu nps shows significant anti bacterial activity against *pseudomonus aeruginosa*, *serratia marcescens*, *E-coli* and methicillin resistant *staphylococcus aureus* (MRSA) bacteria and also conclude the smaller particle size contributes to more effective interaction with microbes.



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