ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072

CASSIA OLEORESIN MEDIATED SYNTHESIS, CHARACTERIZATION AND BIOMEDICAL APPLICATION OF COPPER OXIDE NANOPARTICLES

Westeous Dominic Pereira
Saveetha Dental College and Hospitals, Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University, Chennai -77, Email: 151301101.sdc@saveetha.com

Anitha Roy
Associate Professor, Department of Pharmacology, Saveetha Dental College and Hospitals Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University Chennai -77, Email:anitharoy@saveetha.com

S.Rajeshkumar
Associate Professor, Department of Pharmacology, Saveetha Dental College and Hospitals Saveetha Institute of Medical and Technical Sciences (SIMATS), Saveetha University Chennai -77, Email: rajeshkumars.sdc@saveetha.com

ABSTRACT

Aim and Background: Nanotechnology is one of the most rapidly developing topics in biotechnology, having a wide range of potential uses. Nanoparticles have transformed a variety of fields including health, dietary habits, and energy. Nanotechnology's application in medicine, especially drug delivery, has demonstrated several advantages. Nanoparticles are being used to reduce the side-effects and to eliminate the consequences that drugs may have on the patient. The aim of this study is to create copper oxide nanoparticles out of Cassia oleoresin and test their antimicrobial and cytotoxicity properties once they have been characterized.

Materials and Method: The DPPH free radical scavenging activity of cassia oleoresin was evaluated using CuNPs in a DPPH test. Antimicrobial activity was observed using the Agar well diffusion technique, while cytotoxic activity was measured using ELISA plates containing brine shrimp nauplii.

Result: Cassia oleoresins were easily biosynthesized and had antioxidant, antibacterial, and cytotoxic property that was equivalent to standard. The production of nanoparticles had a peak at 350 nanometers, according to a graphical representation. *Candida albicans* was used to assess the antifungal activity, and at a concentration of 150µl, it showed the greatest inhibition zone. Antimicrobial activity was tested against oral pathogens *Streptococcus mutans* and *Lactobacillus*, with a maximal zone of inhibition seen at 150 µl. Antifungal and antibacterial action is represented by inhibition zones. In concentrations 5µL, 10µL, 15µL and 25µL, 100 percent of living brine shrimps were found, whereas in concentrations of 25µL, the proportion of alive brine shrimp was 80 percent.

Conclusion: As a result, it can be concluded that nanoparticles mediated by cassia oleoresin oil have the potential to be implemented as an efficient antioxidant, antibacterial, and cytotoxic agent. hence it can be used in large-scale manufacturing and in a variety of medical applications.

DOI: 10.5281/zenodo.7089072

Keywords: Cassia Oleoresin, CuNPs, Biosynthesis of nanoparticles, antioxidant activity**.**

INTRODUCTION

Nanotechnology is one of the most advancing fields in biotechnology with a vast array of applications. The introduction of Nanoparticles has revolutionized every field including medicine, nutrition and energy. [1, 2] The use of nanotechnology in medicine, specifically drug delivery has shown to have various benefits. Nanoparticles are being utilized to lessen the side-effects and furthermore to dispense with the results that medications may force to the patient. [3, 4] For the past few years, noble-metal nanoparticles have expanded rapidly owing to their increased characteristics. Copper is a red-brown metal; it is in high demand with a usage which exceeds more than 20 million tons per year. Copper oxide nanoparticles have pulled in a critical enthusiasm because of its wide assortment of utilizations, which incorporates impetuses, gas sensors, High-Tc superconductors, obstruction materials, sun-based energy gadgets, and in the planning of natural and inorganic nanocomposites. [5–7] It has also been used in consumer products such as pillowcases and socks, copper oxide is used for its antimicrobial properties. Copper is additionally considered as a fundamental minor component which is required for the correct working of numerous catalysts in biological systems. Metallic nanoparticles may be synthesized by physical, chemical and organic methods. The physical method being highly expensive and the chemical method being extremely noxious to the environment, the biological method is preferred. [8–11] Copper oxide nanoparticles are integrated by various physical and synthetic strategies they incorporate sol-gel, microwave irradiations, sonochemical, alkoxide based route, thermal decomposition of precursor and liquid-liquid interface techniques involving natural solvents and harsh reducing agents. [12]

Plants are traditionally used for various pharmacological action, and many are explored for scientific evidence. Ethanolic leaf extract of *Caralluma fimbriata* has various health benefits including effects against diabetes and Human Colon Cancer Cells., [13, 14] Plant mediated biological synthesis of nanoparticles, has been picking up interests in drug delivery because of its effortlessness and eco kind disposition. [15] The blend of nanoparticles utilizing plants offers different points of interest, for example, the use of more secure solvents, diminished utilization of risky reagents, milder reaction conditions, attainability, and their flexibility being used for therapeutic and drug applications. [16]

Oleoresins are semi-solid concentrates made out of a tar in arrangement in a fundamental oil, acquired by vanishing of the solvents and utilized for their creation. [17, 18] Naturally occurring oleoresins are known as balsams. Oleoresins are set up from flavors, for instance, basil, capsicum, cardamom, celery seed, cinnamon bark, clove bud, fenugreek, fir golden, ginger, jambu, labdanum, mace, marjoram, nutmeg, parsley, pepper, Pimenta (allspice), rosemary, sagacious, appealing, thyme, turmeric, vanilla and West Indian strait leaves. The oleoresin from Brazilian Copaifera species yielded copalic corrosive and sesquiterpenes. [19]

DOI: 10.5281/zenodo.7089072

Cassia, likewise, called Chinese cinnamon, it is a zest containing the fragrant bark of the Cinnamomum cassia plant of the family Lauraceae. [20, 21] Cassia bark is more impactful and has a less fragile flavor and is thicker than cinnamon bark. Cassia bark is utilized as a seasoning operator in cooking and especially in alcohols and chocolate. Cassia bark is peeled from stems and branches these are then set aside to dry. While drying, the bark curls into quills. Ground cassia is reddish brown in color. Cassia cinnamon is most commonly used for the treatment of diabetes. It is also used for pre-diabetes, gas (flatulence), obesity, and many other conditions, but there is no good scientific evidence to support these uses. In a study conducted by Amit Kumar Dutta et al, cassia fistula was tested against Gram-positive and Gram-negative bacteria and different zones of inhibition were formed. [22, 23]

Ultraviolet-visible (UV-Vis) spectroscopy presently is one of the most well-known expository procedures since it is exceptionally adaptable and ready to identify practically every atom. UV-Visible light is passed through a sample and the transmittance of light and then the sample is measured. [24, 25] From the transmittance (T), the absorbance can be determined as A=-log (T). An absorbance range is gotten which shows the absorbance of a compound at various wavelength. The measure of absorbance at any wavelength is seen because of the substance structure of the molecule.

TEM is a valuable and important technique used for the characterization of nanoparticles. It is used to obtain quantitative measures of particle, size distribution, and morphology. The magnification of TEM is mainly dictated by the proportion of the separation between the target focal point and the example, and the separation between the target focal point and its picture plane. TEM is important because it can provide a better spatial resolution and the capability for additional analytical measurements. TEM works on the essential standards as the light magnifying instrument however utilizes electrons rather than visible light. The wavelength of electrons is much smaller than that of light, hence the optimal resolution attainable for TEM images is many orders of magnitude better than that from a conventional light microscope. This makes TEMs suitable to reveal the finest details of internal structure. [26]

Previously our team had conducted numerous studies which included in vitro studies on natural product-based drug discovery. [4, 27–31] Many plant-derived medicines are used as traditional medicinal systems to treat infections, and their potency against oral microbial pathogens has recently been investigated, so the goal of this study is to prepare copper oxide nanoparticles using Cassia oleoresin and analyze their antimicrobial and cytotoxicity effects after characterization.

MATERIALS AND METHODS:

SYNTHESIS OF NANOPARTICLES

For nanoparticle production, 90mL of 1mM copper oxide solution in double distilled water and 10mL of Cassia oleoresin were combined and stored in an orbital shaker with magnetic stirrer.

SEYBOLD Report
ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072

The colour transformation was visually assessed, and images were taken at regular intervals. The NP solution was centrifuged at 8000 rpm for 10 minutes in a Lark refrigerated centrifuge, and the pellet was obtained and washed twice with distilled water. The purified pellet was collected and dried for 24 hours at 60°C before being kept in an airtight Eppendorf tube.

CHARACTERIZATION OF NANOPARTICLES

The synthesized nanoparticle solution was preliminary characterized by using UV-Vis-spectroscopy. 3 mL of the solution was taken in curettes and scanned in double beam; the maximum absorbance was measured within the range of 300–700 nm using a UV- visible spectrophotometer (ELICO SL 210 UV-Vis spectrophotometer) [32]. For the graphical analysis, the findings were recorded

Transmission Electron Microscope (TEM)

TEM was used to examine the particle size and morphology of biosynthesized nanoparticles, using an accelerating voltage of 80 and 200 kV. CuONP samples were prepared for TEM analysis by depositing a tiny amount of the aqueous solution onto carbon-coated copper TEM grids followed by vacuum drying them.

DPPH RADICAL ASSAY:

The DPPH (1,1-diphenyl-2-picryl-hydrazil) free radical scavenging activity of Cassia oleoresin mediated copper nanoparticle was determined by the method of Rajeshkumar,2017 [33]. Different concentrations 10, 20, 30, 40 & 50µg/mL) of extract were mixed with 1 mL of 0.1 mM DPPH in methanol solution and 450 µL of 50 mM Tris-HCl buffer (pH 7.4) and incubated for 30 minutes. The decrease in the amount of DPPH free radicals was determined using the absorbance at 517 nm after incubation. As a control, BHT was employed. Using the equation below, the percent inhibition was computed.:

$$\%\,Inhibition = Absorbance\ of\ control - Absorbance\ of\ test\ sample \div Absorbance\ of\ control \times 100$$

CYTOTOXICITY STUDY:

Brine water was added halfway into ELISA plates with a minimum of 6 wells. With the use of pipettes, 10 brine shrimp babies were precisely measured and deposited inside. The salty water was used to fill the rest of the well. In micro pipettes, concentrations of 5, 10, 15, 20, and 25 µL were collected and added along the 5 wells. Without any change in nanoparticles, 10 brine shrimp babies along with saline into the well, which was utilized as the control.

Table 1 summarizes the observed values in percentage form.

ANTIBACTERIAL ACTIVITY OF NANOPARTICLES AGAINST ORAL PATHOGENS

The agar well diffusion method was used to determine the antibacterial activity of copper oxide nanoparticles [34, 35]. CuNPs at various concentrations were evaluated against *Streptococcus mutans* and *Lactobacillus*. The freshly prepared bacterial suspension was distributed across the surface of Muller Hinton agar plates. The wells were filled with different concentrations of nanoparticles (50, 100, 150 µL), and the plates were incubated at 37°C for 24 hours. Antibiotics served as a positive control. In each plate, the zone of inhibition was measured.

ANTIFUNGAL ACTIVITY

Candida albicans was implanted on Rose Bengal agar plates, and wells were made with concentrations of 50µL, 100µL and 150µL of Cassia oleoresin mediated copper nanoparticle and antibiotic, and the wells were incubated for 48 hours at 37°C. The zone of inhibition was evaluated after the incubation time, and each pathogen's mean value were recorded and displayed in millimeters. Flucanozole was used as a standard.

RESULT AND DISCUSSION:

Copper oxide has a monoclinic structure and is a semiconducting chemical. CuO has captured the attention since it is the most basic member of the copper compound family and also possesses a number of potentially significant physical features such as high temperature superconductivity, electron correlation effects, and spin dynamics.

VISUAL OBSERVATION:

It is realized that copper nanoparticles show. an adjustment in the shade of the arrangement from blue to brownish yellow was observed, with the development of brown colored powder; demonstrating the development of Cu-NPs [36]. The colour is produced as a result of the copper nanoparticles' surface plasmon resonance (SPR) being stimulated. When oleoresin was added to the copper oxide solution, the colour of the solution changed from white to dark brown. Figure 1 depicts this shading shift. This colour shift indicates the configuration of copper oxide nanoparticles, which is further validated using a UV–Visible spectrophotometer.

Figure :1 Color Change of the solution from blue to brownish yellow

UV-Visible Spectroscopy and TEM:

The use of UV-Visible spectroscopy to ascertain the production and stability of copper nanoparticles in aqueous solution is a viable approach. The UV–visible spectra of the produced

SEYBOLD Report
ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072

nutmeg oleoresin copper oxide solution were recorded (fig 2). The spectra show that the CuNP's SPR peak appears around 350 nm with high absorbance, which is typical of copper nanoparticles. This demonstrates the production of copper nanoparticles mediated by cassia oleoresin. The TEM picture clearly reveals the formation of clusters of spherical nanoparticles.

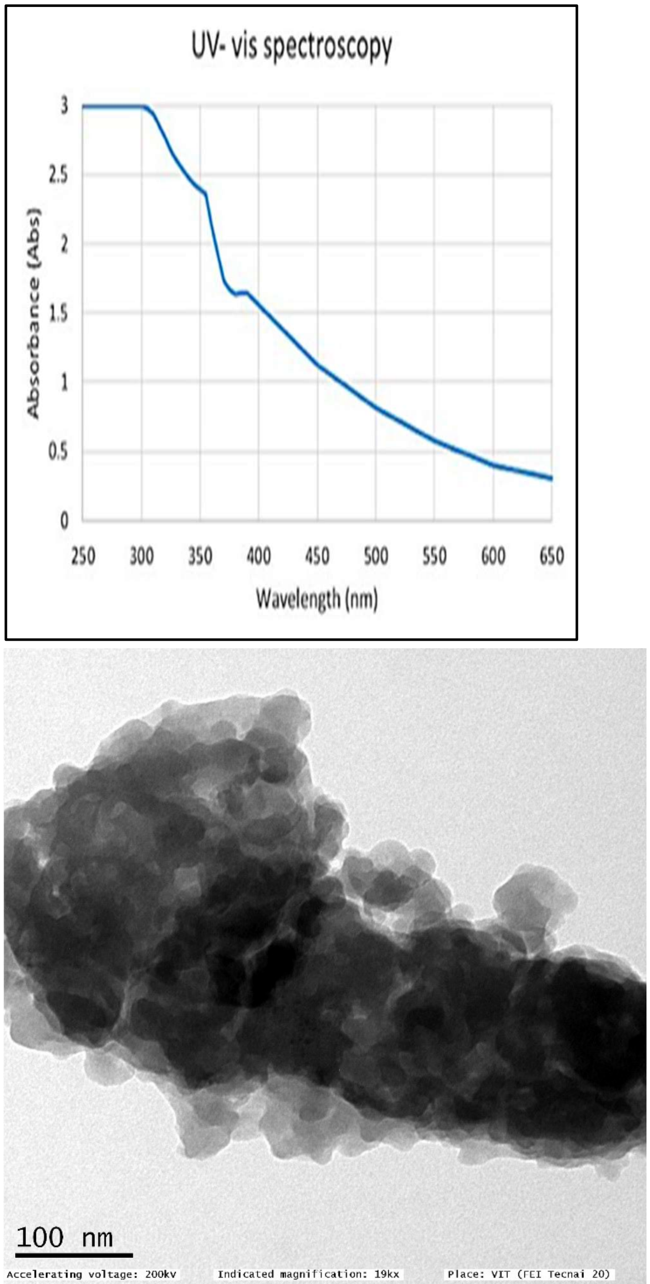


Figure 2 UV-vis spectroscopy and TEM image

ANTIBACTERIAL ACTIVITY:

Evaluation of the antimicrobial activity of the extracts was conducted according to the agar well diffusion method. [37] The antibacterial activity of the synthesized cassia oleoresin mediated CuNPs against the oral pathogens *Streptococcus mutans* and *Lactobacillus* was

DOI: 10.5281/zenodo.7089072

investigated. The diameter of the zone of inhibition (ZOI) around each cell in millimetres was measured in different concentrations of cassia oleoresin mediated CuNPs against bacterial species. *(Figure 3-4)*

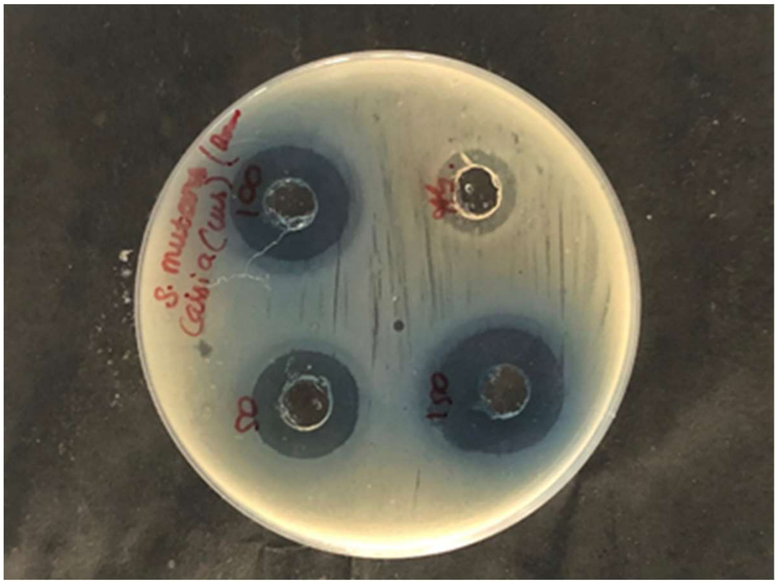


Fig 3 Streptococcus mutans (Zone of inhibition)

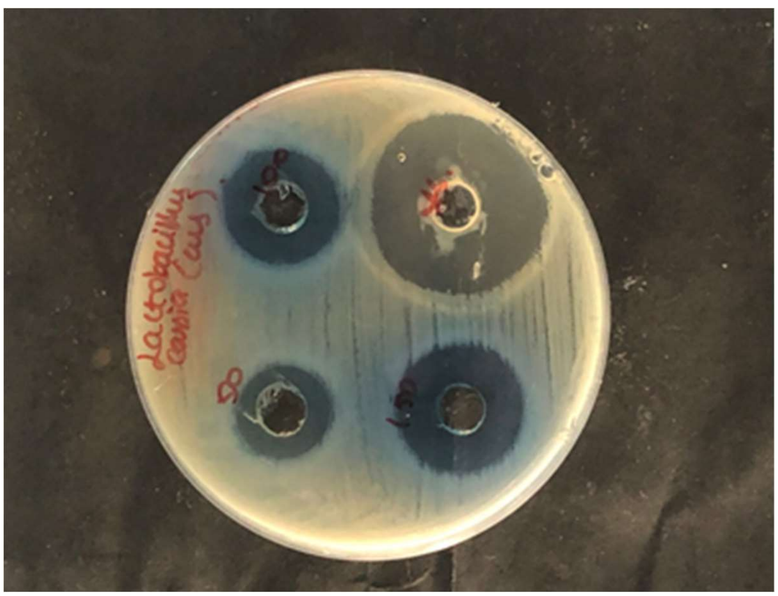


Fig 4 Lactobacillus (Zone of inhibition)

ANTIFUNGAL ACTIVITY

Antifungal activity was determined by inoculating Rose Bengal agar with colonies of Candida albicans, a harmful fungus. Flucanozole was used as the standard. At 50, 100, and 150 µL, the ZOI of the obtained CuNPs against Candida Albicans was found to be 30 mm, 35 mm, and 37 mm, respectively. The Zone of Inhibition grows as the number of nanoparticles rises. At a concentration of 150 µL, the maximum zone of inhibition was detected. The antifungal activity

SEYBOLD Report
ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072

of Cassia oleoresin mediated copper oxide nanoparticles is demonstrated by the zone of inhibition.

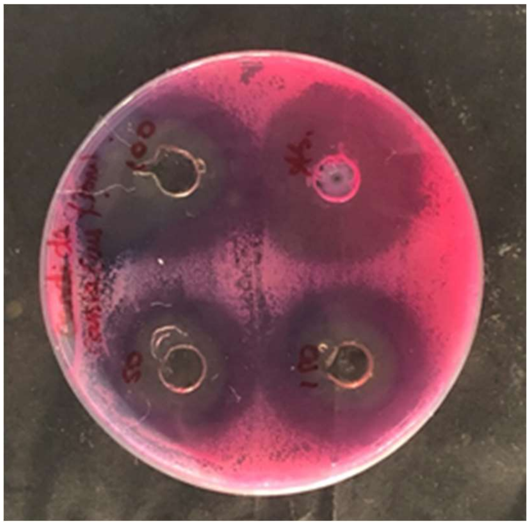


Fig 5 Candida albicans (Zone of inhibition)

The nanoparticles obtained in this study are noticeable and have the potential to be an antagonistic against oral pathogens. The antibacterial impact of the prepared CuNPs was similar against all of the species tested, with a maximum ZOI. The antibacterial activity of CuNP rose when the concentration was increased.

The ZOI of *Streptococcus mutans* was observed to be 20mm, 23mm, and 31mm, respectively. The ZOI values for *Lactobacillus* are 20mm, 23mm, and 26mm, respectively. As the concentration level increased, the zone of inhibition also increased. The data has been tabulated and put as a graph (Figure 6 and 7). The antimicrobial activity of the synthesised copper nanoparticles is because of the copper cations released from them, which cause changes in the membrane structure of microbes, resulting in increased membrane permeability and, eventually, cell death.

The action of metallic copper, copper particles, and colloidal copper nanoparticles is dependent on changes in the structure and capacity of the fungus cell; these particles can alter DNA and disrupt replication and transcription, causing the death of infectious microorganisms.

DOI: 10.5281/zenodo.7089072

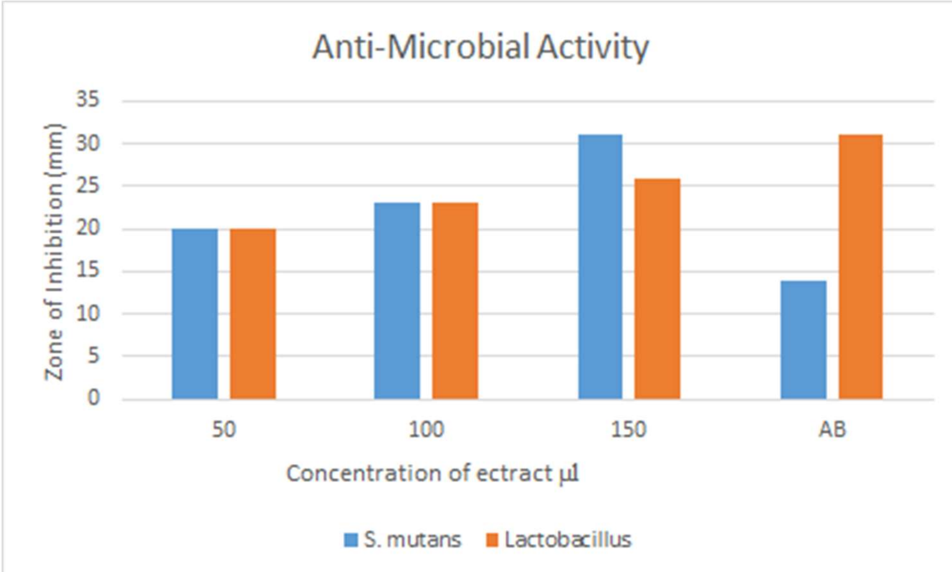


Figure 6 demonstrates Antimicrobial action. The X-axis represents the concentration in µl, while the Y-axis represents the percentage of inhibition. With increasing concentration, the zone of inhibition increases.

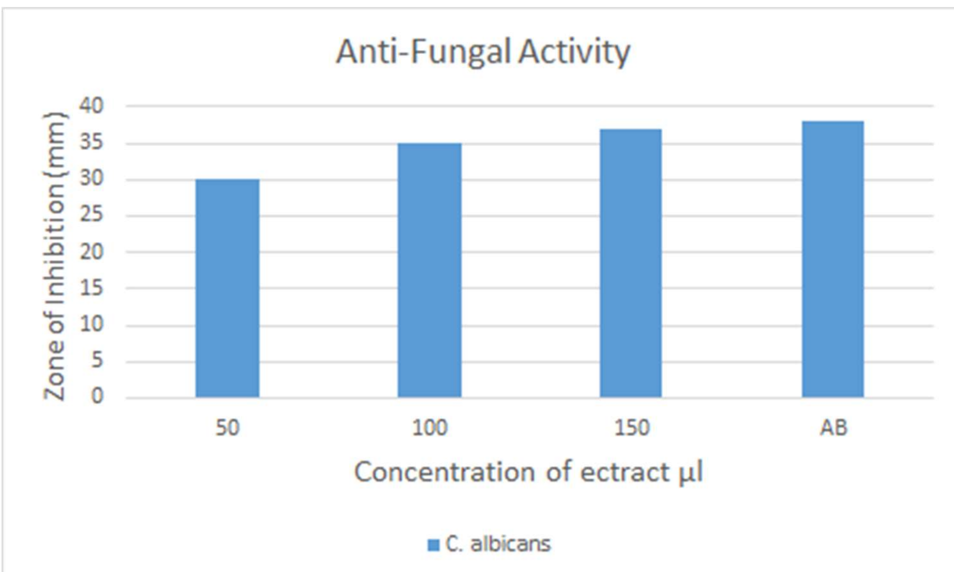


Figure 7 demonstrates Antifungal action. The X-axis represents concentration, whereas the Y-axis represents the zone of inhibition. With increasing concentration, the zone of inhibition increases.

CYTOTOXIC ACTIVITY:

Brine Shrimp Test (BST) is a method utilizing brine shrimp, it is proposed as a simple bioassay for natural product research. It can be used as a low-cost test indicative of antibacterial, cytotoxic, pesticidal, and insecticidal activity and could be used as a simple method for screening antibacterial products [38, 39]. In the current research, the rate of living brine shrimps was 100% in concentrations of 5µL, 10µL, 15µLand 25µL, while the percentage of alive brine

DOI: 10.5281/zenodo.7089072

shrimps was 80% in concentration of 25µL.

Concentration	Percentage of Alive Brime Shrimps
5	100%
10	100%
15	100%
20	100%
25	80%
Control	100%

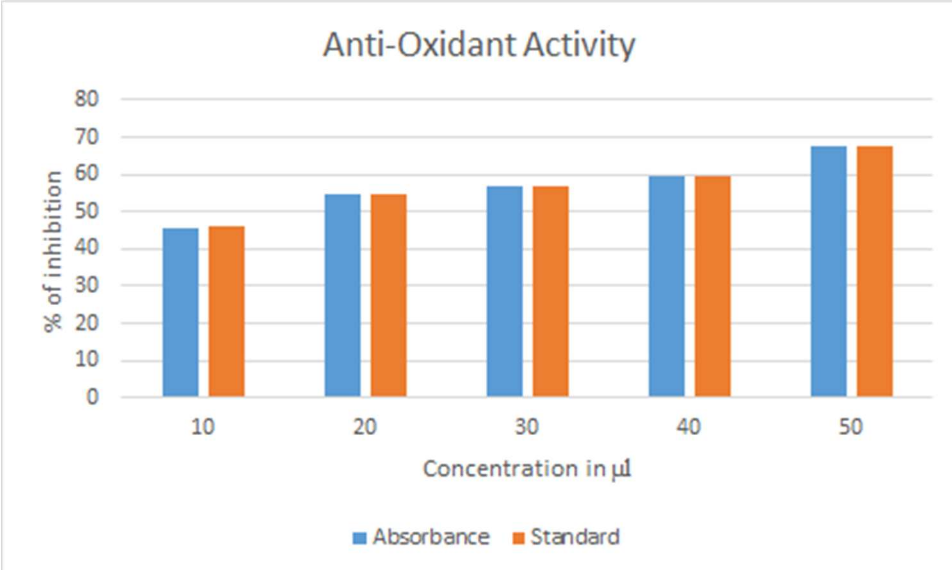
Table 1 (Cytotoxic Activity)

ANTIOXIDANT ACTIVITY:

Hydrogen giving limit or free radical scavenging by the nanoparticles was assessed by the DPPH measure, which relies upon the decrease of the methanolic hued revolutionary sort of the DPPH to the non-colored solution.[40] After combining with cassia oleoresin oil extract and incubation, copper ions were reduced to CuNPs in the present study. The hue changed from yellow to reddish brown, and the same has been observed by various investigators before.

Copper nanoparticles are employed in modern biomedical applications on a regular basis. DPPH has been widely utilized as a stable free to assess reducing compounds and as a suitable reagent for studying the component's free radical scavenging capability. Figure 7 depicts copper nanoparticles' DPPH free radical scavenging capability. Copper nanoparticles have a free radical scavenging action that is quite similar to that of ascorbic acid.

SEYBOLD Report
ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072



CuNPs Anti-oxidant activity is depicted in Figure 7. The X-axis represents the concentration in µl, while the Y-axis represents the percentage of inhibition. The maximum zone of inhibition was seen at a concentration of 50 µl.

CONCLUSION:

Cassia oleoresin was investigated as a new green reducing and stabilising agent for the synthesis of Copper nanoparticles (CuNP's) using a new green chemistry technique in this work. Color shift from white to dark brown indicated the creation of CuNPs, which was validated using a UV-Vis spectrophotometer and a transmission electron microscope. The CuNPs induced by cassia oleoresin had a lot of antioxidants, cytotoxic, and antimicrobial activity against oral pathogens such as *Streptococcus mutans, Lactobacillus* and *Candida albican*s. Hence, the novel green approach for the synthesis of CuNPs using cassia oleoresin was an ecofriendly technology that can be employed in the production of new drugs against oral pathogens as well as new pharmaceutical research.

ACKNOWLEDGEMENT

Authors express sincere gratitude to Saveetha Dental College for the extended support towards the completion of the research. Authors would also like to thank Synthite Industries Pvt Ltd,Kerala for providing extract to carry out this study.

FUNDING

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

CONFLICT OF INTEREST

No potential conflict of interest relevant to this article was reported.

DOI: 10.5281/zenodo.7089072

REFERENCE.

[1] Mistry KR, Sarker DK. SLNs can serve as the new brachytherapy seed: determining influence of surfactants on particle size of solid lipid microparticles and development of hydrophobised copper nanoparticles for potential insertion. *J Chem Eng Process Technol* 2016; 7: 302.

[2] Gayathri K, Roy A, Lakshmi T, et al. Controlling of oral pathogens using ginger oleoresin mediated silver nanoparticles. *International Journal of Research in Pharmaceutical Sciences* 2019; 10: 2988–2991.

[3] Jain A, Anitha R, Rajeshkumar S. Anti inflammatory activity of Silver nanoparticles synthesised using Cumin oil. *Research Journal of Pharmacy and Technology* 2019; 12: 2790.

[4] Ka MTA, Roy A, Rajeshkumar S, et al. Anti-inflammatory activity of cinnamon oil mediated silver nanoparticles-An in vitro study. *International Journal of Research in Pharmaceutical Sciences* 2019; 10: 2970–2972.

[5] Li Y, Liang J, Tao Z, et al. CuO particles and plates: Synthesis and gas-sensor application. *Mater Res Bull* 2008; 43: 2380–2385.

[6] Kruis FE, Fissan H, Peled A. Synthesis of nanoparticles in the gas phase for electronic, optical and magnetic applications—a review. *J Aerosol Sci* 1998; 29: 511–535.

[7] Cheon J, Lee J, Kim J. Inkjet printing using copper nanoparticles synthesized by electrolysis. *Thin Solid Films* 2012; 520: 2639–2643.

[8] Burdock GA. Fenaroli's Handbook of Flavor Ingredients. Epub ahead of print 2016. DOI: 10.1201/9781439847503.

[9] Bajaj G, Chaudhary A, Naaz H, et al. Laser ablation synthesis of Zn/ZnO core-shell nanoparticles. In: *2007 International Workshop on Physics of Semiconductor Devices*, pp. 940–942.

[10] Jia FL, Zhang LZ, Shang XY, et al. Non-Aqueous Sol--Gel Approach towards the Controllable Synthesis of Nickel Nanospheres, Nanowires, and Nanoflowers. *Adv Mater* 2008; 20: 1050–1054.

[11] Tanori J, Pileni MP. Control of the Shape of Copper Metallic Particles by Using a Colloidal System as Template. *Langmuir* 1997; 13: 639–646.

[12] Hosseinzadeh R, Mohadjerani M, Mesgar S. Green synthesis of copper oxide nanoparticles using aqueous extract of Convolvulus percicus L. as reusable catalysts in

DOI: 10.5281/zenodo.7089072

cross- coupling reactions and their antibacterial activity. *IET Nanobiotechnology* 2017; 11: 725–730.

[13] Ashwini S, Ezhilarasan D, Anitha R. Cytotoxic effect of Caralluma fimbriata against human colon cancer cells. *Pharmacognosy Journal*; 9, https://www.phcogj.com/article/252 (2017).

[14] Ashwini S, Anitha R. Antihyperglycemic Activity of Caralluma fimbriata: An In vitro Approach. *Pharmacogn Mag* 2017; 13: S499–S504.

[15] Kandhan TS, Roy A, Lakshmi T, et al. Green synthesis of Rosemary oleoresin mediated silver nanoparticles and its effect on Oral pathogens. *Research Journal of Pharmacy and Technology* 2019; 12: 5379–5382.

[16] Abdel-Halim ES, El-Rafie MH, Al-Deyab SS. Polyacrylamide/guar gum graft copolymer for preparation of silver nanoparticles. *Carbohydr Polym* 2011; 85: 692–697.

[17] Muggeridge M, Clay M. Quality specifications for herbs and spices. *Handbook of Herbs and Spices* 2001; 13–21.

[18] Suderman DR, Cunningham FE. Batter and breading, http://agris.fao.org/agris-search/search.do?recordID=US8817544 (1983).

[19] Basile AC, Sertié JA, Freitas PC, et al. Anti-inflammatory activity of oleoresin from Brazilian Copaifera. *J Ethnopharmacol* 1988; 22: 101–109.

[20] Ravindran PN, Nirmal-Babu K, Shylaja M. *Cinnamon and Cassia: The Genus Cinnamomum*. CRC Press, https://play.google.com/store/books/details?id=KZa8aPxR_-wC (2003).

[21] Hajimonfarednejad M, Ostovar M, Raee MJ, et al. Cinnamon: A systematic review of adverse events. *Clinical Nutrition* 2019; 38: 594–602.

[22] Dutta AK, Madharia P. In-vitro Evaluation of Antibacterial Activity of Cassia fistula against different Gram-positive and Gram-negative Bacteria. *Biomedical & Pharmacology Journal* 2012; 5: 185.

[23] Preety R, Anitha R, Rajeshkumar S, et al. Anti-diabetic activity of silver nanoparticles prepared from cumin oil using alpha amylase inhibitory assay. *International Journal of Research in Pharmaceutical Sciences* 2020; 11: 1267–1269.

[24] Gurunathan S, Han JW, Kim ES, et al. Reduction of graphene oxide by resveratrol: a novel and simple biological method for the synthesis of an effective anticancer nanotherapeutic molecule. *Int J Nanomedicine* 2015; 10: 2951–2969.

DOI: 10.5281/zenodo.7089072

[25] Sapsford KE, Tyner KM, Dair BJ, et al. Analyzing nanomaterial bioconjugates: a review of current and emerging purification and characterization techniques. *Anal Chem* 2011; 83: 4453–4488.

[26] Popov VS, Sopilniak A. Quartz Crystal Microbalance Application for Characterization of Nanomaterials In Situ. *In-situ Characterization Techniques for Nanomaterials* 2018; 351–382.

[27] Nivesh Krishna R, Roy A, Ezhilarasan D. Aqueous extract of Tamarindus indica fruit pulp exhibits antihyperglycaemic activity. *Avicenna Journal of Phytomedicine*, http://ajp.mums.ac.ir/article_14787_0.html (2020).

[28] Aneesa NN, Anitha R, Varghese S. Antidiabetic Activity of Ajwain Oil in Different In Vitro Models. *J Pharm Bioallied Sci* 2019; 11: 142–147.

[29] Keerthiga N, Anitha R, Rajeshkumar S, et al. Antioxidant activity of cumin oil mediated silver nanoparticles. *Pharmacognosy Journal*; 11, http://www.phcogj.com/article/930 (2019).

[30] Aafreen MM, Maajida Aafreen M, Anitha R, et al. Anti-Inflammatory Activity of Silver Nanoparticles Prepared from Ginger Oil—An Invitro Approach. *Indian Journal of Public Health Research & Development* 2019; 10: 145.

[31] Reddy JM, Meghana Reddy J, Anitha R, et al. Characterisation of Cumin oil mediated silver nanoparticles using UV-visible spectrophotometer and TEM. *Research Journal of Pharmacy and Technology* 2019; 12: 4931.

[32] Sharma D, Sabela MI, Kanchi S, et al. Green synthesis, characterization and electrochemical sensing of silymarin by ZnO nanoparticles: Experimental and DFT studies. *J Electroanal Chem* 2018; 808: 160–172.

[33] Rajeshkumar S, Rinitha G. Nanostructural characterization of antimicrobial and antioxidant copper nanoparticles synthesized using novel Persea americana seeds. *OpenNano* 2018; 3: 18–27.

[34] Malarkodi C, Rajeshkumar S, Paulkumar K, et al. Biosynthesis and Antimicrobial Activity of Semiconductor Nanoparticles against Oral Pathogens. *Bioinorganic Chemistry and Applications* 2014; 2014: 1–10.

[35] Pranati T, Anitha R, Rajeshkumar S, et al. Preparation of Silver nanoparticles using Nutmeg oleoresin and its Antimicrobial activity against Oral pathogens. *Research Journal of Pharmacy and Technology* 2019; 12: 2799–2803.

[36] Suárez-Cerda J, Espinoza-Gómez H, Alonso-Núñez G, et al. A green synthesis of copper

SEYBOLD Report
ISSN: 1533 - 9211
DOI: 10.5281/zenodo.7089072

nanoparticles using native cyclodextrins as stabilizing agents. *Journal of Saudi Chemical Society* 2017; 21: 341–348.

[37] Vignesh S, Anitha R, Kumar SR, et al. Evaluation of the Antimicrobial activity of Cumin oil mediated silver nanoparticles on Oral microbes. *Research Journal of Pharmacy and Technology* 2019; 12: 3709–3712.

[38] Sathasivam P, Lakshmi T. Brine shrimp lethality assay of Sesbania grandiflora ethanolic extract--in vitro study. *Journal of Advanced Pharmacy Education & Research*; 7, https://www.speronline.com/japer/Articlefile/JAPER_8_2017.pdf (2017).

[39] Meyer BN, Ferrigni NR, Putnam JE, et al. Brine shrimp: a convenient general bioassay for active plant constituents. *Planta Med* 1982; 45: 31–34.

[40] Rajeshkumar S, Menon S, Venkat Kumar S, et al. Antibacterial and antioxidant potential of biosynthesized copper nanoparticles mediated through Cissus arnotiana plant extract. *J Photochem Photobiol B* 2019; 197: 111531.