JETIR.ORG ISSN: 2349-5162 | ESTD Year : 2014 | Monthly Issue

JOURNAL OF EMERGING TECHNOLOGIES AND INNOVATIVE RESEARCH (JETIR)

An International Scholarly Open Access, Peer-reviewed, Refereed Journal

SYNTHESIS, CHARACTERIZATION AND PHYTOCHEMICAL ANALYSIS OF SILVER NANOPARTICLES FROM STEM OF *CISSUS QUADRANGULARIS* L.

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Abstract: The arena of green synthesis of nanoparticles has been vastly explored for the past two decades due to its enhanced biological activity and non-toxic method of preparation. The extract from the stem of the plant *Cissus quadrangularis* L belonging to the family Vitaceae is a complex concoction of various phytochemicals responsible for several biological activities. The silver nanoparticles synthesized using the plant extract exhibit enhanced properties compared to their original constituents. The preparation of nanoparticles using silver metal is expected due to its low level of toxicity for humans and high antibacterial properties, thus making it ideal for pharmacological studies. The current paper deal with the synthesis, characterization and phytochemical analysis of silver nanoparticles using the stem extract of *Cissus quadrangularis* L. The qualitative screening of the phytochemicals reports the presence of phenols, alkaloids, saponins and terpenoids. The GC-MS analysis revealed vital compounds that possess a broad spectrum of biological functions. The FTIR spectral studies provided detailed information regarding the functional groups present in the synthesized silver nanoparticles. The characterization study gave a detailed insight into the structure, size and stability of silver nanoparticles prepared from the stem of extract of *Cissus quadrangularis* L.

Keywords: AgNPs, *Cissus quadrangularis* L., Phytochemicals, GC-MS and FTIR.

I. INTRODUCTION

Natural products have been utilized to treat numerous ailments and illnesses since antiquity and in folklore (Dias et al., 2012). *Cissus quadrangularis* L, commonly known as Vitis quadrangularis Wall, is a perennial climber in the Vitaceae family that grows best in a warm tropical region (Garima Mishra et al., 2010).In Ayurveda, the stem of *Cissus quadrangularis* L is used as an alternative, anthelmintic, dyspeptic, digestive, tonic, and analgesic in treating eye and ear ailments, irregular menstruation, asthma, and back and spinal complaints (Eswaran et al., 2012).

Nanoparticles are substances with diameters between 1 to 100 nanometers with remarkable potential than the bigger constituents from which they are prepared. The nanoparticle is the most fundamental component in producing a nanostructure since it deals with matter on the scale of one billionth of a metre (Bawazeer et al., 2021). In the synthesis of nanoparticles, plant extracts may act as both reducing and stabilizing agents. The selection of silver for synthesizing the metal nanoparticles is predominantly due to its antibacterial and pharmaceutical properties. The characterization of the synthesized plant-based nanoparticles can be achieved through a set of spectral analyses like the SEM, XRD, Zeta Potential, UV- Vis and FTIR. These provide a detailed insight into the nanoparticles' chemistry and mode of action. Therefore, the nanoparticles can interact with the cells and tissues at the molecular level, thus providing a high degree of specificity in functioning (Saini et al., 2010).

Because of their cost-effectiveness, compatibility for medical and pharmaceutical applications, and large-scale commercial production, AgNPs have prospective uses in the biomedical arena (Mallikarjuna et al., 2010). Plant-produced nanoparticles are more stable and have a higher rate of synthesis than microorganism-produced nanoparticles, and they have a greater variety of shape and size (Iravani, 2011).

II. MATERIALS AND METHODS

2.1 Preparation of Plant extract

10gm of the fresh stem of *Cissus quadrangularis* L. is cut and thoroughly washed under running water to remove surface contaminants and dirt. It is then sterilized with absolute alcohol for complete disinfection. The stem is cut into small pieces, and 100ml of distilled water is added to it in a beaker and is boiled for approximately 20 minutes. After the plant extract cools down, a filtrate is obtained by filtering the extract through Whatman filter paper No.1. Store the filtrate at 4 degrees Celsius for the synthesis of silver nanoparticles.

2.2 Preparation of Silver Nanoparticles

0.1 M standard aqueous solution of Silver nitrate ($AgNO_3$) was prepared (17gm of silver nitrate in 1000ml of distilled water). This solution is used in the synthesis of nanoparticles. This is followed by adding 5ml of plant extract of *Cissus quadrangularis* L. in 45ml standard 0.1M $AgNO_3$ solution in an Erlenmeyer flask. The process is carried out at room temperature, and the change in colour from white to dark brown indicates the formation of silver nanoparticles (Fig. 1).

The solution is centrifuged at 10,000rpm and rinsed with 70% alcohol to separate the nanoparticles from the aqueous solution.

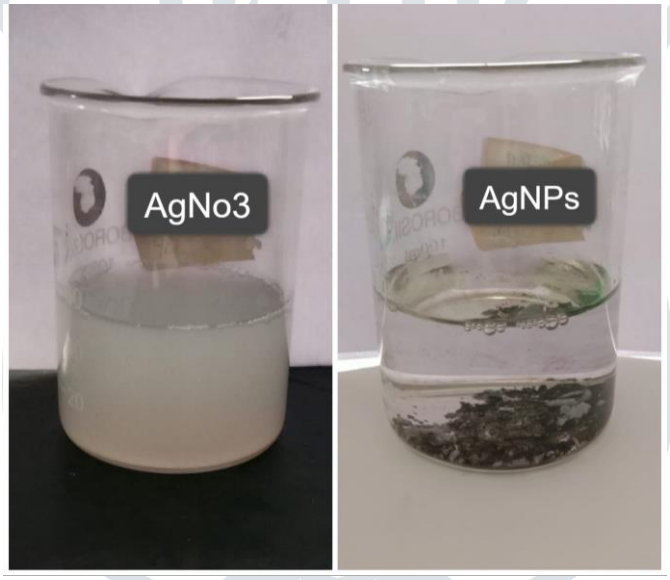

Figure:1 Bioreduction of $AgNO_3$ into AgNPs from the stem extract of *C. quadrangularis*

2.3 Phytochemical Analysis

A series of chemical assays are used to screen the bioactive components of silver nanoparticles formed from stem extract of *Cissus quadrangularis* L. The tests are carried out according to Trease and Evans' established protocols (2002).

2.3.1 Test for Alkaloids: 3ml of the extract was combined with 3ml of 1% HCL in a hot water bath. The liquid was then divided into two test tubes, each receiving 1ml. 1 ml of Dragendroff's reagent was introduced in the first test tube. The appearance of orange-red precipitate was a positive indication.

2.3.2 Test for Flavonoids: A strong yellow hue was obtained by mixing 2 ml of each extract with a few 20 percent sodium hydroxide drops. The yellow tint vanished once a few drops of 70% dilute hydrochloric acid was introduced. The presence of flavonoids in the sample is shown by the production and disappearance of yellow colour. Evans and Trease (1983 and 1996).

2.3.3 Test for Saponins: First, 5ml of distilled water was used to dilute 0.5ml of plant extract. For a few minutes, the suspension was vigorously shaken. The foam developed and lasted for almost 10 minutes, confirming the presence of saponins.

2.3.4 Test for Steroids: 1 ml of plant extract was dissolved in 10 ml of $CHCl_3$, and an equal volume of concentrated sulphuric acid was added to the test tubes through the sidewalls. The solution's top layer becomes red, while the sulphuric acid layer takes on a yellowish colour with green fluorescence. This indicates the presence of steroids.

2.3.5 Test for Terpenoids: 1 ml concentrated sulphuric acid was added to 1 ml crude extract, and the mixture was heated for 2 minutes. The grey tint would indicate the presence of terpenoids.

2.3.6 Test for Quinines: 1 ml extract was added to 1 ml of 1 percent NaOH and thoroughly mixed. The appearance of bluish-green or red colour indicates the presence of quinines.

2.3.7 Test for Phenols: 5ml distilled water was added to 1gm of extract, and the solution was then treated with a 5 percent ferric chloride solution. The presence of phenols is confirmed by forming a dark green hue.

2.4 GC-MS Analysis

The GC-MS analysis of the silver nanoparticle stem extract was done at the Central Analytical Facility, University College of Technology, Osmania University using the standard GC-MS model.

The GC-MS analysis is performed using a SHIMADZU model GC-2010, MS-QP2010 equipment. Helium is employed as the carrier gas, with a 1 ml/min flow rate. The HP5 column is used, which has a 30 mm length, a 0.32 mm internal diameter, a 0.25 mm film thickness, and a temperature range of -60 to 325 degrees Celsius (350 degrees Celsius). The film GC ran 35 minutes in total. The oven temperature was increased at 8 degrees Celsius per minute from 70 to 280 degrees Celsius. The sample volume injected through the injector is 1µl. The MS was performed at 70 eV. By comparing the spectrum of unknown chemicals to the spectrum of known compounds in their library, the name, molecular weight, and structure of unknown compounds were determined.

2.5 Characterization of AgNPs of stem extract of *Cissus quadrangularis*

2.5.1 Fourier transform infrared spectrophotometer Analysis

The functional groups included in the plant extract components are identified using a Fourier transform infrared spectrophotometer (FTIR). The wavelength of absorbed light, which is characteristic of the chemical bond, is given in the annotated spectrum. Analyzing the infrared absorption spectra can reveal the chemical bonds.

For FTIR analysis, the silver nanoparticles were centrifuged for 30 minutes at 12,000 rpm. Then the pellets were rinsed three times with 25 ml of de-ionized water (Muthukrishnan et al. 123). For the study, the dried powder of the silver nanoparticles of *Cissus quadrangularis* L. is used. A transparent sample disc is created by encapsulating 10mg of the sample in 100mg of KBr pellet (Perkin Elmer). The *Cissus quadrangularis* L. stem nanoparticles extract was then put into an FTIR spectroscope.

2.5.2 UV-Vis Spectroscopy

The silver nanoparticles of *Cissus quadrangularis* L. were subjected to UV-Visible spectrophotometric examination using a UV-Visible spectrophotometer (UV-8400 Series) with a 1.0 nm slit width. The test was carried out using UV and visible light with wavelengths ranging from 190.00 to 1100.00 nm.

The extract is first centrifuged at 3000 rpm for around 10 minutes before filtering using Whatman filter paper No.1. After that, the sample is diluted in a 1:10 ratio using the same solvent.

2.5.3 SEM Analysis

Scanning electron microscopy (SEM) analysis of synthesized AgNPs was performed on the Hitachi S-3700N SEM machine.

The powder of AgNPs was sonicated for 5 minutes prior to analysis, and a drop of properly diluted sample was transferred onto a carbon-coated copper grid. This has a magnifying effect to determine the shape and size of the nanoparticles.

2.5.4 Zeta Potential Analysis

The surface charge property and stability of nanoparticles are measured by the Zeta Potential, which reflects the electrical potential of the nanoparticles. The analysis was carried out at Horiba Sz-100 with the conductivity set at 9.606 mS/cm, and the zetametry readings were performed at 2.2V and 25.1 °C.

Nanoparticles with a zeta potential greater than (±)10 mV are neutral in suspension because the surface charge prevents the particles from aggregating. Highly cationic nanoparticles have more than +30 mV zeta potentials, while strongly anionic nanoparticles have less than -30 mV.

2.5.5 Particle size of Nanoparticles

The size of the particles was assessed using the Microtrac S3500 particle size analyzer and a dynamic light scattering approach. Brownian nanoparticles in colloidal solutions are commonly measured using DLS with a 90-degree light scattering angle at a temperature of 25°C.

2.5.6 XRD Analysis

The crystalline structure of the purified SNPs was examined using X-ray diffraction analysis. A thin coating of the sample was placed on a glass slide in the technique for preparing samples for XRD by dropping 100 µL of sample and drying for 30 minutes. The XRD pattern was recorded using Shimadzu XRD-7000 diffractometer with a voltage of 40 kV at a 30 mA current strength. The diffracted intensities were measured at 2θ angles and ranged from 20° to 50°.

Scherrer Formula

$Dp = (0.94 \times \lambda) / (\beta \times \cos\theta)$

Where, Dp = Average Crystallite size,

β = Line broadening in radians,

θ = Bragg angle,

λ = X-Ray wavelength

The FWHM (Full width at half maximum)is the distance between the curve points at the peak half maximum level. On a data graph, a vertical line is drawn from the peak maximum to the baseline.Multiply the length of the line by two to find the line's centre.

III. RESULTS AND DISCUSSIONS

3.1 Phytochemical Analysis

The phytochemical analysis of the synthesized silver nanoparticles of *Cissus quadrangularis* L. stem revealed the presence of secondary metabolites like alkaloids, saponins, terpenoids and phenols (Table 1).

Table 1: Phytochemicals present in the AgNPs of the stem of *Cissus quadrangularis* L

S.No.	Phytochemical	SNP Extract
1.	Test for Alkaloids	Present
2.	Test for Flavanoids	Absent
3.	Test for Saponins	Present
4.	Test for Steroids	Absent
5.	Test for Terpenoids	Present
6.	Test for Quinones	Absent
7.	Test for Phenols	Absent

3.2 GC-MS Analysis

The GC-MS spectral analysis chromatogram revealed peaks of a number of compounds from the GC fractions of the silver nanoparticle extract of the *Cissus quadrangularis* L. plant stem. The observations showed the presence of bioactive compounds like Thiazolidinedione, Cycloheptatrienone, Ethylcyclopentane, 8-Nonynoic acid, cis-1,2-Dimethylcyclohexane, Gentisic acid (tms), Mandelic acid di(tert-butyldimethylsilyl) and 6-Acetyl-.beta.-d-mannose. Fig. 2 shows the chromatogram, and table 2 depicts the names of the chemical compounds, their molecular formula, molecular weight, peak area percentage and their retention time. The majority of these compounds are reported to have antimicrobial, anti-mycobacterium, anticonvulsant, antiviral, anticancer, anti-inflammatory and antioxidant properties. Gentisic acid (GA) is a phenolic acid that has been linked to anti-inflammatory, antigenotoxic, hepatoprotective, neuroprotective, antimicrobial, and mainly antioxidant effects on human health (Abedi et al., 2020).

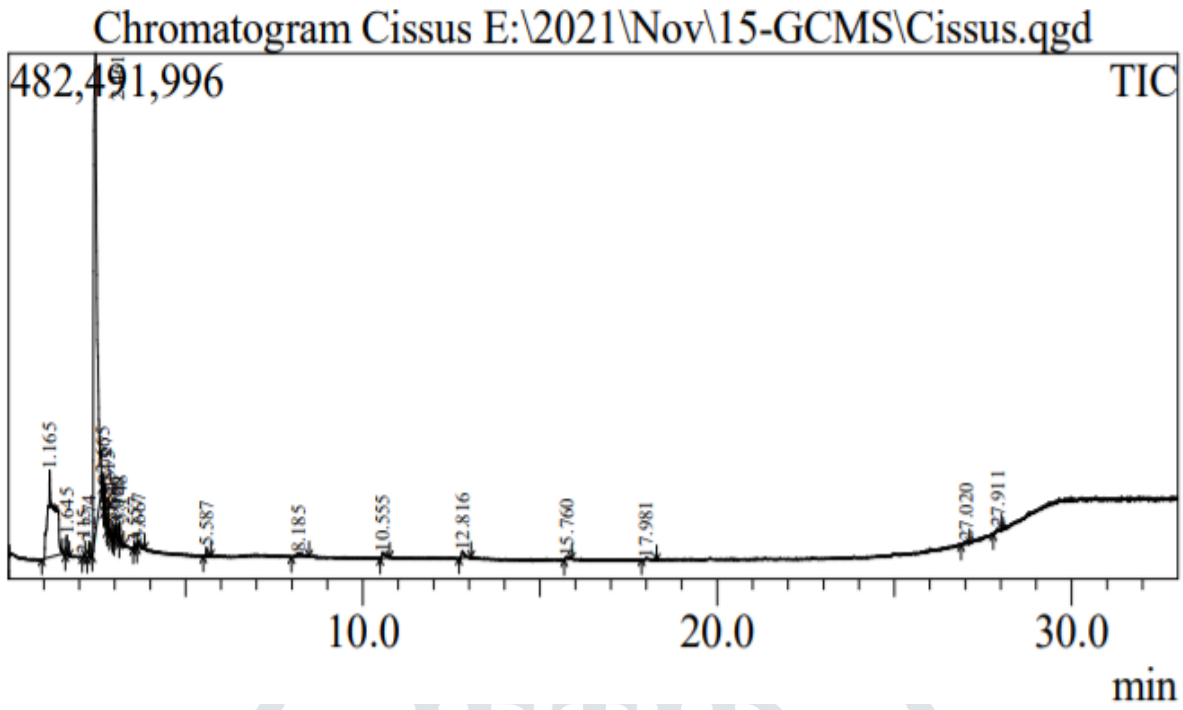

Figure:2 GC-MS chromatogram of the AgNPs of the stem of *Cissus quadrangularis* L

Table: 2 Phytocompounds from GC-MS analysis of AgNPs of the stem of *Cissus quadrangularis* L

S.No.	RT Time	Name of the Compound	Formula	MW	Peak Area(%)	Nature of Compound
1.	1.165	Thiazolidinedione	$C_3H_3NO_2S$	117	0.45	Heterocyclic Compounds
2.	1.64	Cycloheptatrienone	C_7H_6O	106	0.49	Tropolones
3.	2.115	5-Hexenoic acid, 5-methyl-	$C_7H_{12}O_2$	128	0.02	Fatty acid
4.	2.27	Norbornane	C_7H_{12}	96	0.31	Saturated Hydrocarbon
5.	2.461	8-Nonynoic acid	$C_9H_{14}O_2$	154	66.48	Fatty acid
6.	2.725	cis-1,2-Dimethylcyclohexane	C_8H_{16}	112	0.88	Saturated Hydrocarbon
7.	2.885	Ethylcyclopentane	C_7H_{14}	98	0.10	Saturated Hydrocarbon
8.	5.585	Octamethylcyclotetrasiloxane	$C_8H_{24}O_4Si_4$	296	0.33	Organosilicon compound
9.	8.185	Gentisic acid (tms)	$C_{16}H_{30}O_4Si_3$	370	0.85	Fatty acid ester
10.	10.55	Cyclohexasiloxane, dodecamethyl-	$C_{12}H_{36}O_6Si_6$	444	0.78	Organosilicon compound
11.	15.75	Cyclooctasiloxane, hexadecamethyl-	$C_{16}H_{48}O_8Si_8$	592	0.38	Organosilicon compound
12.	17.95	Mandelic acid di(tert-butyldimethylsilyl)-	$C_{20}H_{36}O_3Si_2$	380	0.12	Organosilicon compound
13.	27.91	6-Acetyl-.beta.-d-mannose	$C_8H_{14}O_7$	222	0.14	Carbohydrates

3.3 Fourier transforms infrared spectrophotometer Analysis

The FTIR study aimed to determine biomolecules that get trapped on the surface of nanoparticles and are responsible for the reduction of metal salts to their corresponding nanoparticles (Gujral, 2014). The Fourier transforms infrared spectrophotometer (Fig. 3) reveals the types of chemical bonds present in the silver nanoparticle extract of the *Cissus quadrangularis* L. plant stem. The functional groups of the elements were separated on the basis of the ratio of the peak when the extract was passed through the FT-IR (Pakkirisamy, 2017). Table 3 depicts the occurrence of the alcohols, carboxylic acid, thiocyanates, isothiocyanates, allenes, alkenes, aldehydes, aromatic,

nitro, fluoro and halo compounds at their respective frequency range of the peaks. Some functional groups may vanish due to bioreduction during the formation of AgNPs.

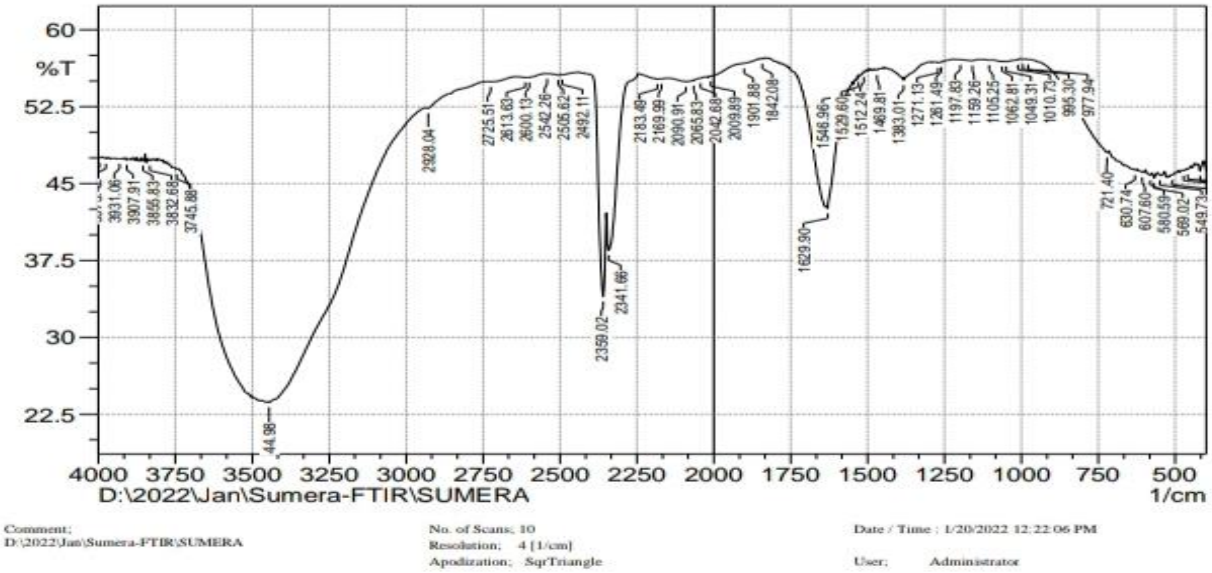

Figure:3 FTIR Spectrum of AgNPs of the stem of *Cissus quadrangularis* L

Table:3 FTIR peak values and functional groups of AgNPs of the stem of *Cissus quadrangularis* L

Peak	Compound class	Group
3444.98	Alcohol	O-H stretching/Strong bond
2928.04-2505.62	carboxylic acid	O-H stretching/ Strong
2169.99	thiocyanate	S-CΞN stretching/ strong
2090.91-2009.89	isothiocyanate	N=C=S stretching/ strong
1901.88	allene	C=C=C stretching/ Medium
1842.08	aromatic compound	C-H bending/ weak
1629.9	alkene	C=C stretching/ medium
1546.96-1512.24	nitro compound	N-O stretching/ Strong
1383.01	aldehyde	C-H bending/ Medium
1271.13-1010.73	fluoro compound	C-F stretching/ Strong
977.94	alkene	C=C bending/Strong
721.4	alkene	C=C bending/strong
630.74-549.73	halo compound	C-Br stretching/ Strong

3.4 UV-Vis Spectroscopy

One of the most straightforward approaches for determining the production and stability of gold and silver nanoparticles in an aqueous solution is the UV-Vis spectroscopy technique (Gujral,2014). The peaks in the UV-Visible spectroscopy spectrum (Fig. 4) depict the absorbance of the synthesized nanoparticles at the respective wavelength. The UV-Vis spectrum profile was chosen at a wavelength of 190nm to 1100nm due to the clarity of the peaks and a suitable baseline. The maximum absorbance of 4.000 was shown at a wavelength of 210 -214 nm, and the least absorbance of 0.172 at 1091 nm (Table 4).

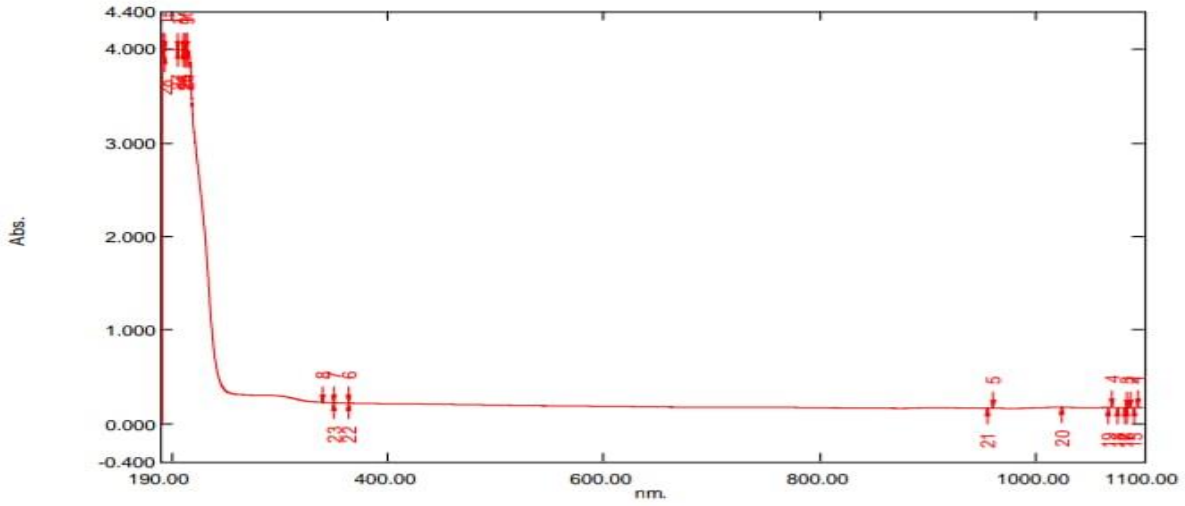

Figure:4 UV-VIS spectrum of AgNPs of the stem of *Cissus quadrangularis* L

Table:4 UV-VIS spectrum peak values of AgNPs of the stem of *Cissus quadrangularis* L

S.No.	Wavelength (nm)	Absorbance
1.	1095.10	0.177
2.	339.70	0.231
3.	214.70	4.000
4.	212.10	4.000
5.	210.50	4.000
6.	206.20	4.000
7.	193.90	4.000
8.	191.30	4.000
9.	1091.20	0.172
10.	349.90	0.226
11.	214.20	3.989
12.	205.70	3.995
13.	192.70	3.930

3.5 SEM Analysis

Surface deposited silver nanoparticles may be seen clearly at a higher magnification in *C.quadrangularis* extract treated with silver nitrate. The images of the Scanning electron microscopy (Fig. 5) portray that the silver nanoparticles of the *Cissus Quadrangularis* L. stem were uniform in size and the shape was found to be spherical. The nanoparticles appear to be aggregated and evenly dispersed. The diameter of the synthesised nanoparticles ranged between 80 - 129 nm.

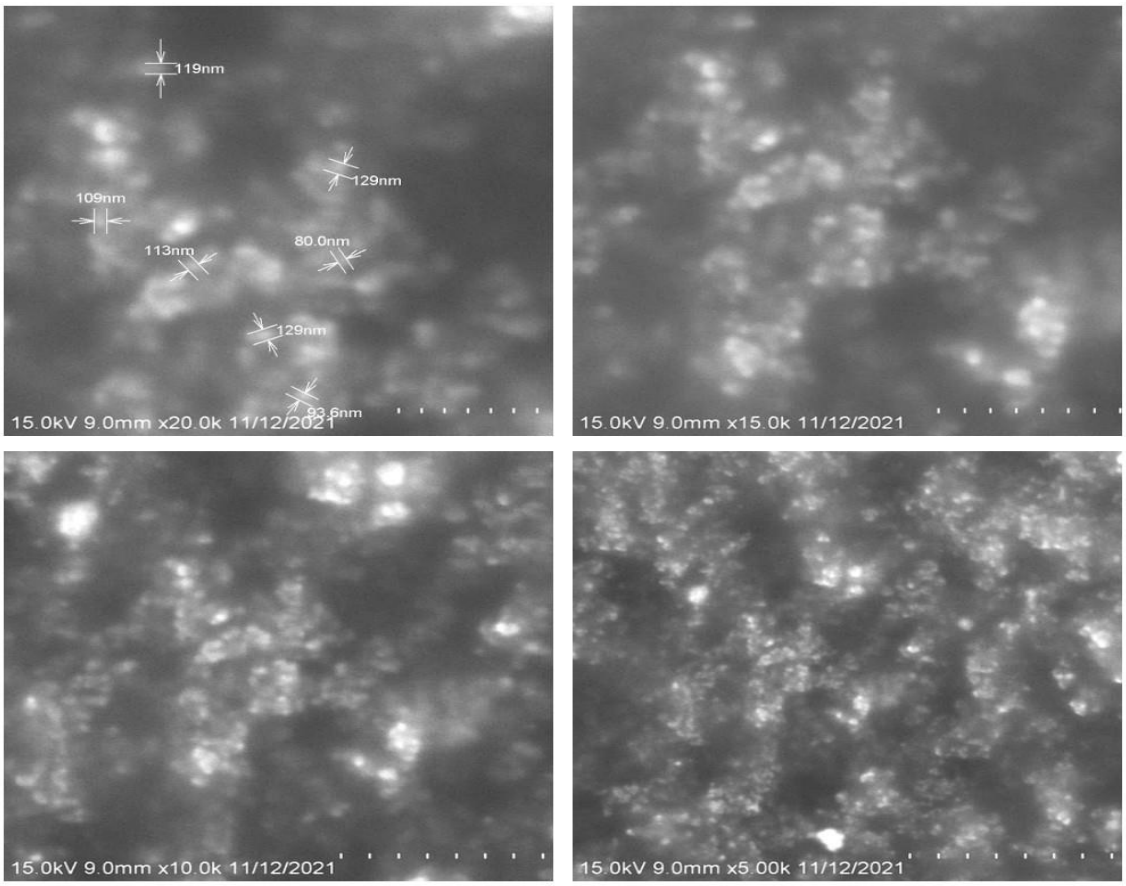

Figure:5 SEM Micrographs showing the size of AgNPs of the stem of *Cissus quadrangularis* L

3.6 Zeta Potential Analysis

The zeta potential and electrophoretic mobility mean of the zeta potential of synthesized AgNPs of *Cissus quadrangularis* L are shown in table 5. The electrophoretic mobility mean was determined to be 0.000044 cm2/Vs, and the zeta potential was found to be 5.7mV. Fig. 6 depicts the linear graph with the zeta potential (mV) values on the x-axis and the intensity (a.u.) on the y-axis.

Table:5 Zeta Potential and electrophoretic mobility mean of AgNPs of the stem of *Cissus quadrangularis* L

Peak No.	Zeta Potential	Electrophoretic Mobility
1	5.7 mV	0.000044 cm2/Vs
2	--- mV	--- cm2/Vs
3	--- mV	--- cm2/Vs

Zeta Potential (Mean) : 5.7 mV

Electrophoretic Mobility mean : 0.000044 cm^2/Vs

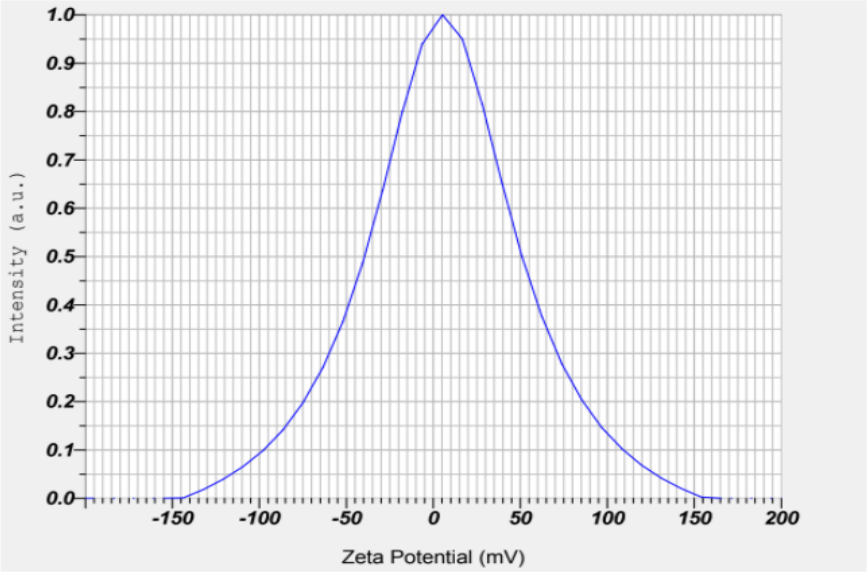

Figure:6 Graph of the Zeta Potential of AgNPs of the stem of *Cissus quadrangularis* L

3.7 Particle size Distribution of Nanoparticles

Particle size can be resolved by measuring the random fluctuations in the intensity of light scattered from a suspension or solution. Fig. 7 shows the number of frequency histograms of particle size data on a linear scale. The smooth curve created by the histogram is a legitimate size-frequency curve if enough particles are counted, and the size interval is at least 10. Hundreds of particles should be measured to get statistically reliable mean size data. The Dynamic Light Scattering (DLS) results depicted the mode value of 483.0 nm and the mean value of 665.1 nm (Table 6). The nanoparticles' polydispersity(PI) index was 10.496, which indicates the aggregation of the particles.

Table:6 Details of Particle size

Peak No	1
S.P.Area Ratio	1.00
Mean	665.1 nm
Median	564.7 nm
Mode	483.0 nm
Standard deviation	343.7 nm
Z-average	926.2 nm
PI	10.496
Count	83 KCPS
Minimum	5.0 nm
Maximum	90.0 nm

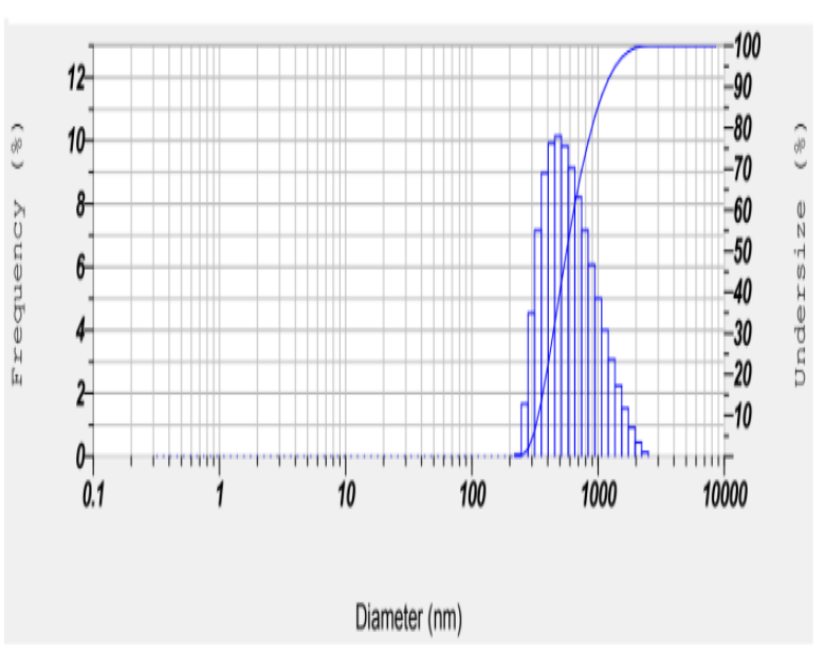

Figure 7 Particle Size Distribution

3.8 XRD Analysis

X-ray crystallography revealed the crystalline nature of nanoparticles. Figure 8 depicts the diffraction pattern of the synthesized nanoparticles, showing various peaks corresponding to the intensity or counts. The XRD patterns can be used to check the nature of materials by looking at the nature of Bragg's peaks that occur in the pattern. Silver nanoparticles had four peaks in their XRD pattern at 2θ corresponding to (35.27) and (32.88) planes of standard XRD peaks of silver crystals. If the peak is broadly humped, the material will be amorphous with short-range ordering. However, the sharp peaks of the diffractogram are an indication of the crystalline nature. Table 7 encloses all the peaks' FWHM, crystalline size, and D values.

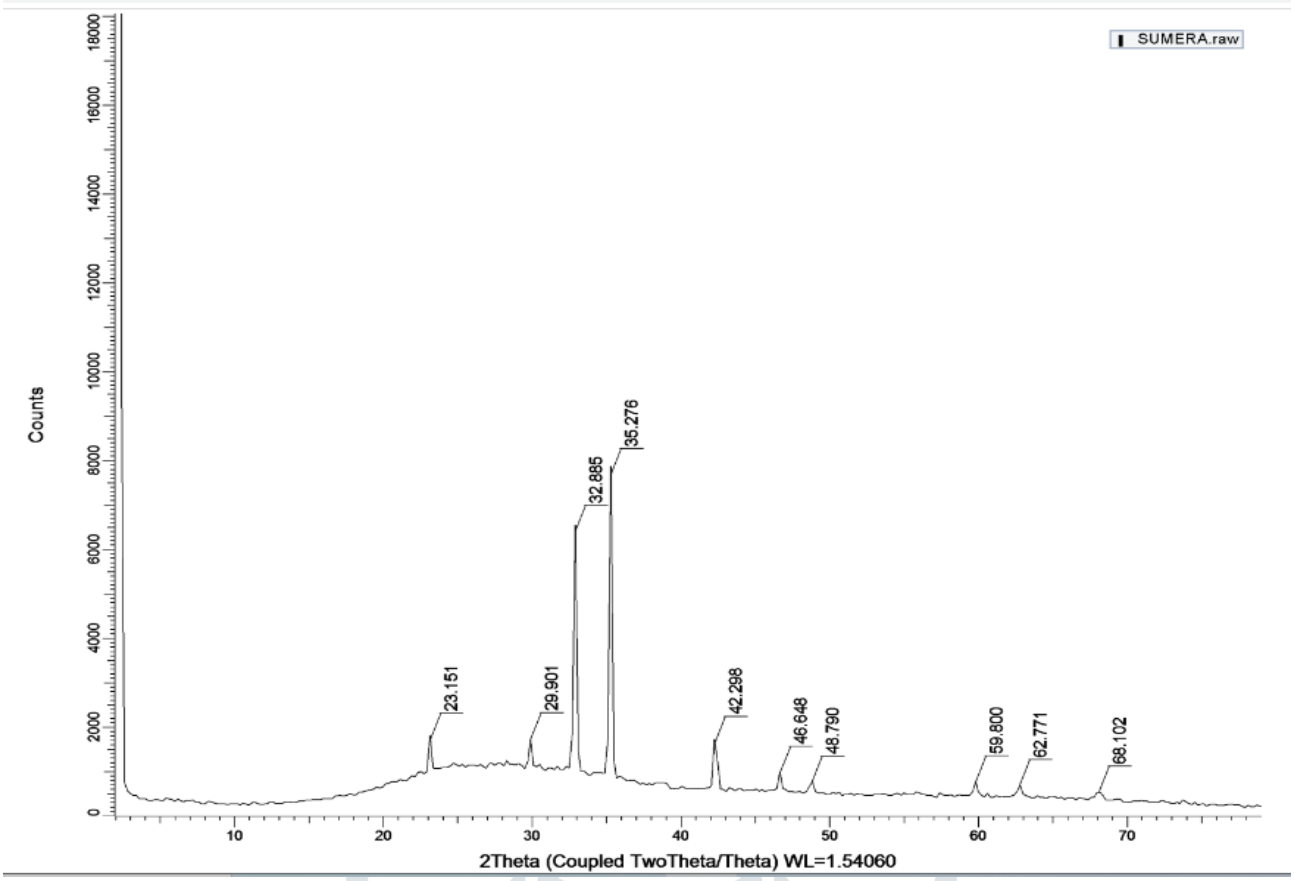

Figure:8 XRD Diffractogram of AgNPs of stem of *Cissus quadrangularis* L

Table:7 FWHM, crystalline size and D values of all the peaks.

2 θ	FWHM	Crystalline Size	D-Value
23.1506	0.75	112.88	3.83890
29.9005	0.75	114.46	2.98588
32.8846	2.25	38.44	2.72142
35.2757	2.65	32.84	2.54224
42.2979	0.65	136.82	2.13501
46.6478	0.35	258.08	1.94555
48.7901	0.3	303.59	1.86501
59.7995	0.3	318.95	1.54528
62.7706	0.25	388.68	1.47909
68.1022	0.2	500.60	1.37570

IV. CONCLUSION

In the present study, a green strategy for the biosynthesis of AgNPs from *Cissus quadrangularis* L. stem extract was described. The preliminary test for identifying the phytochemicals present was carried out, followed by the GC-MS analysis, which reported the occurrence of a number of biologically active compounds. The Spectral analysis, including the FTIR and UV-VIS spectroscopy, revealed various functional groups at the respective wavelengths. The characterization of the synthesized nanoparticles from the *Cissus quadrangularis* L. stem extract was performed

using the SEM, Zeta potential, particle size and the XRD analysis. The characterization study reported the crystallinity of the synthesized naanoparticles, their size, shape, uniformity, surface charge and polydispersity index, which indicates the aggregation of the nanoparticles. The properties mentioned above of the AgNPs make them a suitable potential drug candidate for discovering novel drugs in the medical and pharmaceutical industry.

REFERENCES

Abedi, F., Razavi, B. M., & Hosseinzadeh, H. (2019). A review on gentisic acid as a plant-derived phenolic acid and metabolite of aspirin: Comprehensive pharmacology, toxicology, and some pharmaceutical aspects. *Phytotherapy Research*, *34*(4), 729–741. https://doi.org/10.1002/ptr.6573

Bawazeer, S., Rauf, A., Shah, S. U. A., Shawky, A. M., Al-Awthan, Y. S., Bahattab, O. S., Uddin, G., Sabir, J., & El-Esawi, M. A. (2021). Green synthesis of silver nanoparticles using *Tropaeolum majus*: Phytochemical screening and antibacterial studies. *Green Processing and Synthesis*, *10*(1), 85–94. https://doi.org/10.1515/gps-2021-0003

Dias, D. A., Urban, S., & Roessner, U. (2012). A Historical Overview of Natural Products in Drug Discovery. *Metabolites*, *2*(2), 303–336. https://doi.org/10.3390/metabo2020303

Dynamic Light Scattering Particle Size Distribution Analysis. (n.d.). Horiba Scientific. https://www.horiba.com/sgp/scientific/technologies/dynamic-light-scattering-dls-particle-size-distribution-analysis/dynamic-light-scattering-dls-particle-size-distribution-analysis/#:%7E:text=Particle%20size%20can%20be%20determined,elastic%20light%20scattering%20(QELS).

Iravani, S. (2011). Green synthesis of metal nanoparticles using plants. *Green Chemistry*, *13*(10), 2638. https://doi.org/10.1039/c1gc15386b

Eswaran, R., Anandan, A., Doss, A., Sangeetha, G., & Anand, S. P. (2012). Analysis of the chemical composition of Cissus quadrangularis linn by GC-MS. *Asian j pharm clin res*, *2*, 139-40.

Gujral, S. S. (2014). Synthesis and characterization of Gold and Silver nanoparticles using different leaf extracts namely Catharanthus roseus, Datura metal, and Azadirachta indica, and estimation of antimicrobial activity of silver nanoparticles using disc diffusion method. *Int J Drug Dev Res*, *6*, 222-32.

Kim, YK, Lee, YS, Jeong, DH, Cho, MH, et al.: Antimicrobial effect of silver nanoparticles. Nanomed 3, 95–101 (2007)

Krithiga, N., Rajalakshmi, A., & Jayachitra, A. (2015). Green Synthesis of Silver Nanoparticles Using Leaf Extracts of Clitoria ternatea and Solanum nigrum and Study of Its Antibacterial Effect against Common Nosocomial Pathogens. *Journal of Nanoscience*, *2015*, 1–8. https://doi.org/10.1155/2015/928204

Mishra, G, Srivastava, S, Nagori, BP: Pharmacological and therapeutic activity of Cissus quadrangularis: an overview. Inter J Pharm Tech Res 2, 1298–1310 (2010)

Pakkirisamy, M., Kalakandan, S. K., & Ravichandran, K. (2017). Phytochemical Screening, GC-MS, FT-IR Analysis of Methanolic Extract of Curcuma caesia Roxb (Black Turmeric). *Pharmacognosy Journal*, *9*(6), 952–956. https://doi.org/10.5530/pj.2017.6.149

Saini, R., Saini, S., & Sharma, S. (2010). Nanotechnology: The future medicine. *Journal of Cutaneous and Aesthetic Surgery*, *3*(1), 32. https://doi.org/10.4103/0974-2077.63301

Umoren, S. A., Obot, I. B., & Gasem, Z. M. (2014). Green synthesis and characterization of silver nanoparticles using red apple (Malus domestica) fruit extract at room temperature. *J Mater Environ Sci*, *5*(3), 907-914

Zhang, X. F., Liu, Z. G., Shen, W., & Gurunathan, S. (2016). Silver Nanoparticles: Synthesis, Characterization, Properties, Applications, and Therapeutic Approaches. *International Journal of Molecular Sciences*, *17*(9), 1534. https://doi.org/10.3390/ijms17091534