RESEARCH PAPER

Formulation and Evaluation of Copper Nanoparticles Using *Daucus Carota Linn* Leaves Extract for Cancer

Arzoo[1], Aakash Deep[1*], Harish Kumar[1], Rimmy Nandal[2], Anil Kumar[3], Bhagwati Devi[4], Teena Bhardwaj[5], Sangeeta Sharma[6]

[1*]Department of Pharmaceutical Sciences,
Chaudhary Bansi Lal University, Bhiwani, Haryana, India-127021.
Mobile: +919896096727, Email: aakashdeep82@gmail.com.
[2]Faculty of Pharmaceutical Sciences,
Assam Down Town University, Assam 781026, India.
[3]Geeta Institute of Pharmacy, Geeta University, Panipat.
[4]School of Pharmaceutical Sciences, CGC University, Mohali Punjab 140307.
[5]BM college of Pharmacy, Farukhnagar, Gurugram.
[6]Department of Psychology,
Chaudhary Bansi Lal University, Bhiwani, Haryana, India-127021.

ABSTRACT

The introduction of this research topic develops into the burgeoning field of nanotechnology and its applications in cancer, focusing on the synthesis of copper nanoparticles using Daucus carota (carrot) extract. The goal of our further research is to thoroughly test these nanoparticles by UV Spectroscopy, X-ray Diffraction, SEM, FTIR, Gas Chromatography and SBR Assay. In this study, the materials and methods employed are centered on the synthesis of copper nanoparticles using Daucus carota (carrot) extract and the subsequent evaluation of their anti-cancerous activity. Fresh *Daucus Carota Linn* leaves were collected, cleaned, and processed to obtain an aqueous extract. 25 gm of grinding of leaves material added into the 250 ml ethanol extracting using soxhlet method. After, the extraction, then it filtered using whatmann filter paper no. 1. 5 mL of an aqueous plant extract were mixed with 50 mL of a copper sulphate pentahydarte solution (5 mM). For later use, a dark green powder that was obtained was kept at temperature. Centrifugation was used to separate the synthesized copper nanoparticles for 10 min. at 6,000 rpm. Phytochemical analysis was performed for the following components such as alkaloids, amino acids, carbohydrates, phenolic compounds, saponins, flavanoids, and terpenoids. The result showed good activity against cancerous cells and furthermore clinical study is needed to bring out the full potential of this study.

Keywords: Nanoparticles, *Daucus Carota Linn*, Characterization, anticancer activity

How to cite this article: Arzoo, Aakash Deep, Harish Kumar, Rimmy Nandal, Anil Kumar, Bhagwati Devi, Teena Bhardwaj, Sangeeta Sharma, "Formulation and Evaluation of Copper Nanoparticles Using *Daucus Carota Linn* Leaves Extract for Cancer" Int J Drug Deliv Technol. 2026;16(50s): 1186-1195. DOI: 10.25258/ijddt.16.50s.125.

DOI: xxxx

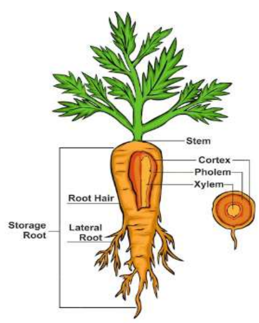
INTRODUCTION

Green synthesis of nanoparticles (NPs) has been a recent development in the field of nanotechnology. This kind of synthesis is more eco-friendly approach compared to the chemical and physical methods. It involves the use of natural ingredients in the bio-reduction of metallic ions to nanoparticles. This simple green eco-friendly method has been utilized to obtain metal nanoparticles using the leaf extract as a precursor. Safer and easily metabolized drugs are natural compounds retrieved from plant materials as compared to other synthetic medicinal compounds. Metallic nanoparticles are mostly synthesized from precious metals viz., silver (Ag), gold (Au), platinum (Pt), and lead (Pb). From these metals, silver is the most preferred metal in the area of the biological system due to its wide range of activities over other metals. The development of these green nanoparticles has been intensified in recent research not only for their fundamental scientific relevance but also for diverse technological applications (Ejidike and Clayton, 2022). Nanomedicine, the application of nanotechnology to medicine, aims to overcome problems, related to diseases, at the nanoscale where most of the biological molecules exist and operate. Particularly nanotechnology application to cancer aims to bring significant breakthroughs in diagnosis, treatment and monitoring of cancer. Today, there is a strong focus on nanotechnology application to cancer. Cancer Nanotechnology is a new field of interdisciplinary research cutting across biology, chemistry, engineering and medicine aiming to lead major advances in cancer detection diagnosis and treatment (Wang and Thanou, 2010). We can see cancer treatment modalities by dividing them into conventional (traditional) and advanced or novel or modern categories (Debela et al., 2021). The most cutting-edge and creative cancer therapeutic approaches include targeted therapy, nanomedicine, gene therapy, siRNA shipment, immunization and molecules of antioxidants enhance the bio-distribution of newly created or tested chemotherapeutic agents around the particular tissue that needs to be treated (Dashtizadeh et al., 2021).

Both physical and chemical methods can be used to produce different types of copper nanoparticles (Thakur, Rai and Sharma, 2014). The biomedical sciences have made substantial use of copper nanoparticles made through green synthesis using medicinal plants to treat a variety of diseases (Liu *et al*., 2021, Tahvilian *et al*., 2019).

arrot (Daucus carota L.) are rich in dietary fiber, anti-oxidants, anthocyanins, carotenoids, vitamins A, B, and C, and minerals. Daucus carota, an edible carrot is an important root vegetable plant grown all over the world, and they belong to the family Apiaceae. The leaves are bristly pinnate in a pattern that separates into thin segments. The aerial parts are used as livestock feed in some parts of Africa and also as vegetables for human beings after harvesting or thrown away. Traditionally, parts of D. carota have been utilized as anti-diabetic, diuretic, muscle and back pain reliever, and aphrodisiac. Ethnomedically, the roots of D. carota L. have been reported to be useful in the treatment of digestive problems, tonsillitis or constipation, and intestinal parasites (19). Aqueous hot leaf extract is taken orally as a uterine stimulant during parturition and has abortifacient potential (29,30). The four most common kinds of phytochemicals dominant in carrots are polyacetylenes, carotenoids, phenolics, and ascorbic acid

Carrot (Daucus carota L.) are rich in dietary fiber, anti-oxidants, anthocyanins, carotenoids, vitamins A, B, and C, and minerals. Daucus carota, an edible carrot is an important root vegetable plant grown all over the world, and they belong to the family Apiaceae. The carrot is composed of the umbel, the stem, and the root (Figure 1A). The leaves are bristly pinnate in a pattern that separates into thin segments. The aerial parts are used as livestock feed in some parts of Africa and also as vegetables for human beings after harvesting or thrown away (Mandrich et al., 2023). Traditionally, parts of D. carota have been utilized as anti-diabetic, diuretic, muscle and back pain reliever, and aphrodisiac. Ethnomedically, the roots of D. carotaL. have been reported to be useful in the treatment of digestive problems, tonsillitis or constipation, and intestinal parasites. Aqueous hot leaf extract is taken orally as a uterine stimulant during parturition and has abortifacient potential. The four most common kinds of phytochemicals dominant in carrots are polyacetylenes, carotenoids, phenolics, and ascorbic acid (Fareed *et al*., 2023)



(a)

(b)

(c)

(d)

(e)

Figure 1.(A) Schematic representation of Daucus Carota L. Its different parts are reported: petiol, hypocotyl, and root and its cross-section, where the phloem and xylem are represented. (B) Carrot white flowers; (C)D. carota subsp. carota (wild carrot); (D) D. carota subsp. Sativus (domestic orange carrot); (E) D. carota subsp. Sativus (domestic violet carrot).

MATERIAL AND METHOD

Materials

Daucus Carota Linn leaves was collected from Bhiwani (Haryana). The specimen of the plant sample was submitted and authenticated in Department of Botany, MDU, Rohtak. After collecting the leaves, tap water was used to wash them, and then deionized water. Dried leaves were ground by using mortar pestle. 25 gm of ground leaves were added into the 250 ml ethanol for extraction using Soxhlet method. Once the extraction was completed, it was filtered through filter paper no. 1 and kept at room temperature (Anaja 2016).

Method of Preparation

The green synthesis approach was used to formulate copper nanoparticles. 50 mL of a Copper sulphate pentahydarte solution (5 mM) and 5 mL of an aqueous plant extract were used to produce copper nanoparticles from *Daucus carota Linn*. NaOH (1 N) solution was added to extract to maintain pH at 7 resulting in formation of green color. The produced copper nanoparticles were separated using centrifugation for 10 minutes at 6,000 rpm. Pellets were separated from the mixture after centrifugation, and they were dried in a hot air oven for overnight at 60°C. The dark green powder was kept at room temperature for further use (Sundaram 2020).

EVALUATION OF COPPER NANOPARTICLES

UV-Visible spectroscopy

UV-vis spectroscopy is an inexpensive, simple, flexible, non-destructive, analytical method appropriate for a wide class of organic compounds and some inorganic species. UV-vis spectrophotometers measure the absorbance or transmittance of light passing through a medium as a function of the wavelength (Rocha et al., 2018). The synthesized copper nanoparticles were optically measured using double beam UV-VIS spectroscopy. It immediately alters the color of substances by using visible range absorption spectroscopy. The optical properties of the synthesized CuNPs were investigated at various wavelengths between 250 and 350 nm (Mali 2020).

Scanning Electron Microscope

Scanning electron microscope (SEM) is one of the most versatile instruments used for the examination and analysis of the microstructure morphology and chemical composition characterizations (Zhou et al., 2007). Scanning electron microscopy (SEM) produces images at 500 000 times magnification and better than 1 nm resolution to characterize inorganic and organic solid morphology, surface topography, and crystallography. An electron beam interacts with the material and generates secondary electrons (SE) and backscattered electrons (BSE) that detectors capture. Although SEM instrument capability continuously evolves with higher magnification and better resolution, desktop SEMs are becoming standard in laboratories that require frequent imaging and lower magnification. Hand-held cameras (800–1500×) have the advantage of low cost, ease of use, and better colors (Davies et al., 2022).

X Ray Diffraction

X-ray diffraction (XRD) is a powerful nondestructive technique for characterizing crystalline materials. XRD instrumentation has undergone significant advancements (Surdu and Gyorgy, 2023). It provides information on structures, phases, preferred crystal orientations (texture), and other structural parameters, such as average grain size, crystallinity, strain, and crystal defects. X-ray diffraction peaks are produced by constructive interference of a monochromatic beam of X-rays scattered at specific angles from each set of lattice planes in a sample. The peak intensities are determined by the distribution of atoms within the lattice (Bunaciu et al., 2015). X-ray diffraction patterns ensure the formation of copper nanoparticles by the diffraction angle, intercity and miller indices (Tabassum et al., 2024).

Fourier transforms infrared spectroscopy

With the help of Fourier transform infrared spectrophotometer (FTIR), the functional groups present in the plant extract components are detected. The annotated spectrum provides the wavelength of absorbed light, which is indicative of the chemical bond. Chemical bonding can be identified by analyzing infrared absorption spectra. The silver nanoparticles were spun at 12,000 rpm for 30 minutes in preparation for FTIR analysis. Following that, 25 ml of de-ionized water was used to rinse the pellets three times (Nazneen and Sultana, 2024).

Gas Chromatography Mass Spectroscopy (GCMS)

The GC–MS technique was adopted for the detection and identification of phytochemical compounds present in copper nanoparticles. The equipment used for the GC-MS analysis is PerkinElmer GC Clarus 500 system (Waltham, MA, USA). The GC–MS detection was executed using electron impact mode with an ionization energy of 70 eV. The carrier gas was helium (99.999%) with a constant flow rate of 1 mL/min and the injection volume was 1 µL. The temperature parameters were 250°C for the injector and 200°C for the ion source (Pei, Jiang and Sun, 2023). From 70 to 280 degrees Celsius, the oven's temperature was raised at a rate of 8 degrees per minute. One liter of sample is injected through the injector. At 70eV, the MS was conducted. The name, molecular weight, and structure of unknown compounds were detected by comparing the spectra of unknown chemicals to the spectrum of known compounds (Nazneen and Sultana, 2024).

Anticancer Activity (SBR Assay)

In this investigation, cell lines were grown in a medium enhanced with 2 mM glutamine and 10% fetal bovine serum. For the screening test, 5000 cells were added per well in 96-well microtiter plates and incubated for 24 hours. Experimental drugs were prepared by diluting frozen stocks with water to obtain concentrations of 10 µg/ml, 20 µg/ml, 40 µg/ml, and 80 µg/ml. The test was stopped using cold trichloroacetic acid (TCA) after the drugs were incubated for 48 hours. Fixed cells were then treated with 0.4% sulforhodamine B (SRB) solution in 1% acetic acid, and after 20 minutes, the plates were washed, dried, and dye was eluted with 10 mM trizma base. Absorbance was measured at 540 nm with a reference wavelength at 690 nm. The percentage growth was computed by comparing test wells against control wells. The concentration that resulted in a 50% decrease in net protein growth was identified as the GI_{50}. If the targeted activity levels were reached, values for GI_{50}, TGI, and LC_{50} were found; if not, results were shown as either surpassing or falling below the tested concentration range.

RESULT AND DISCUSSION

UV-Visible Spectroscopy (Calibration Curve)

The maximum concentration λmax of the drug *Daucus Carota Linn* was determined by scanning the produced solution in the 250-300 nm wavelength range. A wavelength of 280 nm was found to be the maximum. Set the pH of *Daucus carota linn* to pH 8–9. The curve was formed using a concentration range of 2–10 µg/ml, and the regression coefficient was found to be linear i.e. (R^2) is 0.9997.

Table No.

S.No	Concentration (µg/mL)	Absorbance value
1.	*0*	0
2.	2	0.039
3.	4	0.082
4.	6	0.121
5.	8	0.159
6.	10	0.201

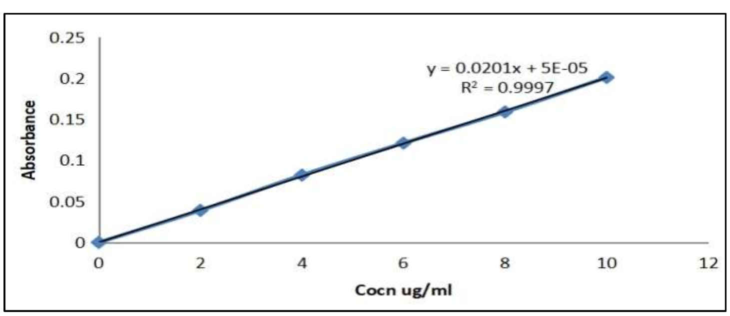


Fig.1 Calibration curve of *Daucus Carota linn*

Scanning Electron Microscopy

Scanning electron microscopy was used to analyze the particle morphology by scanning. The SEM image clearly showed the spherical form of the nanoparticles. Products with different morphologies may arise from the natural component, copper (II), with differing grades of variability. The material with the greatest absorption peak value is copper nanoparticles. The image demonstrates the estimated 200 nanoscale magnitudes of the preparation.

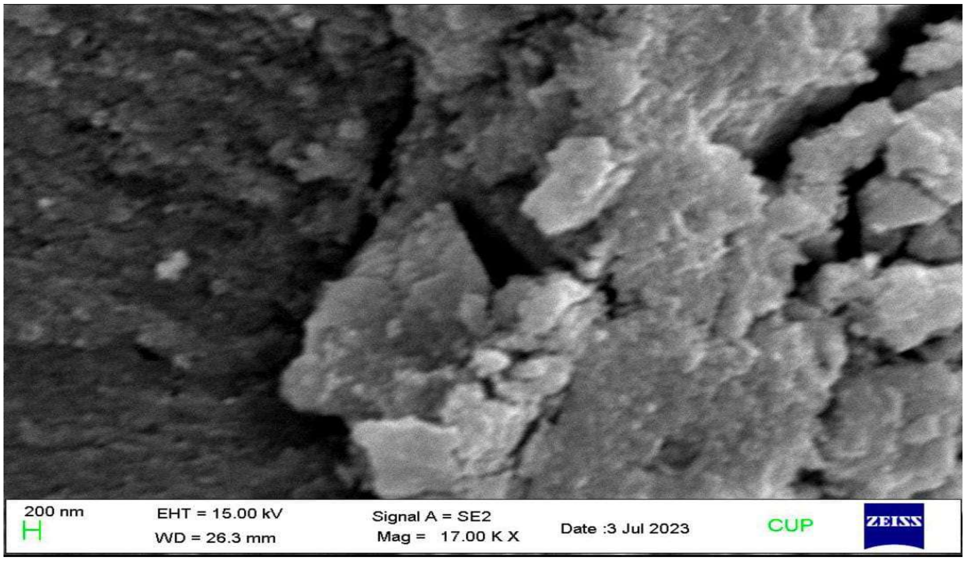


Fig.2 SEM Image Daucus Carota Linn at 200nm

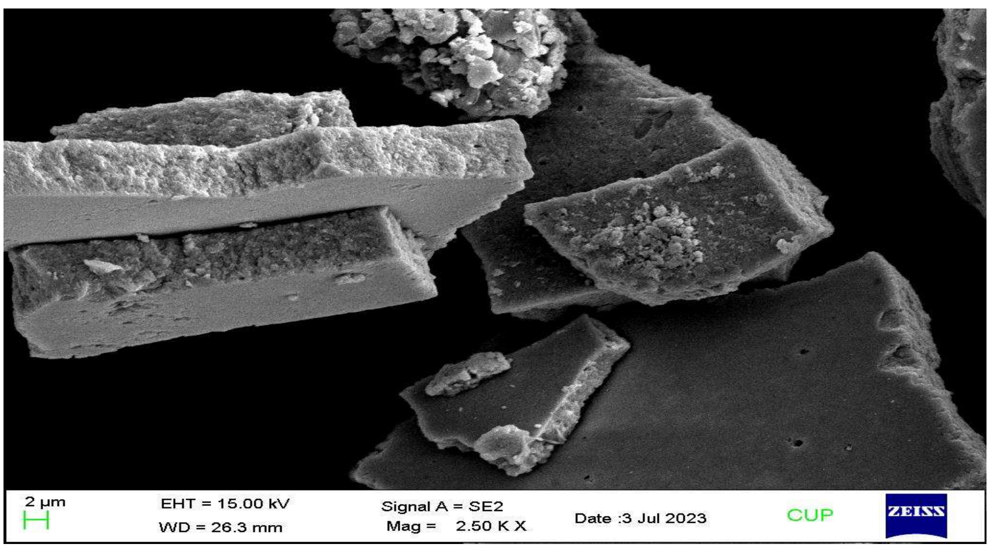


Fig 3 SEM Image Daucus Carota Linn at 2µm

XRD Analysis

For the XRD study, the Rigaku Miniflex-600 diffractometer model was utilized. A Cu K source operation (40 kV, 15 mA) was used at room temperature. Scherer's equation ($D = K\lambda / \beta Cos\theta$), is used to calculate the crystalline size of nanoparticles from X-ray diffraction (XRD) data was utilized to compute the dimensions of the nanoparticles where K is the Scherer coefficient (0.98), D is the average crystal size of nanoparticles, λ is the wavelength of X-ray, β is the full width at half maximum and (θ) is the Bragg's angle corresponding to the (hkl) reflection (Sen et al., 2020). The (111), (200), and (220) set of lattice planes are the ones that correspond to the various Bragg's reflections that copper displays. On the basis of these Bragg reflections, we can conclude that synthesized copper nanoparticles are cubic face-centered and essentially crystalline. The set of lattice planes (111), (200), and (220) were found to be extremely weak and widened relative to each other. According to Bragg's reflection (111), this property demonstrates that biologically nanocrystals are extremely anisotropic and that the nanoparticles are (111) oriented.

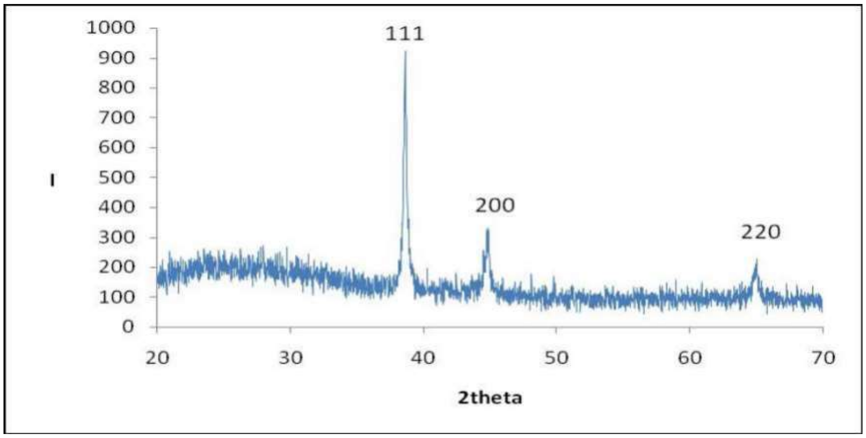


Fig. 4 X-ray diffraction spectrum of copper nanoparticle

FTIR Analysis

FT-IR spectra of the synthesized copper nanoparticles were analyzed with plausible functional groups. FT-IR spectrum of prepared nanoparticles showed intensive bands in the region of 1509-3917 cm^{-1}. The peak showed at 1509 cm^{-1} of aromatic -C=C, at 1608 cm^{-1} of alkene C=C bonds, at 3648 cm^{-1} of alcohol O-H, at 1713 cm^{-1} of carbonyl -C=O, at 3010 cm^{-1} of aromatic C-H.

the nanoparticles showed several distinctive peaks, as depicted in Figure. These peaks were observed in the region of 1509-3917 cm-1. The absorption peak at 1509 cm-1 can be attributed to the presence of aromatic -C=C bonds. Peaks around 1608 cm-1 are associated with alkene C=C bonds, while the peak at 1713 cm-1 is likely due to carbonyl groups of -C=O. The peak observed at 3010 cm-1 may be attributed to aromatic C-H bonds. Additionally, the peak at 3648 cm-1 is likely due to the presence of alcohol -O-H functional groups.

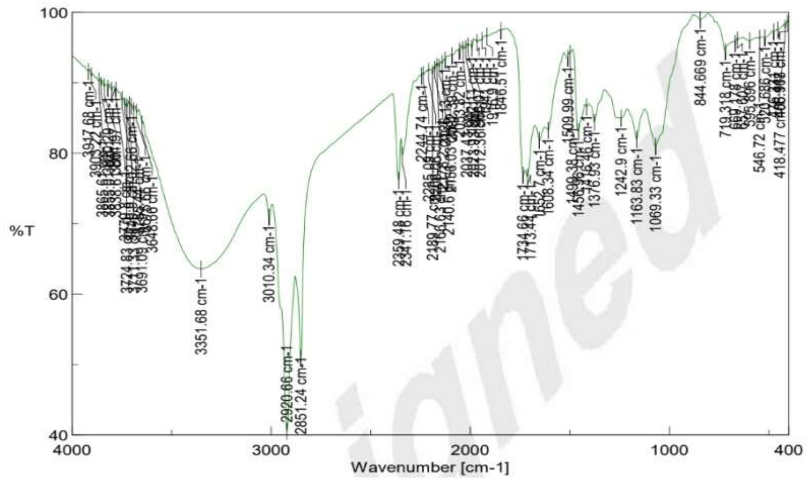


Fig. 5 FTIR absorption spectrum obtained from copper nanoparticles.

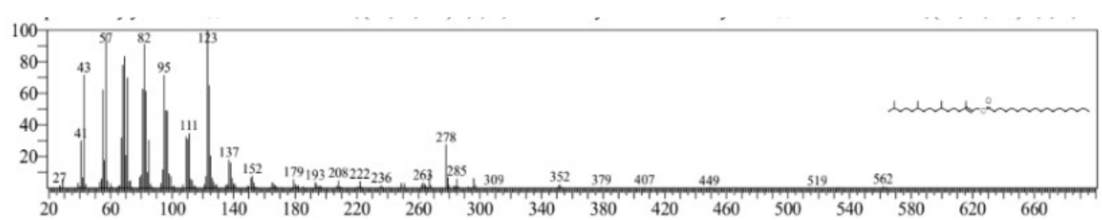
The presence of oxygen atoms may also play a role in facilitating the absorption of these heterocyclic components on the surface of the copper nanoparticles and stabilizing them. The spectra exhibit small shifts and variations in peak amplitudes, which may be explained by the distinct capping species and the degree of coordination between them and the copper nanoparticles' metal surface. FTIR study, taken as a whole, provided valuable new insights into the biomolecules responsible for the stability and reduction of the synthesized copper nanoparticles.

Phytochemical compounds identified by GC-MS analysis:

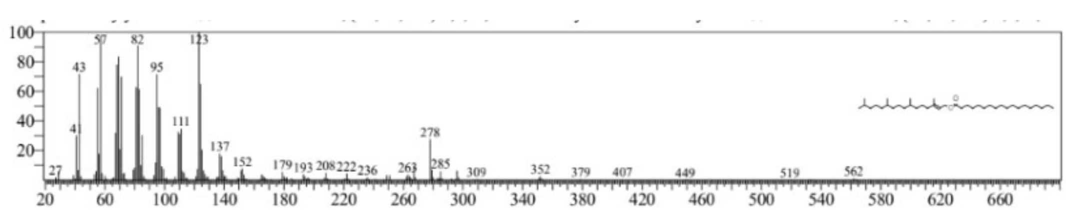
The active principles of the bioactive components of Daucas Carota Linn were identified by GC-MS analysis, together with their molecular formula, molecular weight, retention duration, and peak area percentage. Phytol, Hexadecanoic acid-methyl ester, n-Hexadecanoic acid, Phytyl stearate, and Octadecanoic acid are the main chemical components responsible for their anticancer properties.

Table : Phytochemicals Identified by GC MS Analysis of *Daucus Carota Linn.*

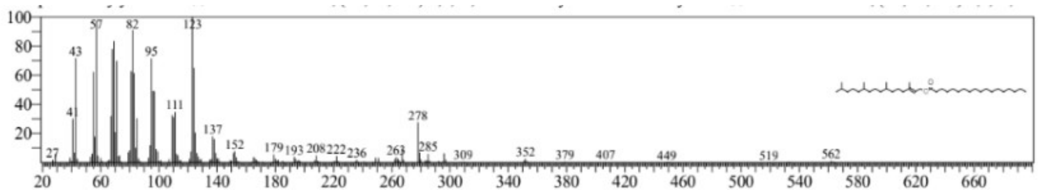
S.No	Compound	Molecular Formula	Molecular Weight	Retention Time	Area %	Peak
1	Naphthalene, 1,2-dihydro-1,1,6 trimethy	C13H16	172	18.743	0.19	157
2	DL-Proline,5-oxo-,ethyl ester	C7H11NO3	157	21.702	0.23	84
3	Phenol, 4-(3-methyl-2 buteny)	C11H14O	162	22.265	0.17	147
4	Decanoyl Chloride	C10H19CLO	190	23.171	0.59	55
5	Beta Carotene	C40H56	536	50.981	0.34	69
6	Lathosterol	C27H460	386	53.999	0.28	255
7	Diosgnine	C27H4203	414	54.922	1.16	139
8	**Phytyl stearate**	**C38H74O2**	**562**	**55.557**	**O.14**	**57**
9	Hydroxy-Beta-damascone	C13H20O2	208	25.659	0.06	69
10	Longifolene	C15H24	204	26.798	0.22	161
11	8-pentadecanone	C15H30O	226	26.962	0.07	57
12	Cinnamic acid, 4-hydroxy-3-methoxy	C31H40O15	652	27.857	0.34	105
13	1-hexadecanol	C16H34O	242	29.732	0.17	55
14	2-pentadecanone, 6,10,14-trimethyl	C18H36O	268	30.826	0.59	58
15	1H-2-benzopyran-1-one,3,4-dihydro-8	C11H12O4	208	30.893	0.73	164
16	2-cyclohexan-1-one,3-(3-hydroxybutyl	C13H22O2	210	31.003	0.10	109
17	10-Nanodecanone	C19H38O	282	31.503	0.07	155
18	**Hexadecanoic acid, methyl ester**	**C17H34O2**	**270**	**32.580**	**0.24**	**74**
19	9,12,15-octadecatrienoic acid,(Z,Z,Z)-	C18H30O2	278	33.248	4.73	79
20	**n-Hexadecanoic- acid**	**C16H32O2**	**256**	**33.248**	**12.85**	**73**
21	9,12-octadecadienoic acid (Z,Z)-methyl	C19H34O2	294	35.958	0.25	67
22	9,12,15-octadecatrienoic acid, methyl	C21H26O2	292	36.081	0.41	79
23	**Phytol**	**C20H40O**	**295**	**36.395**	**8.61**	**71**
24	9,12,15- octadecadienoic acid (Z,Z,Z)	C18H30O2	278	37.373	17.34	
25	**Octadecanoic acid**	**C18H36O2**	**284**	**37.672**	**0.37**	**73**
26	2-butenoic acid,2-methyl-,2-(acetyloxy-	C27H38O8	490	39.655	0.87	
29	Octacosanol	C28H58O	410	43.135	0.51	55
30	Cyclohexane, bromo	C6H11br	162	43.616	1.09	83
31	Cyclohexane, bromo	C6H11br	162	43.720	1.24	83
32	1-hexacosanol	C26H54O	382	46.343	0.44	97
33	Octacosanol	C28H58O	410	49.435	0.26	55
34	Diosgnine	C27H42O3	414	51.645	0.47	139
35	Dl-alpha-tocopherol	C29H50O2	430	52.504	0.55	430
36	**Phytyl stearate**	**C38H74o2**	**562**	**52.730**	**0.27**	**57**



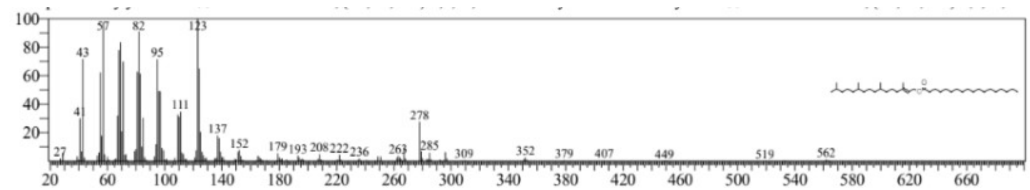
Compound:1



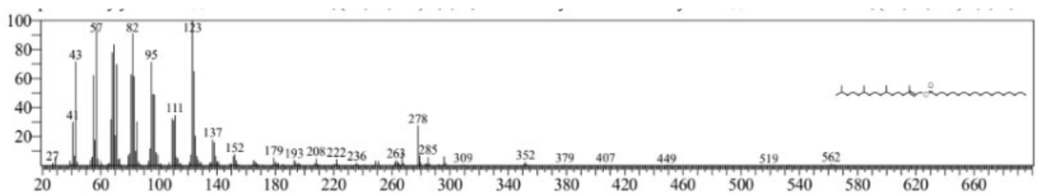
Compound: 2



Compound: 3



Compound: 4



Compound: 5

Anticancer activity

The anticancer efficacy of plant extract and produced copper nanoparticles was performed on Hep G-2 cancer cell line. On comparison to standard drug (Adriamycin), which had a GI50 value of <10 µg/mL, the ***Daucas Carota Linn*** leaves extract and copper nanoparticles, having GI50 values of >80 µg/mL and >80 µg/mL, respectively, shown increased activity on the Hep G-2 cancer cell line (Table).

Table: Anticancer potential of Plant extract, copper nanoparticles and Standard Drug

S. No.	Hep-G2	LC50	TGI	GI50*
1.	A1	NE	NE	>80
2.	A2	NE	NE	>80
3.	ADR	16.6	<10	<10

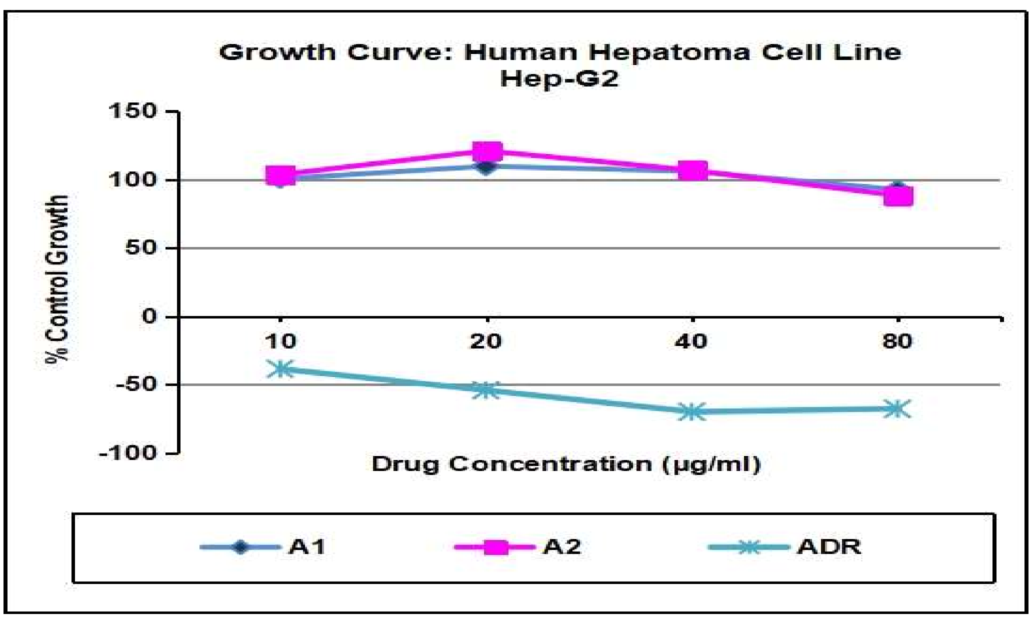


Fig 3 Drug concentrations (µg/ml) calculated from graph

CONCLUSION

Copper nanoparticles were synthesized by an economical, efficient and environmentally friendly method green synthesis of silver nanoparticles by *D carota* L. leaf extract (DCLE).

Analytical techniques like UV–Vis spectrophotometer, XRD, FT-IR, and SEM techniques had been used to study the formation of copper nanoparticles. Spherical shapes of the synthesized copper nanoparticles were determined from SEM micrographs, and face-centered cubic phase with a mean particle size distribution. SBR assay was used to evaluate the effectiveness of the drug for anticancer activity. With GI_{50} values of >80µg/ml, >80µg/ml, and <10µg/ml for plant extract, copper nanoparticles and to standard drug (Adriamycin), respectively the study showed increased activity of copper nanoparticles against standard drug.

FUTURE PERSPECTIVES

The study showed that *Daucus carota* extracts could be an eco-friendly reducing agent for functional nanoparticle synthesis. The plant-extract based nanoparticles showed the advantages of cheap, environmentally friendly, self-capping and stabilization, which would be useful for practical application in anticancer management.

REFERENCES

1. Zhou W, Apkarian R, Wang ZL, Joy D. Fundamentals of scanning electron microscopy (SEM). Scanning microscopy for nanotechnology: techniques and applications. 2007:1-40.
2. Wang M, Thanou M. Targeting nanoparticles to cancer. Pharmacological research. 2010 Aug 1;62(2):90-99.
3. Vijayakumar. G , Kesavan. H , Kannan. A , Arulanandam. D , Kim.J.H , Kwang Jin Kim.K.J , Song.H.J , Kim. H.J and Rangarajulu. S.K, “Phytosynthesis of Copper Nanoparticles Using Extracts of Spices and Their Antibacterial Properties,” MDPI, 2021 ,P-1-18.
4. Valsalam. S, Paul. A, Arasu. M.V, Al-Dhabi. N.A., Ghilan. A.K.M, Kaviyarasu. K, Ravindran. B, Chang. S.W, Arokiyaraj. S, “Rapid biosynthesis and characterization of silver nanoparticles from the leaf extract of Tropaeolum majus L. and its enhanced in vitro antibacterial, antifungal, antioxidant and anticancer properties,” Journal of Photochemistry & Photobiology, B: Biology,2018,P-33.
5. Thakur. S, Rai. R, Sharma. S, “Study The Antibacterial Activity of Copper Nanoparticles Synthesised Using Herbal Plant Leaf Extract,”International journal of Biotechnology and Reaserch, 2014, 4:(5):P-21-34.
6. Tahvilian. R, Zangeneh. M.M ,Falahi. H, Sadrjavadi. K, Jalalvand.A.R, Zangeneh.A, “Green synthesis and chemical characterization of copper nanoparticles using Allium saralicum leaves and assessment of their cytotoxicity, antioxidant, antimicrobial, and cutaneous wound healing properties”,Wiley, Applied orangometalli chemistry, 2019,P-16.
7. Tahvilian S, Kuban JD, Yankelevitz DF, Leventon D, Henschke CI, Zhu J, Baden L, Yip R, Hirsch FR, Reed R, Brown A. The presence of circulating genetically abnormal cells in blood predicts risk of lung cancer in individuals with indeterminate pulmonary nodules. BMC Pulmonary Medicine. 2023, 5;23(1):193.
8. Tabassum M, Alam MA, Mostofa S, Bishwas RK, Sarkar D, Jahan SA. Synthesis and crystallinity integration of copper nanoparticles by reaction medium. Journal of Crystal Growth. 2024,15;626:127486.
9. Surdu VA, György R. X-ray diffraction data analysis by machine learning methods—a review. Applied Sciences. 2023, 4;13(17):9992.
10. Sundaram. C.S, Sivakumar. J, Kumar. S.S, Ramesh .P.L.N, Zin. T, Rao. US.M, “Antibacterial and Anticancer Potential of Brassica Oleracea Var Acephala using Biosynthesised Copper Nanoparticles,” Med J Malaysia, 2020,65:(6),P-677-684.
11. Sen SK, Barman UC, Manir MS, Mondal P, Dutta S, Paul M, Chowdhury MA, Hakim MA. X-ray peak profile analysis of pure and Dy-doped α-MoO3 nanobelts using Debye-Scherrer, Williamson-Hall and Halder-Wagner methods. Advances in Natural Sciences: Nanoscience and Nanotechnology. 2020,15;11(2):025004.
12. Sarkar. S and Kotteeswaran. V, “Green synthesis of silver nanoparticles from aqueous leaf extract of Pomegranate (Punica granatum) and their anticancer activity on human cervical cancer cells”, Advances in Natural Sciences: Nanoscience and Nanotechnology, 2017,P- 11.
13. Rocha FS, Gomes AJ, Lunardi CN, Kaliaguine S, Patience GS. Experimental methods in chemical engineering: Ultraviolet visible spectroscopy—UV-Vis. The Canadian Journal of Chemical Engineering. 2018;96(12):2512-7.
14. Nazneen S, Sultana S. Green Synthesis and Characterization of Cissus quadrangularis. L stem mediated Zinc Oxide Nanoparticles. Plant Science Archives. 2024;05.
15. Mandrich L, Esposito AV, Costa S, Caputo E. Chemical composition, functional and anticancer properties of carrot. Molecules. 2023 Oct 19;28(20):7161.
16. Mali. S.C, Dhaka. A, Githala. C.K, Trivedi.R, “Green synthesis of copper nanoparticles using Celastrus paniculatus Willd. leaf extract and their photocatalytic and antifungal properties,” Elsevier, 2020,P-1-9.
17. Liu H, Wang G, Liu J, Nan K, Zhang J, Guo L, Liu Y. Green synthesis of copper nanoparticles using Cinnamomum zelanicum extract and its applications as a highly efficient antioxidant and anti-human lung carcinoma. Journal of Experimental Nanoscience. 2021,15;16(1):410-23.
18. Fareed N, Nisa S, Bibi Y, Fareed A, Ahmed W, Sabir M, Alam S, Sajjad A, Kumar S, Hussain M, Syed A. Green synthesized silver nanoparticles using carrot extract exhibited strong antibacterial activity against multidrug resistant bacteria. Journal of King Saud University-Science. 2023,1;35(2):102477.

19. Ejidike IP, Clayton HS. Green synthesis of silver nanoparticles mediated by Daucus carota L.: antiradical, antimicrobial potentials, in vitro cytotoxicity against brain glioblastoma cells. Green Chemistry Letters and Reviews. 2022, 3;15(2):298-311.
20. Devi. R.S, Jeevitha. M, Preetha.S and Rajeshkumar. S, "Free Radical Scavenging Activity of Copper Nanoparticles Synthesized from Dried Ginger", Journal of Pharmaceutical Research International, 2020,Vol:32 :19:P-7.
21. Debela. D.T, Seke G.Y., Muzazu. S.K.Y, Heraro. K.D, Ndalama. M.T, Mesele. B.W, Haile.D.C Kitui. S.K., and Manyazewal. T, "New approaches and procedures for cancer treatment: Current perspectives", SAGE open medicine, (2021),Vol:9:P-1-10.
22. Davies TE, Li H, Bessette S, Gauvin R, Patience GS, Dummer NF. Experimental methods in chemical engineering: Scanning electron microscopy and X-ray ultra-microscopy—SEM and XuM. The Canadian Journal of Chemical Engineering. 2022, 100(11):3145-59.
23. Dashtizadeh. Z, Kashi. F. J , Ashrafi. M "Phytosynthesis of copper nanoparticles using Prunus mahaleb L. and its biological activity " Materials Today Communicaton, 2021, Vol: 27 : P- 8.
24. Das. P.E,A-Yousef. I.A, Majdalawieh. A.F, Narasimhan. S and Poltronieri. P,"Green Synthesis of Encapsulated Copper Nanoparticles Using a Hydroalcoholic Extract of Moringa oleifera Leaves and Assessment of Their Antioxidant and Antimicrobial Activities", Multidisciplinary Digital Publishing Institute. Molecule, 2022,Vol:25:P-17.
25. Bunaciu AA, UdriŞTioiu EG, Aboul-Enein HY. X-ray diffraction: instrumentation and applications. Critical reviews in analytical chemistry. 2015, 2;45(4):289-99.
26. Anaja. D, Kavitha, S,"A Study of phytochemical, antioxidant activity and fTIR analysis of Daucus carota (thunb) A. Henry" European journal of pharmaceutical and medical research, 2016, P-390-394.