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Fabrication of Silver Nanoparticle Using Aqueous Leaf Extract of *Talinum portulacifolium* and Its Antimicrobial Activities

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Abstract

The escalating issue of antibiotic resistance has prompted a quest for innovative antimicrobial solutions. Among these, nanoparticles produced through environmentally friendly methods have garnered significant interest for their potential antimicrobial attributes. Nanoparticles are used immensely due to its small size, orientation, physical properties, which are reportedly shown to change the performance of any other material which is in contact with tiny particles. These particles can be prepared easily by different chemical, physical and biological approaches. But the biological approach is the most emerging approach of preparation, because this method to easier than the other methods, ecofriendly and less consuming. This investigation centres on the production of silver nanoparticle using the leaves of Talinum portulacifolium The biosynthesized silver nanoparticle derived from Talinum portulacifolium leaves were subjected to comprehensive characterization through various techniques. In the UV-visible spectrum analysis, the silver nanoparticles exhibited maximum absorption peak at 380 nm. XRD analysis indicated that the silver particles generated in our experiments existed in the form of nanocrystals, as evidenced by peaks at 2θ values of 17.852, 21.001, 23.141, and 27.901 and 28.796 corresponding to the height of 216.20, 172.18, 308.26, 317.09 and 128.53cts respectively. The corresponding "d" spacing value of silver nanoparticle were 4.968Å, 4.230Å, 3.4843Å, 3.197Å and 3.100Å. SEM spectral analysis unveiled the formation of cube-shaped silver nanoparticles within a size range spanning from 200 nm to 1 µm. The antimicrobial efficacy of metallic nanoparticles derived from Talinum portulacifolium was evaluated using the disc diffusion method against both gram-positive and gram-negative pathogens, including E.coli, Staphylococcus aureus, Bacillus subtilis, as well as the fungi Aspergillus flavus and Candida albicans. Notably, metallic Ag nanoparticle exhibited substantial zones of inhibition against the selected pathogens.

Keywords: *Synthesis, Talinum portulacifolium,* $AgNo_3$ *Nanoparticles, Characterization, Antimicrobial Activity.*

1. Introduction

Nanobiotechnology is the recently growing techniques in the scientific world. The nanoparticle was used in different fields of scientific research. It may be synthesized chemically

or biologically with living organisms. There is an enormous interest in the synthesis of nanoparticles due to their unusual optical (Krolikowska et al., 2003), chemical (Kumar et al., 2003), photo electrochemical (Chandresekharan et al., 2000) and electronic (Peto et al., 2002) properties. The plant mediated green synthesis of silver nanoparticles plays an important role in medicinal field especially in drug discovery of pharmaceutical industries. Recent studies on the use of plants and microorganisms in the synthesis of nanoparticles are a relatively new and exciting area of research with considerable potential for development. It is well known that many plants can provide inorganic materials either intra- or extracellularly (Mann, 1993) and microorganisms are recently found as possible eco-friendly nano-factories (Beveridge and Murray, 1980).

Physical properties of nanoparticles such as large surface area, energy, spatial confinement and reduced imperfections are the attractive attributes that lay a platform for its application in multiple approaches (Morons et al., 2005). Among the various methods like sol-process, micelle, sol-gel process, chemical precipitation, hydrothermal method, pyrolysis, chemical vapor deposition, bio-based protocol etc., bio-based protocol is the most important and eco-friendly production method (Leela and Vivekananda, 2008). Processes devised by the nature for the synthesis of inorganic materials on nano- and microlength scales have contributed to the development of a relatively new and largely unexplored area of research based on the use of microbes in the biosynthesis of nanoparticles (Sastry et al., 2003).

Green synthesis offers improvement over other methods, i.e., synthetic, chemical or using micro-organisms, as it is reported to be cost-effective, environmentally friendly and non-toxic to the environment and can be used for large-scale synthesis (Ghorbani et al., 2011). The green chemistry method is based on the mechanism of plant-assisted redu00ction due to the presence of phytochemicals (Pavani et al., 2012). The main phytochemicals involved are flavones, aldehydes, terpenoids, ketones, carboxylic acids and amides.

In this manuscript, we present the synthesis of silver nanoparticles (AgNPs) using leaves from *Talinum portulacifolium* a member of the Portulacaceae family and a prominent dicot family within Angiosperms. This study employs a straightforward and cost-effective green synthesis approach. The resulting nanoparticles will be thoroughly characterized through various analytical techniques, including UV-visible spectroscopy, Scanning Electron Microscopy (SEM), X-ray diffraction (XRD), and Antimicrobial Activity assessments.

2. Materials And Methods

Plant Material Collection

Leaves from *Talinum portulacifolium* as depicted in Plate 1, were chosen for the current study. The plant material, belonging to the Portulacaceae family, was collected from the Appaneri village, Kovilpatti taluk of Tuticorin district, Tamil Nadu. Taxonomic attributes were verified using references such as the 'Flora of Presidency of Madras' (Gamble, 1928) and the 'Flora of the Tamil Nadu Carnatic' (Mathew, 1981).

Isolation of Nanoparticles

Fresh and healthy *Talinum portulacifolium* leaves were carefully gathered and meticulously cleansed with tap water followed by distilled water to eliminate any dust or visible impurities. Subsequently, 10 grams of fresh leaves were boiled in 100 milliliters of double-distilled water, and the resulting extract was filtered through Whatman no.1 filter paper, collecting the filtrate in a conical flask. This obtained extract was utilized for the synthesis of various nanoparticles.

Preparation of Silver Nitrate Solution

1mM silver nitrate solution is prepared by the concentration of 0.0169 g in 100 ml double distilled water and stored.

Metal-Plant Extracts Interaction

90 ml of silver nitrate solution was placed in a conical flask, to which 10 milliliters of the leaf extract were added. The Silver Nitarte solution underwent a color shift from light green to brownish green. The conical flask was subjected to light exposure for a 72-hours incubation period.

Characterization

UV- Spectrophotometer Analysis

For UV-spectrophotometer analysis, approximately 1 ml of the sample suspension was transferred into a quartz tube. The sample was then diluted with 2 milliliters of distilled water to facilitate monitoring of nanoparticle synthesis. UV-Visible spectra scans were conducted in the wavelength range of 200-900 nanometers using a UV-visible spectrophotometer (Shimadzu UV 1800, Germany).

XRD Analysis

To characterize the purified synthesized nanoparticles, freeze-dried powder samples were employed for XRD analysis, which was conducted at 40 kV/20 mA using continuous scanning in 2 delta mode (Absar, 2003). The nanoparticle solution was first purified through repeated centrifugation at 5000 rpm for 20 minutes, followed by redispersion of the nanoparticle pellet into 10 milliliters of deionized water.

SEM Analysis of Silver Nanoparticles

SEM analysis was carried out using a ZEISS machine. Thin films of the sample were prepared on a carbon-coated copper grid by dispensing a small amount of the sample onto the grid. Any excess solution was removed using blotting paper, and the films on the SEM grid were allowed to dry by exposure to a mercury lamp for 5 minutes.

Antimicrobial Activity of Nanoparticles

The effectiveness of leaves extracts from *Talinum portulacifolium* against the growth of various human pathogens was evaluated using the Agar well diffusion method in a clinical laboratory (Scudder Diagnostic Centre, Nagercoil). The Kirby-Bauer method was employed to assess the antibacterial activity of isolated plant extraction pellets. Overnight grown cultures of the respective bacteria and fungi were inoculated onto Nutrient agar, Sabouraud's Dextrose Agar (SDA), and Potato Dextrose Agar (PDA) plates. Wells were created in all plates, including those designated for controls. Leaf extracts (acetone, benzene, distilled water, petroleum ether, and ethanol) were prepared at a concentration of 50 mg/mL. Antibiotics (Amikacin and Nystatin) at the same concentration were used as controls. Bacterial plates (E.coli, Pseudomonas aeruginosa, Bacillus subtilis, and Staphylococcus aureus) were incubated at 37°C for 24 hours, while fungal plates (Aspergillus flavus and Candida albicans) were incubated at 35°C for 48 hours. The diameter of the inhibition zone was measured in millimeters (Peraman Muthu Kumaran, 2014).

3. Results and Discussion

Synthesis and Characterization of Nanoparticles

The leaves of *Talinum portulacifolium* when exposed to Silver Nitrate in aqueous solutions, exhibited distinct color changes, signifying the formation of three distinct types of nanoparticles. These nanoparticles were subsequently confirmed through spectral studies involving UV-Visible Spectroscopy, X-Ray Diffraction, and SEM analysis, and they were employed in various biological activities. The meticulous control of starting materials is imperative for ensuring the reproducible quality of herbal products. Consequently, recent years have seen a heightened emphasis on the standardization of medicinal plants with therapeutic potential. Despite the availability of modern techniques, the identification and evaluation of plant-based drugs through pharmacognostical studies remain a more reliable, accurate, and cost-effective approach. According to the World Health Organization (WHO, 2000), the macroscopic and microscopic description of a medicinal plant constitutes the initial step toward establishing its identity and purity, and this should precede any other testing procedures (Anonymous, 2002).

UV- Visible Spectrum Analysis

The reduction of metal ions to metal nanoparticles, facilitated by exposure to *Talinum portulacifolium* leaves, was observable through color changes and subsequently confirmed via UV-Vis spectroscopy. UV-Vis spectroscopy is a widely employed technique for investigating size and shape-controlled nanoparticles in aqueous suspensions (Wiley et al., 2006). The presence of nanoparticles was verified by obtaining spectra in the visible range using a UV-visible spectrophotometer, with absorption wavelengths spanning from 200 to 900 nanometers (Figures 1).

Upon introducing *Talinum portulacifolium* leaf extract to a 1 mM Silver Nitrate solution, the solution transitioned from light green to dark green within 10 minutes, indicative of Ag nanoparticle formation.

Silver Nanoparticles Analysis of *Talinum portulacifolium* Leaves

Absorption spectra of silver nanoparticles formed in the reaction medium exhibited an absorbance peak within the 200 to 400 nm range, with the characteristic peak of silver nanoparticles observed at 240 nm (Figure 1). It is known that when the surface plasmon vibrations in silver nanoparticles are excited, the silver nanoparticles exhibit some yellowish brown color in 30 the aqueous solution (Krishnaraj *et al*., 2010). Our findings align with the research of Dubey et al. (2010), who investigated the significant effect of metal salt concentration on the synthesis of silver nanoparticles.

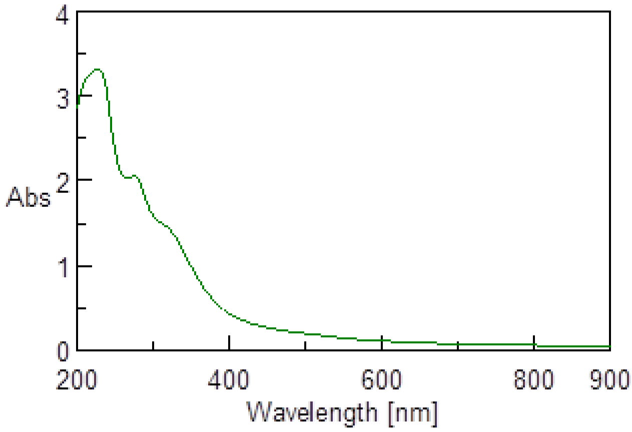


Figure 1: UV-Vis Spectrum of Silver Nanoparticles in Leaves of *Talinum portulacifolium*

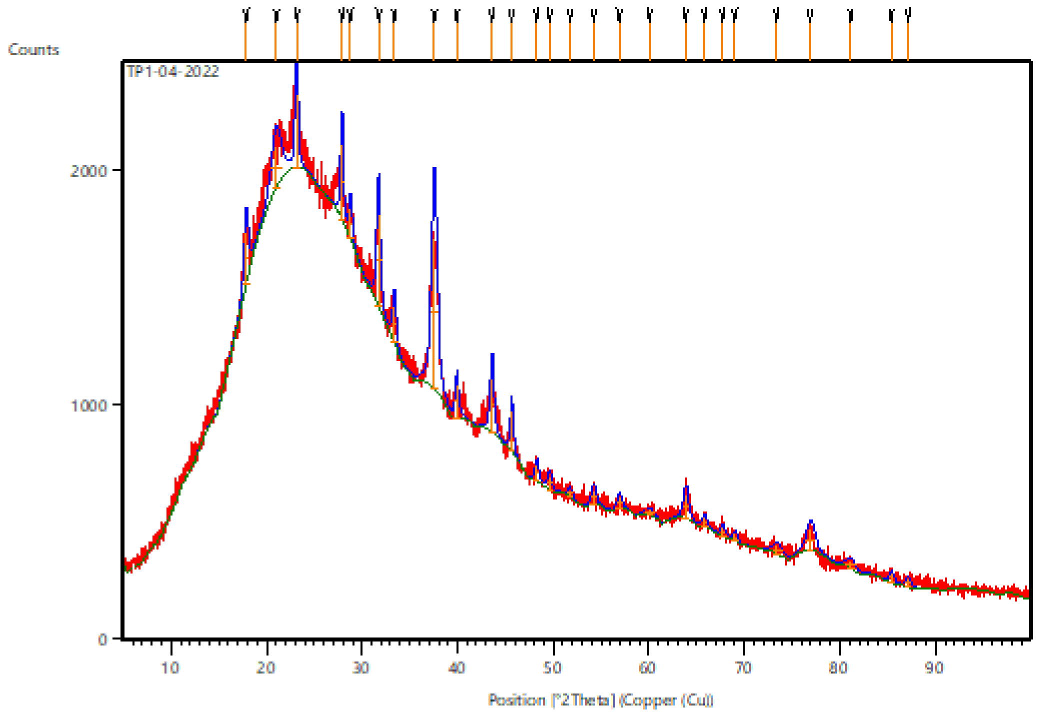
XRD Analysis

The biosynthesized nanoparticles, derived from *Talinum portulacifolium* leaf extracts, were meticulously characterized and validated through X-ray diffraction (XRD) analysis (Figure 1). The XRD analysis aimed to confirm the crystalline nature of the nanoparticles. A comparative assessment of our XRD spectrum against established standards substantiated that the nanoparticles generated in our experiments indeed existed in the form of nanocrystals, as indicated by the discernible peaks at specific 2θ values. In the case of silver nanoparticles, the XRD analysis revealed eight distinctive peaks in the XRD image, ranging from 0 to 90 Å (Figure 2, Table. 1). These peaks corresponded to 2θ values of 17.852, 21.001, 23.141, 27.901, 28.796, 31.725, 33.369 and 17.852 corresponding heights of 216.20, 172.18, 308.26, 317.09, 128.53, 385.34, 152.76 and 216.20cts respectively. The corresponding "d" spacing value of silver nanoparticle were measured by 4.968Å, 4.230Å, 3.4843Å, 3.197Å, 3.100Å, 2.820Å, 2.685Å and 4.968Å. The average size of the silver nanoparticles synthesized using *Acalypha indica* leaf extracts by (Krishnaraj et al, 2010) was nearly 35 nm. The nanoparticles synthesized using *C. citronella* and *S. verbascifolium* were around 36 and 41 nm respectively presented by Joy (Joy and Johnson, 2015).

Table 1: XRD Pattern of AgNPs Synthesized by Leaves of *Talinum portulacifolium* Extract with $AgNO_3$ Solution

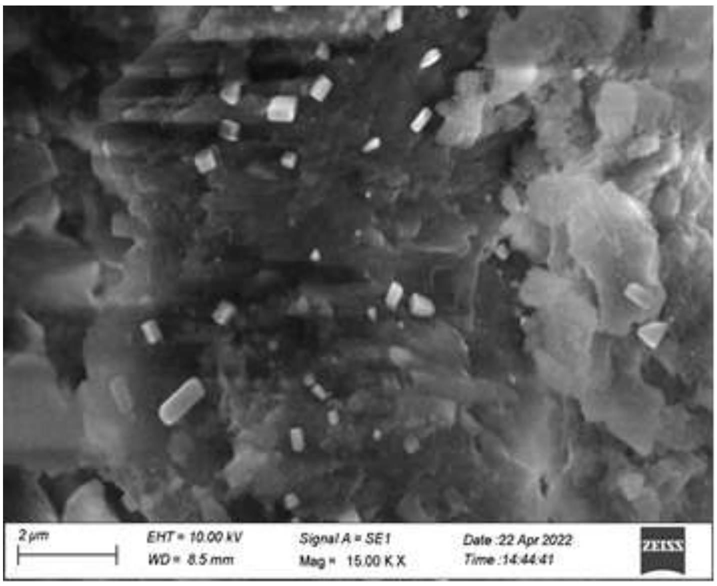
Pos.[°2Th.]	Height [cts]	FWHM Left [°2Th.]	d-spacing [Å]	Rel. Int. [%]
17.852	216.20	0.519	4.968	33.78
21.001	172.18	1.212	4.230	26.90
23.141	308.26	0.346	3.843	48.16
27.901	317.09	0.346	3.197	49.54
28.796	128.53	0.433	3.100	20.08
31.725	385.34	0.433	2.820	60.21
33.369	152.76	0.692	2.685	23.87
17.852	216.20	0.519	4.968	33.78

Figure.2 : XRD Spectrum of AgNPs in Leaves of *Talinum portulacifolium*



SEM Analysis of Silver Nanoparticles

Examination of the Scanning Electron Microscopy (SEM) images provided insights into the formation and morphology of stable silver nanoparticles derived from *Talinum portulacifolium* leaf extract. The SEM analysis revealed the uniform distribution of silver nanoparticles on the cell surfaces (Figure 3). These silver nanoparticles exhibited a cubic shape, with particle sizes ranging from 2 to 20µm. The presence of larger silver particles may be attributed to the aggregation of smaller ones, potentially occurring during the SEM measurements. Most of the silver nanoparticles were uniformly distributed, but in the later time nearly 14 w, the particles formed an aggregation The organic compounds or the reducing agents present in the extract may be responsible for the shape and size of the nanoparticles, where the compounds interlink with nanoparticles and reduce them (Phatak RS and Hendre AS 2015).



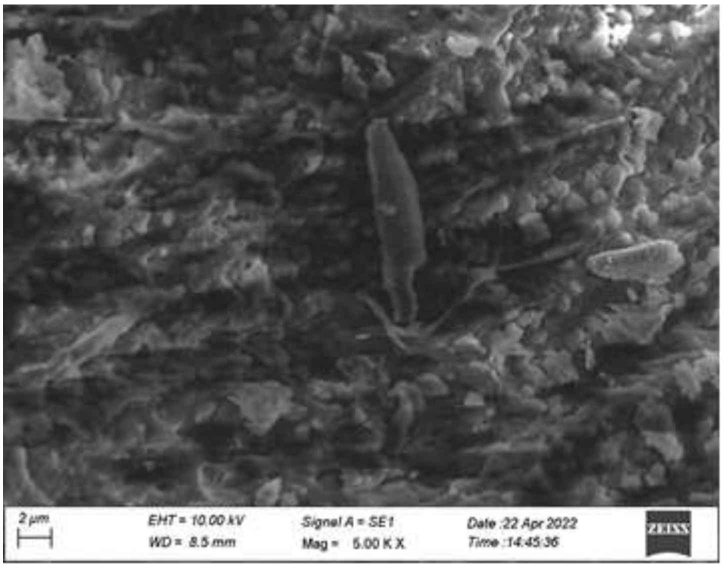


Figure 3: SEM image of AgNPs synthesized by Leaves of *Talinum portulacifolium* extract with $AgNO_3$ Solution

Antimicrobial Activity Studies

Infections caused by a range of bacterial agents, including pathogenic strains of *Escherichia coli*, *Salmonella* spp., and *Staphylococcus aureus*, are pervasive and on the rise. In recent years, the global emergence of drug resistance in human pathogenic bacteria has become a concerning trend, as documented by various researchers (Piddock and Wise, 1989; Singh et al., 1992; Mulligen et al., 1993). The relentless use of antibiotics has led to the development of resistance among microorganisms, posing substantial challenges in clinical settings for the treatment of infectious diseases (Davis, 1994).

Plant-based materials continue to be a vital resource in combating serious diseases worldwide. According to the World Health Organization (WHO, 1993), a staggering 80% of the global population relies on traditional medicine, with a significant portion of these therapies incorporating plant extracts or their active components. Despite their longstanding use, the systematic study of plants to identify their antimicrobial properties represents a relatively recent field of research.

The findings from this study have demonstrated the substantial antimicrobial activity exhibited by various metal nanoparticles against a diverse array of microorganisms. This trend is particularly pertinent in light of the gradual escalation in drug resistance among microorganisms (Amin et al., 2012). While synthetic drugs have been employed to combat a wide range of diseases caused by pathogenic microbes in humans, they often entail side effects, especially when overdosed. As a result, several medicinal plants have been identified and harnessed for their effectiveness in treating bacterial infections in humans. In the present investigation, the antimicrobial activity of *Talinum portulacifolium*. Leaves were assessed against six pathogens, and the results, including the zones of inhibition, are depicted in Plate 1 and tabulated for reference.

a) Antibacterial Activity Assessment

In this study, we assessed the antibacterial potential of various nanoparticle extracts derived from *Talinum portulacifolium* leaves. Our investigation revealed significant antibacterial activity against the following bacterial strains: *Escherichia coli*, *Bacillus subtilis*, and *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

b) Antibacterial Studies on *Talinum portulacifolium* Leaves

The extracts obtained from *Talinum portulacifolium* leaves, containing silver nanoparticle extracts, exhibited in-vitro inhibitory effects on the growth of the tested microorganisms. The results, showcasing the zones of inhibition measured in millimeters (mm), are succinctly presented in Table 2.

Generally, we observed moderate inhibition of the growth of these test organisms, evident by cleared zones on the culture plates (Plate 1).

Table 2: Antimicrobial potential of SiNPs nanoparticles of *Talinum portulacifolium* (Forssk.) Asch. ex Schweinf

Pathogens	Antibacterial activity - Zone of Inhibition (mm)	
	Silver Nanoparticles ($AgNO_3$)	Control (Amikacin)
Escherichia coli	15 ± 0.3	21 ± 0.4
Bacillus subtilis	14 ± 0.2	17 ± 0.3
Staphylococcus aureus	17 ± 0.4	16 ± 0.4
Pseudomonas aeruginosa	15 ± 0.5	18 ± 0.3
Candida albicans	15 ± 0.4	15 ± 0.2
Aspergillus flavus	14 ± 0.5	18 ± 0.3

± Standard Error, + Present, **-** Absent

The overall antimicrobial efficacy varied among the different pathogens. Notably *Staphylococcus aureus* exhibited the largest zone of inhibition (17mm), incontiniuity *E. coli and Pseudomonas aeruginosa* measuring 15 mm, when exposed to silver nanoparticle. *Bacillus subtilis* showed a slightly smaller inhibition zone of 14 mm.

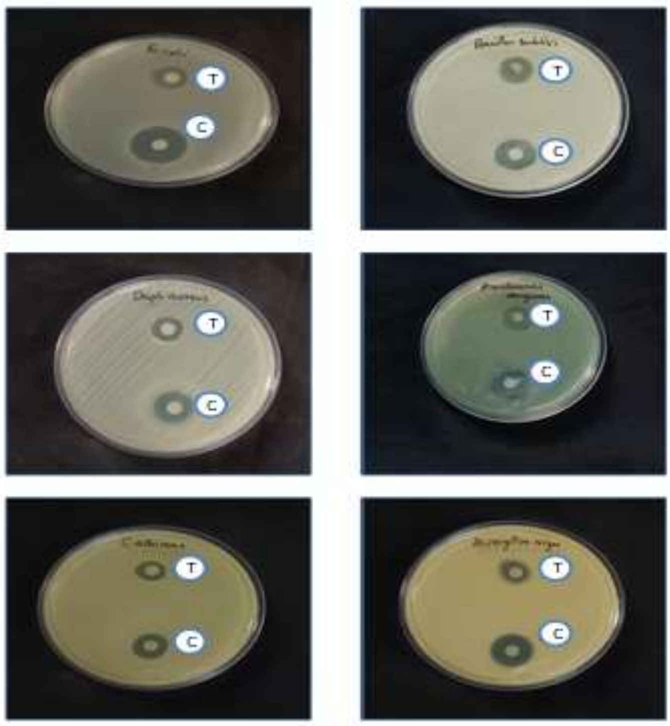


Plate 1: Antimicrobial Activity of SiNPs Nanoparticles from *Talinum portulacifolium* Leaves

The size of metallic nanoparticles plays a crucial role in maximizing the surface area available for interaction with bacterial cells, thereby enhancing the efficacy of bacterial eradication (Parameswari et al., 2010). This phenomenon can be attributed to the interaction between copper nanoparticles and the bacterial cell wall, which is particularly facilitated by the abundance of negative charges present in gram-negative bacteria (Ruparelia et al., 2008). Moreover, the surface properties of nanoparticles lead to interactions with the bacterial outer membrane, ultimately causing membrane disruption and bacterial cell death (Applerot et al., 2009).

(c) Antifungal activity

The antifungal activity of various nanoparticles derived from leaves of *Talinum portulacifolium* was assessed against two strains, namely *Aspergillus flavus* and *Candida albicans*.

(d) Antifungal studies on Leaves of *Talinum portulacifolium*

In silver nanoparticles of *Talinum portulacifolium* were carried out against *Candida albicans* growth is controlled by a maximum zone of inhibition (15 mm) in silver nanoparticles. *Aspergillus flavus* growth was showed a minimum level (14 mm) in silver nanoparticles.

Numerous synthetic drugs have been employed to treat various diseases caused by pathogenic microbes in humans. However, these drugs often result in side effects due to overdose. Several medicinal plants have been identified and utilized to combat bacterial infections in humans. In this study, the antimicrobial activity of leaves from *Talinum portulacifolium* was evaluated against six pathogens, and the corresponding inhibition zones were documented. The medicinal properties of silver have been recognized for over 2,000 years, and since the nineteenth century, silver-based compounds have found applications in various antimicrobial uses. Silver nanoparticles, known for their diverse physical, biological, and pharmaceutical applications, are being employed as effective antimicrobial agents in public spaces such as railway stations and elevators in China, where they have demonstrated notable antimicrobial properties.

4. Conclusion

The present study enumerate that the terrestrial products are factories for the pharmaceutical products. Further analysis required to isolate the pure compound for the preparation of drugs in the pharmaceutical industries. The areas particularly bioprocess product exploitations are only in current focus that in itself has opened vistas in a technology upsurge. The order beneath the chaos at the genomic level is recognizable but not yet been fully understood. Among the different methods for NP synthesis, the chemical reduction method and green synthesis method were widely studied due to their advantage in controlling particle size and morphology. The NMR and wide applications of AgNPs in different fields could be analyzed in future.

References:

1. Absar, A., Shankar, S., & Murali, S. (2003). Geranium leaf biosynthesis of silver nanoparticles. Biotechnology Progress, 19, 1627–1631.
2. Amin, M., Anwar, F., Janjua, M. R. S. A., Iqbal, M. A., & Rashid, U. (2012). Green Synthesis of Silver Nanoparticles through Reduction with Solanum xanthocarpum L. Berry Extract: Characterization, Antimicrobial, and Urease Inhibitory Activities against Helicobacter pylori. International Journal of Molecular Sciences, 13, 9923-9941.
3. Anonymous. (2002). The Drugs and Cosmetics Act and Rule. In The Drugs and Cosmetics Act 1940. The Drugs and Cosmetics Rule 1945, Government of India, Ministry of Health and Family Welfare.
4. Kirby, A. W., Sherris, W. M., & Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. American Journal of Clinical Pathology, 45(4), 493-496.
5. Applerot, G., Lipovsky, A., Dror, R., Perkas, N., Nitzan, Y., Lubart, R., & Gedanken, A. (2009). Enhanced Antibacterial Activity of Nanocrystalline ZnO Due to Increased ROS-Mediated Cell Injury. Advanced Functional Materials, 19(6), 842-852.
6. Beveridge, T. J., & Murray, R. G. E. (1980). Site of metal deposition in the cell wall of Bacillus subtilis. Journal of Bacteriology, 141, 876-887.
7. Chandrasekharan, N., & Kamat, P. V. (2000). Improving the photoelectrochemical performance of nanostructured TiO2 films by adsorption of gold nanoparticles. The Journal of Physical Chemistry B, 104, 10851-10857.
8. Bauer, A. W., Kirby, W. M., Sherris, J. C., & Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. American Journal of Clinical Pathology, 45(4), 493-496.
9. Chandran, S. P., Chaudhary, M., Pasricha, R., Ahmad, A., & Sastry, M. (2006). Synthesis of gold nanotriangles and silver nanoparticles using Aloe vera plant extract. Biotechnology Progress, 22, 577-583.

10. Davis, J. (1994). Inactivation of antibiotics and the dissemination of resistance genes. Science, 264, 375-382.
11. Dubey, S. P., Lahtinen, M., & Sillanpaa, M. (2010). Tansy fruit mediated greener synthesis of silver and gold nanoparticles. Process Biochemistry, 45(7), 1065-1071.
12. Gamble, J. (1928). Flora of Presidency of Madras.
13. Pető, G., Molnár, G. L., Pászti, Z., Geszti, O., Beck, A., & Guczi, A. (2002). Electronic structure of gold nanoparticles deposited on SiOx/Si(100). Materials Science and Engineering: C, 19(1-2), 95-99.
14. Joy, P. H., & Johnson, I. (2015). Plant-mediated biosynthesis and characterization of silver nanoparticles by leaf extracts of Tragia involucrata, Cymbopogon citronella, Solanum verbascifolium, and Tylophora ovata. Karbala International Journal of Modern Science, 1, 237.
15. Krishnaraj, C., Jagan, E. G., Rajasekar, S., Selvakumar, S., Kalaichelvan, P. T., & Mohan, N. (2010). Synthesis of silver nanoparticles using Acalypha indica leaf extracts and its antibacterial activity against water-borne pathogens. Colloids and Surfaces B: Biointerfaces, 76, 50.
16. Leena, V., Hublikar, Sharanabasava, V., Ganachari, Narasimha Raghavendra, Nagaraj, R., Banapurmath, Veerabhadragouda, B., Patil, Yunus Khan, T. M., & Irfan Anjum Badruddin. (2021). Biogenesis of Silver Nanoparticles and Its Multifunctional Anti-Corrosion and Anticancer Studies. Coatings, 11, 1215.
17. Mann, S. (1993). Molecular tectonics in biomineralization and biomimetic materials chemistry. Nature, 365, 499-505.
18. Mathew, K. M. (1981). The Flora of the Tamil Nadu Carnatic, 2, 1459-1460.
19. Mulligen, A. N., Schaefer, H., & Lahaye, R. (1993). Evaluation of candidate DNA barcoding loci for economically important timber species of the mahogany family (Meliaceae). Molecular Ecology Resources, 11, 450-460.
20. Muthukumaran, P., Saraswathy, N., Kogilavani, R., Udhaya Bhaskar, S., & Sindhu, S. Preliminary Phytochemical Screening and Antimicrobial Properties of Pleurotus lorida and Pleurotus eous against some human pathogens: A Comparative Study. International Research Journal of Pharmacy, 5(2).
21. Parameswari, E., Udayasoorian, C., Sebastian, S. P., & Jayabalakrishnan, R. M. (2010). The Bactericidal Potential of Silver Nanoparticles. International Research Journal of Biotechnology, 1(3), 044-049.
22. Pastoriza-Santos, I., & Liz-Marzán, L. M. (2005). Colloidal silver nanoplates. State of the art and future challenges. Journal of Materials Chemistry, 18(15), 1724-1737.
23. Pavani, K. V., Sunilkumar, N., & Grayathramma, K. (2012). Plants as ecofriendly nanofactories. Journal of Biosciences, 6(1), 1-6.
24. Phatak, R. S., Hendre, A. S. Sunlight-induced green synthesis of silver nanoparticles using sun-dried leaves extract of Kalanchoe pinnata and evaluation of its photocatalytic potential. Der Pharmacia Lettre, 7, 313.
25. Piddock, K. J. V., & Wise, R. (1989). Mechanisms of resistance to quinolones and clinical perspective. Journal of Antimicrobial Chemotherapy, 23, 475-483.
26. Prashant Mohanpuria, Nisha K. Rana, Sudesh Kumar Yadav. (2008). Biosynthesis of nanoparticles: technological concepts and future applications. Journal of Nanoparticle Research, 10, 507–517.
27. Ruparelia, J. P., Chatterjee, A. K., Duttagupta, S. P., & Mukherji, S. (2008). Strain specificity in antimicrobial activity of silver and copper nanoparticles. Acta Biomaterialia, 4(3), 707-716.
28. Singh, A. P., Rawat, V. K., Behera, S. K., & Khare, P. B. (1992). Perspectives of pteridophytes biodiversity: a source of economy elevation. In National conference on biodiversity, development and poverty alleviation, Uttar Pradesh State Biodiversity Board, 46-49.
29. WHO. (2000). Methods and data sources for global burden of disease estimates 2000-2011. Geneva: Department of Health Statistics and Information Systems.
30. Wiley, B. J., Im, S. H., Li, Z., McLellan, J., Siekkinen, A., & Xia, Y. (2006). Maneuvering the surface plasmon resonance of silver nanostructures through shape-controlled synthesis. Journal of Physical Chemistry B, 110(32), 15666-15675.