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Biological Potential And Characterization Of Green Synthesized Silver Nanoparticles Using *Chromolaena Odorata (Linn)*

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ABSTRACT
In this research paper, we present a simple and eco-friendly biosynthesis of silver nanoparticles using Chromolaena odorata leaf extract as a reduction agent. For phytochemical screening such as carbohydrates, terpenoids, resins, saponins, tannins and alkaloids in the Chromolaena odorata extract, the current research was conducted using normal protocols and antibacterial activities of the silver nanoparticles extract of Chromolena odorata leaf prepared from extracts. The phytochemical constituents of C.odorata were qualitatively analyzed and the occurrence of different phytochemicals in the ethanol extract was verified. ZETA SIZER, FTIR, SEM, AFM, XRD and UV- Visible spectrophotometers were used to study the characterization of silver nanoparticles. AgNPs of chromolaena odorata leaf extracts were used for antibacterial activity against four bacterial species, namely E.coli, Staphylococcus aureus, Pseudomonas aeruginosa and Bacillus subtilis. Good findings for sugars, terpenoids, tannins and saponins were seen in the leaf extract. Silver ion reduction in the presence of plant extract and the production of silver nanoparticles confirmed a transition from light black to dark black in the color of the solution after treatment with AgNO3.FTIR leaf extract spectrum, clearly displaying maximum peaks at 803.77 cm-1 corresponding to C-C stretch of Alkane, peak at 1243 cm-1 assigned to C-O stretch of Alcohol, peak at 2119.047 cm-1 assigned to C=C stretch of Alkynes terminal, peak at 3051.265 cm-1 corresponding to C-H stretch of Aromatic and relatively strong peak at 3700 cm-1 attributed to Alkynes free O-H stretch, peak at 3051.265 cm-1 corresponding to C-H stretch of Aromatic and relatively strong peak at 3700 cm-1 attributed to Alkynes free O-H stretch, peak at 3051.2655 cm-1. The highest absorbance is found to occur at 190nm. The morphology of the particle and its size were determined by SEM. It is remembered that the shape was structurally spherical. It varies in size from 93-191nm. AFM was able to see the peculiar shapes and the composition of the silver nanoparticles. The XRD spectrum showed different peaks of around 390. The good organism resistant to Chromolaena odorata AgNPs was found to be Pseudomonas aeruginosa, while others showed intermediate quality. It was discovered that the silver nanoparticles synthesized from C.odorata extracts had the strongest antimicrobial efficacy against P. aeruginosa (15 mm), S. (14mm) with aureus, and E. Coli (14mm) and, respectively, the lower antimicrobial activity of silver nanoparticles synthesized by Bsubtilis (13 mm).

Keywords: Chromolaena odorata, Phytochemical screening, silver nanoparticles; ZETA SIZER, FTIR, SEM, AFM, XRD, and UV–Vis spectrophotometer. Antibacterial activity.

1. INTRODUCTION

The quickly evolving field of nanotechnology allows the development of nanometer-scale materials (Kruis et al. 2000). They are very cost-effective and have low maintenance

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requirements. This research was planned for the simple, cost-effective and environmentally pleasant synthesis of nanoparticles from silver (AgNPs) using the reduction and stabilization agent C.odorata leaves extract. A protocol silver nano particle for the synthesis of using radiolysis of silver ions in ethylene glycol was stated by (Soroushian et al). Recently the usage of the antibacterial and anticancer agent Silver nanoparticles is high. Due to their nanoscale structure, silver nanoparticles possess numerous novels and advanced physical, chemical and biological properties and functionality. The industry admires silver nanoparticles for their wide variety of uses in diverse fields.

2. METHODOLOGY

Selection of Plant Material

Leaves of *C.odorata* plants were collected from Kodikanal Mount.

Fig 1. *Chormoleana odorata*

Plant Description

A perennial herb that grows quickly is Chromolaena odorata. It is a multi-stalked shrub that in open fields, is 2.5 m (100 inches) thick and widely planted. These have branches that are fuzzy, but the shrub is woody at the base. It becomes etiolated and behaves like a creeper in shady regions, rising on other vegetation. It can grow to a height of 10 m (33feet). The plant is hairy and glandular and when crushed, the leaves emit a pungent, aromatic odor. The leaves, being triangular or elliptical with serrated edges, are opposite. The leaves are 1-5 cm thick and 4-10 cm long (up to 4-2 inches)

Preparation of Extraction

New C. leaves. Kodaikanal Mount was a set of odorata used in this analysis. The fresh leaves were dried for two consecutive days under direct sunshine, allowed to dry entirely under a ventilator for another 2 days at room temperature and ground into powder. 100 g of the powder was sprayed with 300 ml of ethanol and left to stand at room temperature for 1 day. It filtered the extract.

Phytochemicals Analysis of *C.Odorata* Leaf Extracts (Harbone, 1975)

Alkaloids (Wagner's test), Flavonoids (Alkaline reagent test), Glycosides, Saponins, Tannins (Legal Test), Cardiac glycosides (Keller's killani test), Steroids and Phytosterol (Libermannburchard test), Triterpenoids, Phenols, Anthraquinone.

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Biosynthesis of Silver Nanaoparticles:

- Preparation of 0.1 M silver nitrate solutions
- Addition of ammonium solution to the silver nitrate solution
- Addition of plant extracts to the silver nitrate solutions
- Incubation to allow nanoparticles formation at room

Temperature

UV-VIS SPECTROSCOPY: (SPECORD – 210, GERMANY)
A UV-Vis (Specord-210, Germany) to determine the optical properties of Ag-NPs spectrophotometer was used. From 350nm to 500nm at different intervals up to 24 hours after adding of AgNO3 to the plant extract the spectra were obtained. After 24 hours of AgNO3 insertion, the spectrum was captured.

FTIR Analysis: (AGILENT- CARRY 630)
Using the FTIR spectrometer analysis of the chemical composition of the synthesized silver nanoparticles (Agilent Company, Carry 630 model, Ger). By the KBr pellet process the solutions were dried at 750C and the dried powders were differentiated in the 4000-400 cm-1 range.

XRD Analysis: (PHILIPS PAN ANALYTICAL)
An optimal method for the calculation of the crystallite size of powder samples is X-ray diffraction. The theory requires detailed quantification of the peaks' expansion. X-ray diffraction spectroscopy determined synthesized Silver nanoparticles' phase variety and grain size (Philips PAN analytical).

ZETA SIZER :(HORIBA, JAPAN)
In mortar and pestle, the synthesized silver nanoparticles were crushed to move the amount of up to 100ml with purified water to the beaker. Then kept in an ultrasonicator to form a silver nano particle and the solution was read at Zeta Sizer instrument for the Zeta Sizer and Zeta potential. Zeta sizer was read using Cuvette. Zeta potential was read using Electrode.

Electron Microscopy Scanning: (SEM - TESCAN)
The morphology of the powder samples was analyzed at Anna University, Chennai, using the Scanning Electron Microscope (SEM - Tescan) analysis.

Atomic Force Microscope (APE RESEARCH, ITALY)
Samples were air-dried from the maximum point at which silver nanoparticles were produced and permitted to be characterized by Atomic Force Microscopy (APE Research, Italy) because of their agglomeration of silver, morphology thorough scale. The AFM image was taken with the

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following configuration of silicon cantilevers: 0.02-0.77 N/m force constant, tip height of 10-15 nm, contact mode.

Through Well Diffusion Process, Antimicrobial Activity

The Department of Microbiology, Dwaraka Doss Goverdhan Doss Vaishnav College, Chennai, collected *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus subtilis* and *Escherichia coli*. The well diffusion system tested silver nanoparticles synthesized from *Chromoleana odorata* plant powder.

3. RESULTS AND DISCUSSION

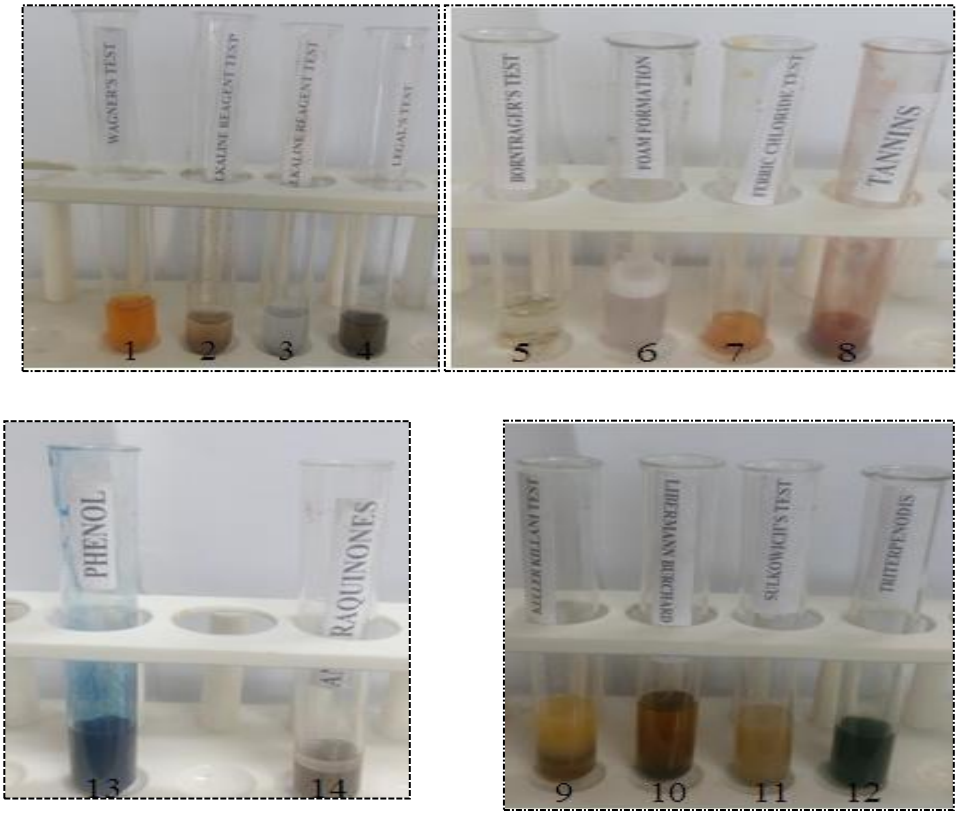

Fig2: Phytochemical Tests for Ethanol Extract

Biosynthesis of Nanoparticles

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Fig3: Silver Nanoparticles' Synthesis
(1)Silver Nitrate Solution (2) After adding ammonium solution (3) After adding Plantextract

Previous experiments have shown that AgNPs can be synthesized by plants such as Azadirachta indica (Shankar, S.S et.al 2003), Capsicum annuum (Ahmad, A et al 2003 a&b) rica papaya (Jha, R et al 2010), Gliricidiasepium, Eucalyptus hybrid (Dubey, S.P et al 2009) and microorganisms such as Aspergillus fumigatus (Balaji, D.S et al 2009) Cladosporium cladosporioides Bhainsa, Fusarium oxysporum, Pseudomonas aeruginosa, Rhodopseudomonas capsulate (He, 2007). After mixing with C, in the current study. Odorata leaf extract, accompanied by 3-h incubation, reduced the silver ions to AgNPs (Silver nanoparticles). The hue shifted to charcoal from dark brown.

UV- VIS Spectroscopy

V is spectrophotometer research, the synthesized silver nanoparticles have been monitored by UV which reveals the absorption spectra of silver nanoparticles being produced by the reaction media having an absorption peak of C at 420nm instead of C for odorata.

ZETA Size Analysis

Table 1: Zeta size analysis ofAgNps Nanoparticles for *c.odorata*

Peak No	S.P Area Ratio	Mean	S.D	Mode
1	1.00	129.5nm	39.2nm	112.2nm

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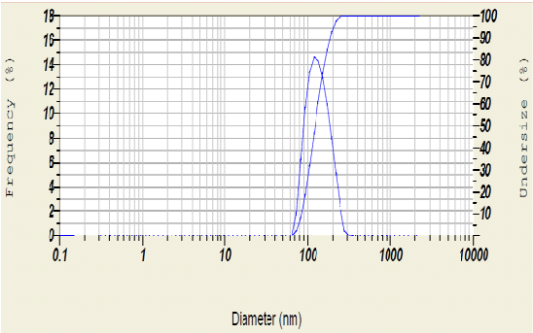
2	---nm	---nm	---nm	---nm
3	---nm	---nm	---nm	---nm
TOTAL	1.00	129.5nm	39.2nm	112.2nm

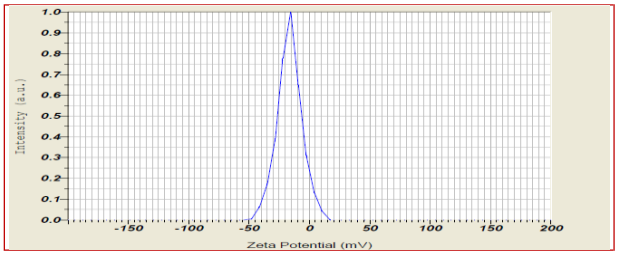


Graph1: ZETA Analysis Graph of Silver

Cumulant Operations

Z-Average : 104.2nm
PI : 0.787

ZETA Potential



Graph 2: Potential Activity Of *C.odorata*

Peak No	Zeta Potential	Electrophoretic Mobility
1	-16.4Mv	-0.000127cm^2/Vs
2	---Mv	---cm2/Vs
3	---mV	---cm2/Vs

Table 2: ZETA Potential

Zeta Potential (Mean): -16.4Mv
Electrophoretic Mobility Mean: -0.000127cm^2/Vs

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FTIR Analysis

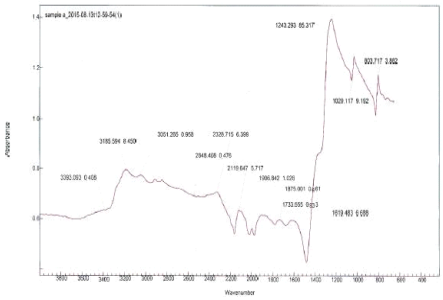

Fig 4 : FTIR Spectra for *C.odorata*

Identification of Electron Microscopy using Scanning

For the study of the morphology and scale of the synthesized C, scanning electron microscopy was used. Nanoparticles Odorata. (Fig.5) illustrates the SEM pictures of C. Odorate the nanoparticles with varying magnifications. The SEM pictures of C. Odorata nanoparticles demonstrate the spherical morphology of well crystallized particles. The SEM study found that C sizes ranged from 93 to 142 nm for the synthesized Agnes. Odorata.

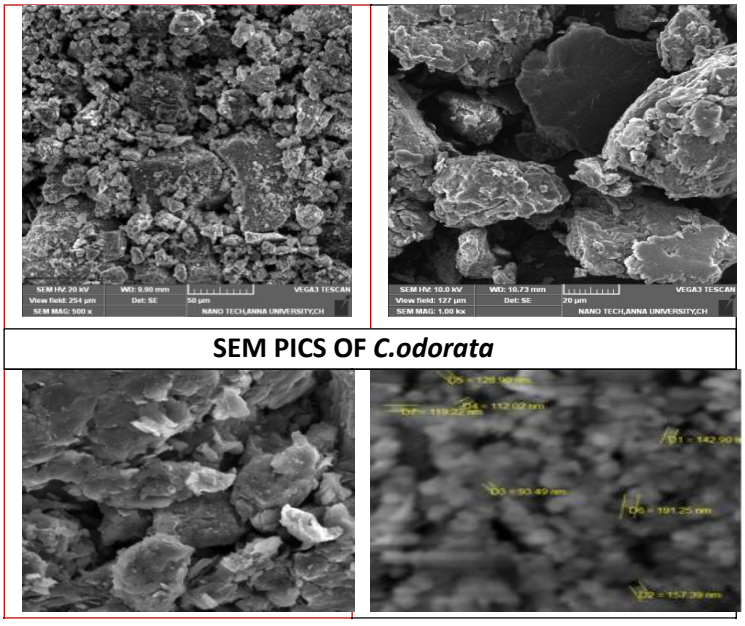

Fig 5: Electron Microscopy Scanning

Identification of Microscopy using Atomic Force

Atomic Force Microscopy (AFM) characterized the silver nanoparticles by their precise size and silver morphology. Fig.6 revealed topographical photographs of unusual silver nanoparticles synthesized with natural plant extract.

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XRD Diffraction

The XRD spectrum (Figure 7) revealed separate diffraction peaks of about 380, indexed by the silver nanoparticles (Ahmad, A et al 2003b). Due to decreased agent stability of the nanoparticles, these sharp Bragg peaks may have occurred. Consequently, on the surface of the silver nanoparticles the solidification of the bio-organic level takes place using the XRD findings. It shows the effect of experimental conditions on the nucleation and growth of the nuclei of crystals which indicates wider peaks with smaller particle size.

It was discovered that the C-synthesized silver nanoparticles. Odorata extracts had the strongest antimicrobial efficacy against P. aeruginosa (15 mm), S. (14mm) aureus, and E. Coli (14 mm) and the reduced antimicrobial activity of synthesized silver nanoparticles against B.subtilis, respectively, and (13 mm). The findings were confirmed by the antibacterial activity of silver nanoparticles against numerous pathogenic plants, including S, Ocimumtenuiflorum, Solanum tricobatum, Syzygiumcumini, Centella asiatica and Citrus sinensis (Peter Logeswari et al 2012) Oh, aureus, P. aeruginosa, E, P. aeruginosa, E. The inhibition region was about 30 mm to 12 mm for coli and B.subtilis, respectively.

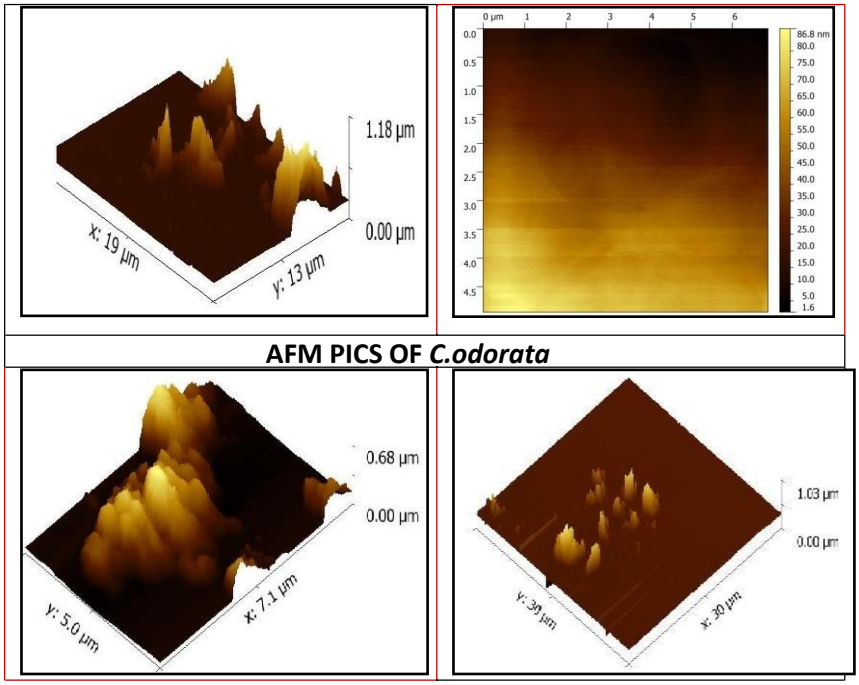

Fig 6: AFM pictures with the synthesized Nanoparticles of Silver by *C. odorata.*

Fig 7: X-RAY Diffraction Pattern of Synthesized Type C Silver Nanoparticles C. Odorata through well Diffusion Process, Antimicrobial Activity

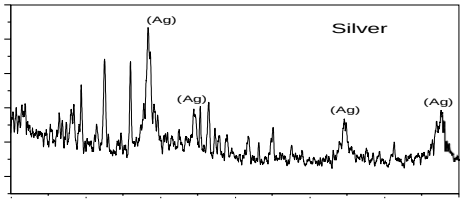

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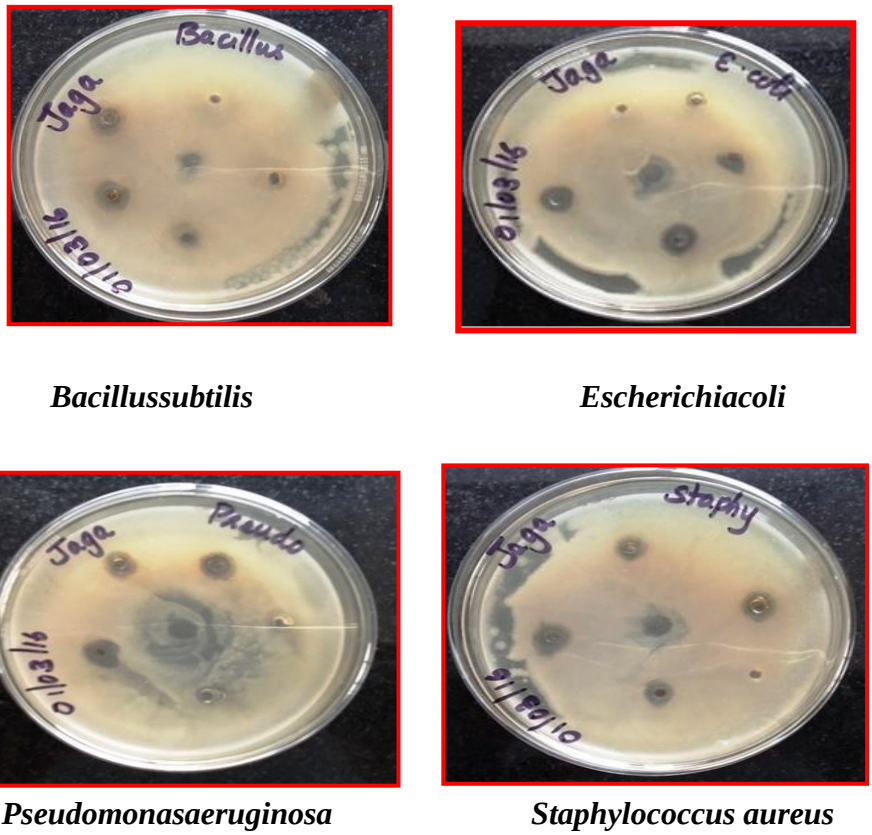
Table 3: Silver Nanoparticle Inhibition Zone By C.odorata Extract

S.No	Microorganism	Control	1 mg/ml	2 mg/ml	3 mg/ml	4 mg/ml	5 mg/ml
1	*E.coli*	-	5	12	12	13	14
2	*B.subtilis*	-	-	-	10	12	13
3	*S.aureus*	-	12	13	10	12	14
4	*P.aeruginasa*	-	5	10	12	15	10

Plant



Bacillussubtilis ***Escherichiacoli***

Pseudomonasaeruginosa ***Staphylococcus aureus***

Fig 8: Antibactertial Activity of AgNps *(C. odorata)*

4. CONCLUSION

This study was aimed to synthesize and characterize the silver nanoparticles of C. odorata and to evaluate the antibacterial and anticancer activity.

The phytochemical characteristics of C.odorata leaf ethanol extraction were examined through phytochemical screening tests for the existence of chemical constituents such as alkaloids, flavonoids, saponins, tannins, cardiac glycosides, triterpinoids, phytosterol, steroids and phenols. The ethanol extract tested positive for tannins, phenols and glycosides of the heart. Qualitative analyzed the phytochemical constituents of C.odorata and confirmed the presence of various phytochemicals in the Ethanol extract. This result concordant with (Harborne, J.B 1995) who given A handbook of Bioactive compounds from plants.

In vitro experiments and characterization of silver nanoparticles indicated a key role in many therapeutic formulations, the results drawn from the analysis suggest. UV-vis spectrophotometer study monitored the composition and optimisation of the reduced silver nanoparticles in the colloidal solution. About 190 nm of sharp bands of silver nanoparticles were detected.

The abnormal shape of AgNPs, and the development of silver nanoparticles, were shown by AFM. The SEM study revealed that the sizes ranged from 93 to 142 nm for the synthesized AgNps. The development of silver nanoparticles was shown by SEM determination of healthy samples of brown color as well scattered nanoparticles could be seen in ***Chromolaena odorata*** samples treated with silver nitrate.

To classify the bimolecular for capping and effective stabilization of the synthesized metal nanoparticles, FTIR calculation was carried out showing that the band between 3500-3000 cm-1 corresponds to a stretch for C-H bonds, the peak about 1500-1000 cm-1 shows the bond stretch for N-H. (C.odorata) and 3176 cm-1 lead to the C-H bond stretch, with peaks around 1500-810 cm-1 reflecting the N-O bond stretch.
It was found that the zeta scale was 129.5 nm and the zeta potential was found to be 16.4 mvolt. For Chromolaena odorata, the XRD spectrum showed separate peaks of about 39º. These sharp bands may have resulted in Bragg peaks due to the stabilization by the capping agent of the nanoparticles. In the antimicrobial screening technique, the zones of inhibition were established suggesting that the Ag NPs synthesized in this phase had effective antimicrobial activity against pathogenic bacteria. Because of their good antimicrobial function, biologically synthesized silver nanoparticles may be of enormous value in the medical field.

C. odorata extracts, which are an inexpensive, effective and eco-friendly procedure, have created silver nanoparticles. The reduction of silver nitrate into silver nanoparticles by UV-vis spectrophotometer, XRD, AFM and SEM techniques have been confirmed. In the indicated antimicrobial screening test, the zones of inhibition have been established; the anticancer activity of Ag NPs synthesized from C. In this scheme against pathogenic bacteria, Odorata has good antimicrobial activity. Chemically synthesized silver nanoparticles for their successful use may be of tremendous importance in the medical industry.

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