Pharmaceutical Breakthroughs
Volume 1, Issue 1, 37-49
DOI: 10.59762/pb724290251120231110131524

Synthesis And Characterisation of Copper Nanoparticles Using Aqueous Leaf Extract of Lagerstreomia Speciose and Their Biological, Antioxidant and catalytic activities.

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ABSTRACT:
Nanotechnology is gaining importance at tremendous rate in every field of science and technology. Researchers have developed a large number of different methods for the synthesis of nanoparticles; among this green route method is of great importance as it is eco-friendly and cost-effective. In this study we synthesized copper nanoparticles through green synthesis using a plant called lagerstroemia speciose. The formation of copper nanoparticles has been observed due to change in colour of extract solution. The sample solution was characterized by different techniques in order to find out the various physiological parameters like shape, size and nature of bonding. Plant extract acts as both reducing and capping agent .The various instruments used for characterization are DLS (Dynamic Light Scattering), SEM (Scanning Electron Microscopy), X-ray Diffractometry (XRD) and Fourier Transform-Infrared (FT-IR). Cups obtained were of size 35nm. The synthesized copper nanoparticles were studied for their anti-microbial activity, antioxidant, antidiabetic and catalytic activities and cytotoxicity.

KEYWORDS: Copper nanoparticles, UV-Visible spectrophotometer, Dynamic Light Scattering (DLS), Scanning Electron Microscopy (SEM), Fourier Transformer-Infra red (FT-IR), inhibition activity, catalytic activity.

INTRODUCTION
The branches of science and technology which are modern and very useful fast growing are Nanoscience and Nanotechnology. These deal with fabrication, characterization and applications. Metallic and non-metallic nanostructured materials of different compositions, sizes, and shapes that are playing an ever increasing role in day to day life Lengke, Maggy F et al, (2007) have been brought into the market as nano-products. Nanotechnology is a growing field in biomedicine as well Prabhu N et al, (2010). The controlled fabrication of nanoparticles has propelled nanotechnology into one of today's most promising and popular fields of scientific research Shenmar, R et al, (2005). Potential future advancement requires the ability to prepare nanomaterials in a reproducible and controlled manner Balzani V et al, (2002). Generally metal and metal oxide nanoparticles were routinely synthesized by various physical and chemical methods. Frequently used synthetic methods are sol-gel technique solvothermal, reduction, non-sputtering, and electrochemical technique Shokuhi R and M. Tripathi et al, (2010). But these methods are toxic, expensive, and potentially threatening, not to mention the difficult separation procedure, energy requirement and high pressure.

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Copper nanoparticles (CuNPs) have found multiple applications i.e in the field of medicine, biosensors, electronic devices, lubricants, reagents in various reactions, and biotechnology Sadowski Z et al, (2000) . Further, copper has also been used as anti-biotic, anti-fungal agents in the treatment of wounds Chatterjee A.K, et al, (2014). Various attempts have been made for the synthesis of CuNPs by the methods of laser ablation, gamma irradiation, electron irradiation, thermal decomposition of metal compounds, chemical reduction, photochemical methods, microwave processing, and biological methods Din M.I, Rehan et al,(2016) . The main disadvantage of these approaches is the use of lethal and costly chemicals. The need of hour is to investigate a new route of synthesis, which should be cost-effective and eco-friendly. Therefore, more and more attempts are being made to bio-synthesize of CuNPs by employing different plant extracts and this process is found to be simple, easy, low cost and eco-friendly and is known as green technique for the production of CuNPs in bulk Saranyaadevi K et al, (2014). The main mechanism considered in such synthesis of nanoparticles is the role of phytochemicals present in the extract which act as the reducing agents to reduce the aqueous metal ions. The phytochemicals that mainly cause the reduction of metal ions are flavonoids, terpenoids, carboxylic acids, quinones, aldehydes, ketones and amides Prabhu, P et al, (2012). The role of plants in the synthesis of nanoparticles is currently under consideration. The previous studies show that the many researchers have synthesized metal nanoparticles from the leaves extracts of the plants like Butea monosperma (Flame of Forest, Palas) Chaturvedi et al, (2015), Piper longum Jacob S et al, (2012), Nerium oleander (Red kanher) Gopinath M et al, (2014) Ocimum sanctum Kulkarni V.D et al, (2013), Tea leaf Vaseeharan B et al, (2010), Glycine Max (Soybean) Vivekanandhan S et al, (2009), Aloevera plant Vivekanandhan S et al, (2006) etc. In the present study the synthesis of CuNPs from the fruit extracts of plant lagerstroemia speciose was selected due to its potential medicinal value. Lagerstroemia speciosa is generally known as crape myrtle as it belongs to the Lythraceae family. This tree is extensively dispersed in Philippines, Malaysia and India. The leaves of the banabá and other parts are commonly used as a tea preparation in the Philippines, Japan and Taiwan. In Vietnam the plant's immature leaves are eaten as vegetables, and its mature leaves and fruits are consumed in medicine for glucose reduction in blood Tanaka et al, (2007). Folk medicines have customarily been prepared from roots, bark and leaves of Lagerstroemia Speciosa and are medication for many diseases and weaknss. It is used as an effective anti-diabetic drug because it contains high content of corosolic acid. Banaba is also recommended for kidney, hypertension and bladder problems. Leaves of the species have been used over thousands of years as folkloric treatment by the native Indians and Japanese for illness, ailments particularly for lowering blood sugar levels and weight loss. The flower extracts of the species has some pharmacological properties like antioxidant and anti-microbial activities, whereas fruit extracts reported antinociceptive, anti-diarrhoea and cytotoxic activities. Research on leaf extracts reveals that anti-bacterial, anti-viral, anti-inflammatory, anti-obesity, anti-diabetic and xanthine oxidase inhibition, diuretic, decongestant activities and roots are applied for treating mouth ulcers. In addition to that bark is used to relieve the abdominal pains Koduru RL et al, (2017). The species also has essential metals like potassium, sodium, magnesium and iron which were clinically proved Saraswathi VS et al, (2011).

Chemical Constituents: The chemical constituents Guang-Hui H et al, (2013) Lagerstreomia Speciosa include corosoli acid Katta VK et al, (2006), lageracetal, amyl alcohol, ellagic acid, gallic acid, 4- hydroxyl benzoic acid, beta sitosterol, 3,3,4-tri-O-methl ellagic acid, 3-O-methyl-3,4-methylene-dioxy ellagic acid, Asiatic acid, alphitolic acid, 3,3,1 –di-O- methyl ellagic acid, 3,4,3,1 ,41 - tetra-O- methyl flavellagic acid, 31 , 41 -di-O-methl-3,4- metheledioxy flavellagic acid, 3-O-methyl ellagic acid, 6,7-dihydroxy coumarin, alanine, isoleucine, alpha amino butyric acid, ellagitannin Klein, Guy et al, (2007) and methionine. In the present study, we used leaves of lagerstroemia plant for the synthesis of copper nanoparticles and their different applications.

Materials and method

Preparation of leaf broth of Lagerstreomia specio L. plant

For the preparation of extract, 10g of dried powder was mixed in 100ml of Milli-Q in a 250ml Erlenmeyer flask and heated at 70°C for half an hour. The mixture was cooled and centrifuged at 13000rpm for 20mins in order to collect pure extract of leaves. The pure extract of leaves was then transferred into glass bottle. The bottle was sealed properly and labelled as "LGL Leaf Extract" and then stored at refrigerated temperature (4°C).

Preparation of copper sulphate solution

Copper sulphate solution (10mM) was prepared by adding 0.0249g of copper sulphate to a beaker containing 20ml of distilled water. The mixture was stirred for complete dissolution of copper sulphate.

Synthesis of copper nanoparticles

For the synthesis of Copper nanoparticles , 1ml Lagerstreomia aqueous leaves extract, maintained at PH 10, was taken in glass beaker which was placed on magnetic stirrer with hot plate at constant rpm 700 and temperature 70°C. 5ml of 0.01M CuSO4 were added dropwise through burrette to the beaker containing leaf extract. After complete addition of copper sulphate solution the the beaker containing mixture is removed from magnetic stirrer and kept at room temperature for overnight for the completion of reaction, which was indicated by the colour change of mixture. The mixture was then subjected to different characterisation techniques.

Characterisation

The following spectrophotometric techniques such as Uv-visible spectrophotometer analysis using nano spectrostar (BMG LAB TECH) ii) Dynamic Light Scaterring (Malvern) iii) scanning electron microscope (SEM) analysis using a JFC1600 instrument (JEOL, Ltd., Tokyo, Japan) equipped with an energy-dispersive X-ray (EDX) attachment; and iii) fourier transform infrared spectroscopy (FTIR) analysis using a Spectrum One FTIR spectrophotometer (PerkinElmer, Inc.) and X-ray diffractometry were used for investigating the morphology, elemental composition, crystalline nature, functional group and stability of synthesized CuNPs.

Encapsulation Efficiency

Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES)

Encapsulation is the incorporation of materials of different or same nature to provide the formulation of required characteristics. Inductive coupling plasma with optic emission spectrometry technic (ICP-OES) was used to determine the metal load efficiency in the mother solution of nanoparticle. It was performed with atomic emission spectrophotometer ICP-OES Varian 725-ES using argon as carrier gas. The samples were ashed at 550°c in furnace. The ash was collected and dissolved in 0.5M nitric acid and then solution was filtered by using ashless filter paper. Volume was made upto 50ml. This volume of sample was then injected into ICP-OES . The results are expressed as mg of copper per gram of lipid. The encapsulation efficiency percentage is calculated by following formula

$$\% EE = Ci \div C0$$

Where, Ci is the real concentration calculated by ICP. and C0 is theoretical concentration of product involved into the process.

Antibacterial activity
Well Diffusion Method

Antibacterial assay of the Lagerstroemia Specio L. leaf extract and freshly prepared silver nanoparticles (CuNPs) was performed using agar well-diffusion procedure against 3 pathogenic bacteria Rios J et al, (1988). Staphylococcus aureus, Bacillus cereus and Eschirichia.coli. Nutrient agar and nutrient broth media were used in bacterial culturing. An overnight culture (107 CFU/mL) and freshly prepared nutrient agar medium were mixed at 45°C and poured into the sterilized Petri plates. To solidify the culture, Petri plates were set aside at room temperature in the laminar flow. 8mm wells were made using sterilized micropipette tips of 1 mL, and a sterilized needle was used to take out the agar plug. About 100µL, 150µL Cuso4 standard, Lagerstroemia Leaf extract, Copper nanoparticles and Control (Nutrient broth) were poured into the prepared wells and then these Petri dishes were incubated at 37°C for 24 h. The bacterial growth reduction was measured through the diameter of the zone of inhibition in millimeter (mm) after 24hours Seeley HW et al, (2001). The diameter was obtained by measuring the clear zones around each well by using scale Hammer KA et al, (1999).

Anti-diabetic activity
Alpha-Amylase Assay

A-amylase inhibition was studied by the method Malik CP and Singh MB (1980) with little changes. Alpha amylase (.5mg/ml) was prepared and 20µl of which was added to each test tube containing various concentrations i.e. 20µl, 40µl, 60µl of test samples (Lagerstroemia Specio L. leaf extract, copper nanoparticles). 10µl of 0.02m phosphate buffer (ph 6.9) was then added to each test tubes and the mixture was incubated for 10minutes. The incubated mixture was then added with 1ml of 1%starch solution and incubated again for 20minutes. Lastly 400µl of DNS reagent was added to halt the reaction. Finally Whole reaction mixture was boiled for 5minutes. The colour change was observed from yellow to orange which indicated α-amylase inhibition activity. The tubes were left to cool, and the absorbance was measured at 540 nm. Control was prepared wherein amylase was not added. The percentage inhibition of αamylase was calculated as

$$\% = [(Ao - Ai)/Ao] \times 100$$

Where
Ao was the absorbance of the standard and Ai was the absorbance of the test samples

In vitro cytotoxicity by MTT assay
Cell lines and culture medium

MCF-7 cancer cell line was bought from the national centre for cell sciences (NCCS), Pune, India. Growth medium with 10% fetal bovine serum (#RM10432, hi media) was used to culture the cancer cells and incubated at a temperature of 37±0.2 °C. MTT reagent was bought from hi media. The assay used to verify the cytotoxic activity of sample against MCF-7 was MTT(3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyltetrazoliumbromide) assay. This assay involves the reduction of the NAD(P) H dependent oxidoreductase enzymes of viable cells on yellow colored water-soluble tetrazolium dye to give a formazan product with deep purple color Mosmann T (1983). The cytotoxicity assay was carried out by seeding 100 µl cell suspension in a 96-well plate at required cell density per well and incubated for 24 h at 37 °C in 5% Co2. Exhausted media was replaced after the incubation period by new media containing different concentrations (15, 30, 50 and 90 µg/ml) of test agent (copper nanoparticles) and incubated for 24 h at 37 °C. Cells in media containing 10% FBS with nothing added were used as controls. After the incubation, the media containing test samples were removed followed by addition of MTT reagent and incubated again. MTT reagent was separated after incubation, and then 50 µl solubilized solution of dimethyl sulfoxide (DMSO) was added to the obtained purple formazan crystals and the absorbance was measured at 570 nm using a spectrophotometer Uma Suganyaa KS et al, (2016). Cell viability and cytotoxicity percentage were calculated using the following equation El-Naggar NEA and Theivasanthi T et al, (2013)

$$\text{Cell Viability } \% = \text{TestOD} \div \text{Control OD} \times 100 \quad \ldots\ldots\ldots\ldots\ldots (1)$$

$$\text{Cytotoxicity } \% = 100 - \text{Viability } \% \quad \ldots\ldots\ldots\ldots (2)$$

Antioxidant Activity
DPPH Assay

The antioxidant assay of the Lagerstroemia Specio L. leaf extract and its green-synthesized copper nanoparticles (GCuNPs) was determined using the method stated by BrandWilliams et al. Brand-Williams W et al, (1995), which is to calculate scavenging activity of the DPPH (1,1-diphenyl-2-picrylhyorazyl) free radical. 0.5 mM DPPH radical solution was prepared by mixing DPPH in methanol and stored in dark for 30 min. Reaction mixture was prepared by adding 20 µL, 40 µL, 60 µL of the Lgl extract and freshly prepared FGCuNPs into 100 µL of 0.5 mM DPPH radical solution. 3 mL of methanol was then added and left for half an hour. The color changed was observed from deep violet to light violet which is indication of antioxidant activity of prepared sample. DPPH gets reduced when reacts with an antioxidant agent that gives hydrogen. Absorbance (Abs.) of the reaction mixture was measured utilizing a UV-Vis spectrophotometer at 517 nm. Methanol (3 mL) dissolved in DPPH radical solution (0.3 mL) was used as the control solution. The level of percentage inhibition by the extract and green-synthesized AgNPs was calculated according to the subsequent formula:

$$\% \text{ Inhibition} = Ac - As/Ac \times 100$$

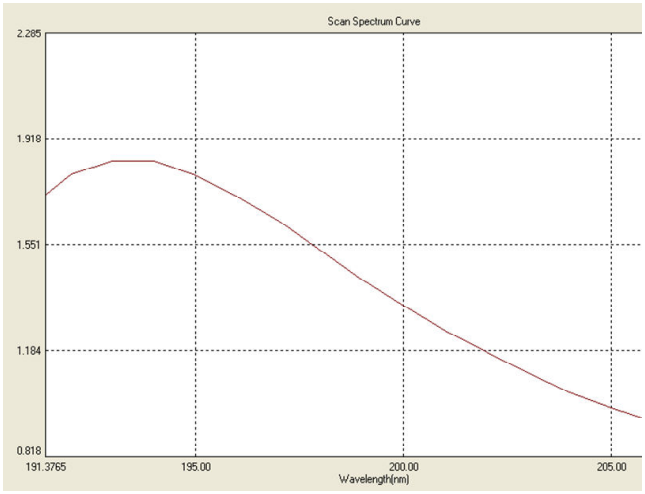
Catalytic reduction
The synthesized metal nanoparticles were used

as catalyst for reduction of nitro compounds and dyes by sodium borohydride Pradhan, N et al, (2002). 0.2M sodium boro-hydride was freshly prepared of which 1ml was taken in a cuvette. Add 1.9ml of 0.2Mm of dye to the cuvette containing sodium boro-hydride. Shake the Cuvette and keep it in the uv-visible spectrophotometer to record the absorbance. Remove the cuvette and add 0.1ml of test sample and shake it vigorously and keep it again in Uv-visible spectrophotometer to record absorbance.

Result and Discussion

UV-VIS Spectroscopic analysis

Copper nanoparticles prepared from lagerstroemia plant (aqueous leaf extract) were confirmed by subjecting the solution mixture to uv-visible spectrophotometer which gives the absorption peak. The peak was observed at a wavelength of 193nm which is similar to the previous report Aher h et al, (2019) confirms the formation of copper nanoparticles. The formation of peak is due to surface plasma resonance of copper nanoparticles shown in (fig.1). The change in colour of solution from black-red to brown is first visual indication of formation of copper nanoparticles.



Maxima at 193nm

Figure. 1
Uv-visible spectrum of Copper nanoparticles

Particle size distribution

Particles size curve of Dynamic Light Scattering (DLS) revealed that the average size of copper nanoparticles present within the sample solution is 35nm.Two peaks of copper nanoparticles were observed with intensities of 98.7% and 1.3%. Poly-distribution index (Pdi) was (0.259) (fig.2) which shows that there is very low agglomeration and variation in particle size Lizunova, A.A et al, (2017). The larger number of values is perhaps due to hydrodynamical shell. The size (radius of hydrodynamic shell is calculated from Stokes-Einstein equation i.e. D= kBT/ 6ππηRh) depends on particle shape, structure and roughness Kätzel, U et al, (2008).

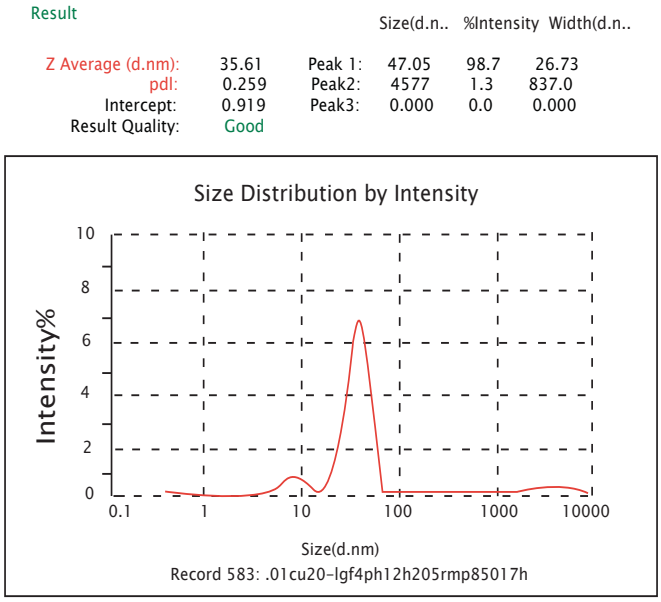


Figure. 2
DLS of average particle size of Copper nanoparticles

Zeta potential gives insight on surface charge and stability of CuNPs. In this study it was found that the capping agents present on the surface of the green synthesized Cu NPs are negatively charged groups which are responsible for the stability of the nanoparticles. Zeta potential of copper nanoparticles was found -28.3 mv (fig.3) which means that the capping agents are negatively charged groups. Hence, it is concluded that Copper nanoparticles have good stability which is indication of presence of large number of adsorption sites on copper nanoparticles similar to previous study Azizi S and Barzinjy, A et al, (2020) depends on surface charge of particles i.e. Zeta potential.

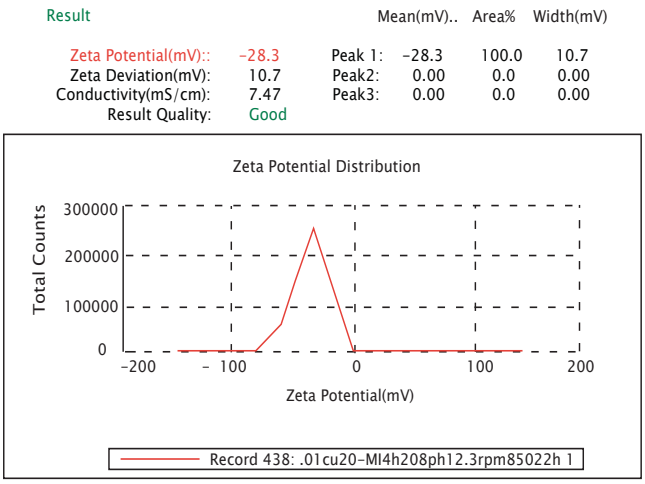


Figure. 3
DLS of Zeta Potential of Copper nanoparticles

Scanning Electron microscopy (SEM)

The size and shape of the copper nanoparticles was examined by scanning electron microscopy (SEM). It was found that shape of particles is spherical with particle size within nanometres range i.e. below 50nm (fig.4). The particles of different size were present in sample. Agglomeration is present which is usual with the green synthesis nanoparticles. It is because of possession of higher surface area and the durable affinity by green synthesized nanoparticles that agglomeration or aggregation Sundrarajan M and Agarwal, H et al, (2017). Fig. shows the scanning electron microscopy of copper nanoparticles synthesized by the plant extract of Lagerstroemia obtained by bio-reduction method.

Fourier Transform – infrared (FT-IR)

FTIR investigation was used to identify the biomolecules which are responsible for reducing, capping and stabilizing agents in the formation of copper nanoparticles with Lagerstroemia Specio L. The absorption bands at 3380, 1633, and 1099 (fig.5) corresponds to the O-H stretching of alcohol and phenol, and also to the presence of amines N–H of amide, C-H stretching of aliphatic group, C=O stretching of ester carbonyl, C=C stretching of the aromatic ring, and C-O stretching of ester, respectively Pradhan, S and Fatma, S and Saranyaadevi, K et al, (2014). The peaks obtained from FT-IR analysis of copper nanoparticles indicates the presence of biomolecules like polyphenols, terpenoids, flavonoids, alkaloids which are responsible for reduction of copper ions to copper nanoparticles because of their ability to act as reducing, capping and stabilising agents Kalainila P and Sahu, K.P et al, (2013).

X-ray diffraction analysis

The crystallinity and phase structure of the nanoparticles were determined by its typical powder XRD diffraction patterns. 2θ values 32°,35°, 38°,46° where peaks are observed corresponds to (110), (111), (202) and (020) planes of CuNp (fig.6) The diffraction peaks are in good agreement with those of the standard JCPDS card no. 04-0836 for the standard spectrum of the pure face centered cubic (FCC) CuNPs. Besides, copper nanoparticle peaks, other peakes are observed which may be due to presence of copper oxide nanoparticles. Agglomeration was

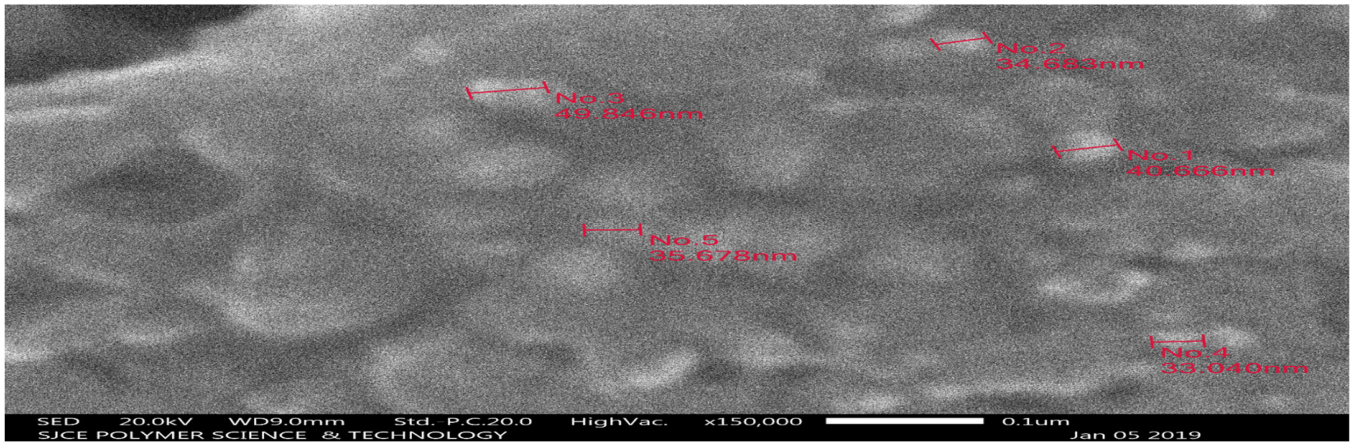


Figure. 4
SEM image of Copper nanoparticles

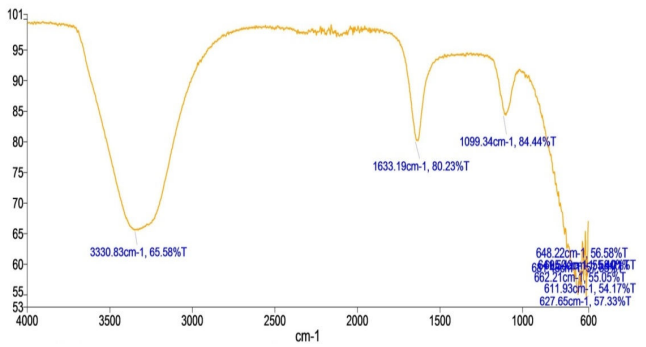


Figure. 5
FT-IR Spectrum of copper nanoparticles

also observed which may be due presence of other particles S. Yallappa and M. Taran et al, (2017).

Encapsulation Efficiency

The ICP-OES spectroscopy was used to determine the Cu ion concentration encapsulated by phytochemicals of plants extract in the solution of CuNPs. Cu content in the solution was determined by ICP-OES equivalent to 1.4g which accounts for 70% of total amount of nanoparticles. Hence, it confirms the formation of high yield of copper nanoparticles.

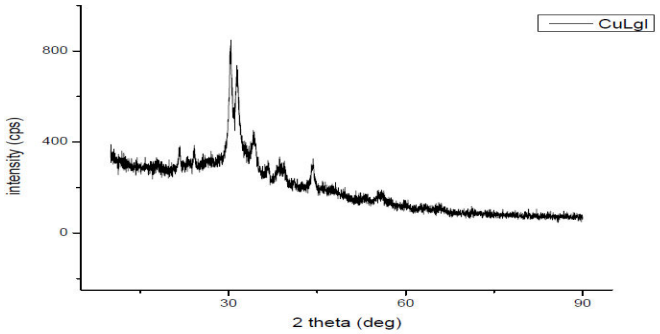


Figure. 6
XRD pattern of synthesized Cu NPs

Anti-bacterial Activity

Antimicrobial activity was done by well diffusion assay against different pathogens like Staphylococcus aureus, Bacillus cereus, and E.coli. 101 diluted culture (106 CFU) and 100µl and 150µl sample were used for the experiment. Well diameter was 8mm. It was observed that copper nanoparticles showed zone of inhibition (table.l) in S. aureus, Bacillus cereus and E.coli. It was also observed that inhibition rate in

Samples	Concentration (µl)	Zone of inhibition (mm)		
		S. aureus	B. cereus	E.coli
Control (Nutrient broth)		No inhibition	No inhibition	No inhibition
Cuso4 standard	100	13	10	11
	150	16	12	14
Lagerstroemia Leaf extract	100	11	09	12
	150	13	11	12
Copper nano particles	100	18	11	15
	150	22	13	18

Table. 1
Concentration versus zone of inhibition of test samples against pathogens

crease with the increase in concentration M. Valodkar, et al, (2011).The reason for being inactive to other pathogens may be the dose concentration and pH of copper nanoparticles.

Anti-diabetic Activity

Copper nanoparticles synthesized through green route showed better anti-diabetic activity while plant leaf extract showed moderate inhibition. The study has confirmed that with the increase in the concentration of Copper nanoparticles the activity of αamylase was more efficiently inhibited (table.2). The Lagerstroemia leaf extract made Copper nanoparticles reduced the amylase level by hydrolysing complex carbohydrates into lower carbohydrates to increase the utilisation of glucose Rajaram K and Abideen S et al, (2015). Our results are in agreement with the previous studies. According to various in vitro studies, for diabetes care, alpha-amylase inhibition is supposed to be one of the most operative methods Balan K and Hamid HA et al, (2015).

Concentration (µl)	Control %	Lagerstroemia leaf %	Copper nanoparticles %
20	0	52	65
40	0	61	70
60	0	67	80

Table. 2
Concentration versus inhibition percentage (%) of test samples

In vitro anticancer activity

MTT Assay and Cell Morphology

In this study, the MTT assay was used to estimate the anticancer capability of CuNPs on breast cancer cell line (MCF-7). From the experiment; it was observed that the cytotoxicity against breast cancer cell line increases with

increasing concentration of CuNPs. The IC50 of CuNPs was found at 95µg/ml against the MCF-7 cancer cell lines. It was determined that 50% inhibition of cells was observed compared to untreated control. The proliferation of MCF-7 cancer cell line exposed to CuNPs was considerably inhibited in a dose-dependent manner(fig.7). The cytotoxic effect of synthesized CuNPs on cancer cells was investigated by visual examination of the morphology of all the cells under an optical microscope. It was concluded that the morphological analysis of CuNPs treated cancer cells show obvious structural changes like change in cells membrane surface, cell contraction and inhibition of cell growth. Thus, confirming that apoptosis has been induced in CuNPs treated MCF-7 cancer cells J. Xu, et al, (2013).

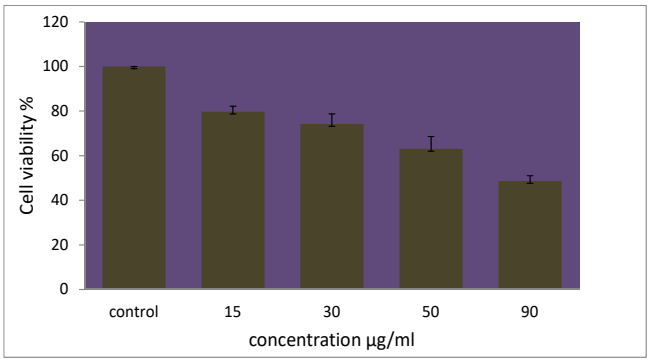


Figure.7
Graphical representation of cytotoxicity of CuNps

Antioxidant Activity of Copper Nanoparticles

The antioxidant activity of the CuNPs is shown in Figure as the DPPH inhibition percentage. The antioxidant activity of the CuNPs increased with the increase in concentration. The inhibition percentage of CuNps (table. 3) was found 55%, 60%, 62% at 20 µg/mL, 40 µg/mL 60 µg/mL respectively. The principle of the DPPH assay is based on the reduction of DPPH in the presence of a hydrogen-donating antioxidant due to the formation of diphenylpicryl hydrazine, where sample compounds alter the DPPH colour due to their hydrogen-donating ability. The colour change of the violet DPPH to yellow clearly demonstrated the effect of CuNPs as an antioxidant H. Y. Lin and C.-C. Chou (2004). Copper nanoparticles exhibited high antioxidant properties compared with

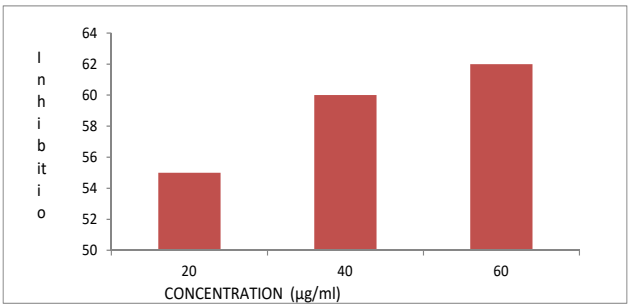


Table.3
Concentration versus inhibition (%) of test samples

the plant extract alone; in order to neutralize the free radicals, the antioxidant itself undergoes oxidation. The activity and stability of the CuNPs, are affected during the anti-oxidation process and also they are oxidized in the presence of air; therefore

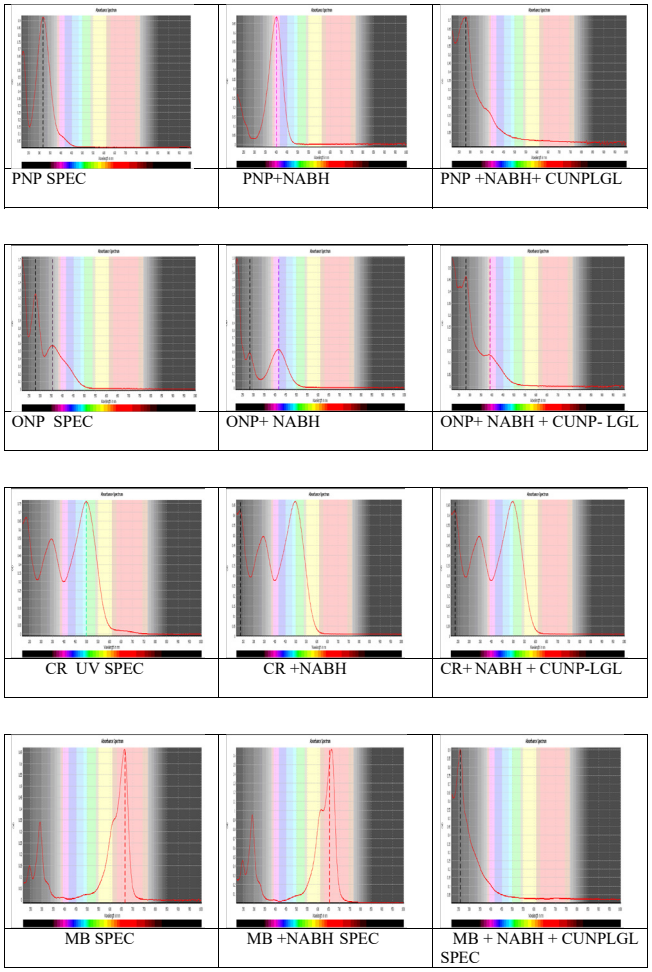


Figure.8
Catalytic activity of copper nanoparticles

metal nanoparticles could be used for treating many diseases caused by oxidative stress Synthesized copper nanoparticles using Lagerstromea specio leaves were used to study the rate of reduction of organic dyes in presence of NABH4 under normal conditions and monitored by UV-Visible spectroscopy. The reaction of organic compounds with NABH4 is thermodynamically favorable but due to the presence of kinetic barrier the feasibility of reaction decreases. Metal nanoparticles reduce the kinetic barrier between donor ions and acceptor ions hence act as catalyst. The absorbance peaks of ortho-nitrophenol, para-nitrophenol, Congo red, methylene blue were observed at 317,355nm, 500nm, 600nm respectively. Upon addition of NABH4 to each compound uv-visible spectra readings were again taken, the absorbace peaks were observed at 400nm, 420nm 550nm, 650nm.This shift in absorbance peaks shows reduction of dyes by NABH4. Samples were kept for several days to observe any further change. No change was observed. The samples were then added with copper nanoparticles, mixed well and uv-visible spectra readings were taken after half an hour. The absorbance peaks were observed at 292nm, 290nm, 250nm, 255nm which showed that further reduction K. Kuroda, T. Ishida, and M. Haruta et al, (2000) has taken place. Hence, it was concluded that copper nanoparticles catalyzed the reaction by reducing the kinetic barrier, thus acts as catalyst.

Conclusion

It was concluded that green synthetic method is better than other method because it is very simple, low-cost and environment friendly. Aqueous extract of leaves of Lagerstroemia speciose were used to synthesize the copper nanoparticles. The so-prepared copper nanoparticles were characterized with Uv-Vis Spectrophotometer, SEM, DLS, FT-IR and XRD to determine their shape, size, stability, chemical composition and nature. In addition to this, the nanoparticles were studied for different applications i.e. anti-diabetic activity, cytotoxicity antimicrobial activity anti-oxidant activity and catalytic activity by different assays. The results calculated displayed strong activities of prepared copper nanoparticles.

Acknowledgements

We are thankful to Director, Defence Food Research Laboratory, Mysore, Karnataka, India, for providing laboratory facilities to work in Nanoscience project under the supervision of Dr. FARHATH KHANUM (Head of Division Nutrition, Biochemistry and Toxicology).

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