Phytofabricated Silver Nanoparticles Derived from *Leea crispa* Leaf Extract: Antituberculosis and Anticancer Activities

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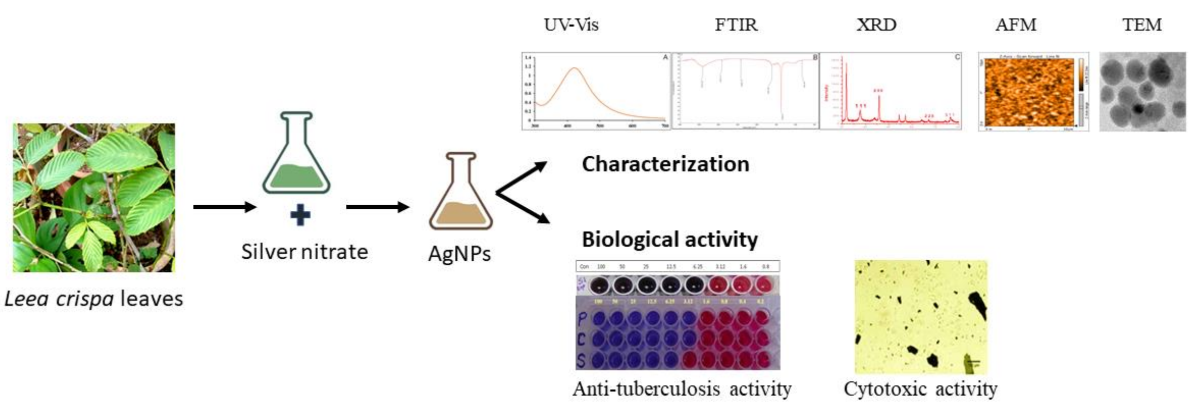
GRAPHICAL ABSTRACT



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Aqueous extracts of *Leea crispa* used for producing silver nanoparticles (AgNPs) with the supplementation of the external capping substance, were determined by UV-Visible spectroscopy. The synthesized nanoparticles were examined for their antituberculosis and anticancer activities. The presence of phytoconstituents available for reducing silver ions and to form the AgNPs was confirmed using FTIR analysis. The XRD and TEM examination validated the AgNPs' spherical particle shape and size of 15 to 85 nm and their face-centered cubic crystal form. Additionally, the FTIR spectrum revealed variation in the band values in the range 1384.0 to 3419.4 cm^{-1}, respectively, and the EDX noted a strong band at 3 keV induced the presence of metallic silver. The AgNPs exhibited comparatively potential anti-tuberculosis activity (0.2 to 100 µg/mL) respectively. Alternatively, various doses of AgNPs, 12.5 to 400 µg/mL documented considerable activity towards the human breast cancer cell lines. The percentage of cell viability increased at 12.5g/mL and declined at 400 ng/mL concentrations of AgNPs solution. The AgNPs synthesized from *L. crispa* exhibited potential activity against life-threatening tuberculosis and cancer cells.

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Keywords: Leea crispa; TEM; AgNO3; MTT Assay; MCF-7 Cell line; Anti-cancer; Anti-tuberculosis

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INTRODUCTION

Green synthesis of metallic nanoparticles has attracted research interest in nanomedicine in recent years because of their potential therapeutic applications (Kurian *et al.* 2023). Silver nanoparticles are extensively used in manufacturing medical products, sterilizing equipment, and drug delivery applications because of their unique biocompatibility such as anti-tuberculosis, anticancer, antiviral, anti-inflammation, and antibacterial agents (Abdel Hafez *et al.* 2020; Jain *et al.* 2021). Green-mediated nanoparticles are cost-effective, safe to handle, energy efficient, and environment friendly

(Aswathi *et al.* 2023). Synthesis of physico-chemical compounds produces hazardous reagents, high conductivity, high temperature, and requires costly equipment, more energy, and produces highly toxic by-products during their synthesis. Therefore, physical and chemical synthesis is inadequate (Venugopal *et al.* 2017). Green synthesis involves cheaper methods, such as plant-based products or plant parts, bacteria, algae, fungi, lichens, *etc.*, for the synthesis of AgNPs (Huq *et al.* 2022). These extracts contain diverse phyto-constituents including proteins, flavonoids, amino acids, alkaloids, saponins, polysaccharides, tannins, enzymes, carboxylic acids, aldehydes, ketones, *etc.* that can reduce Ag+ ions and stabilize the AgNPs into desired shapes and sizes (Saha *et al.* 2017). Many researchers have used plant parts for the synthesis of AgNPs, and these have shown numerous biological properties (Abdussalam-Mohammed *et al.* 2023). In contrast, biogenic AgNPs are used in topical ointments, and creams, to avoid infection from burns and wounds, silver materials including nano-devices, fabrics, polymers, consumer products, silver gel, *etc.* are used in medical-based industries (Song and Kim 2009). Silver is also called the stone of wealth because of its litheness, softening, and gleaming properties (Balaraman *et al.* 2020).

Leea crispa (Syn. *Leea asiatica*) is a well-known medicinal plant in the traditional Ayurveda and Unani systems. It is broadly scattered in the tropical and subtropical regions of the world. The phytochemical compounds from these plants are used in traditional medicine for asthma, as a snake antidote, blood coagulation, diabetes, hepatic disorder, and oxidative stress-related disorders (Hossain *et al.* 2021). Studies have suggested the presence of bioactive compounds in these plants, largely flavonoids and terpenoids (Sen *et al.* 2012), which could be associated with their extensive biological properties. ZnO nanoparticles synthesized from aqueous extract of these plants showed antioxidant, photocatalytic, and anti-cancer properties (Ali *et al.* 2021). *Leea coccinea*, a related species of this genus has shown significant antibacterial potentialities (del Carmen *et al.* 20121). These plant species serve as an excellent source for various phytochemicals. There are no reports of Ag nanoparticle synthesis and assessment of these nanoparticles for their anti-tuberculosis and anticancer effectiveness. Hence the current study aimed to synthesize the Ag nanoparticles using leaf extract and to evaluate the potential of these green synthesized nanoparticles for their antituberculotic and anticancer potentials.

EXPERIMENTAL

Materials

Silver nitrate was obtained for the experiment from Hi-media Laboratories in Mumbai, India. Human breast cancer cell line (MCF-7) was obtained from the National Centre for Cell Science (NCCS) in Pune, India. All chemicals and reagents used were analytical grade.

Collection and Preparation of Plant Extract

Fresh *L. crispa* leaves were procured from the Karnataka University Botanical Garden campus, Dharwad, Karnataka, India (15°26'12.6"N 74°59'00.4"E). The samples were identified, and the specimen copy was submitted to the Department of Botany, Karnatak University (Accession No. KUD-TU-001). The collected leaves were washed twice with tap water followed by Milliq water, soon after the leaves were shade dried for 4 hours under room temperature. Approximately 10 g of fresh leaves were boiled for 40 min at 80 °C in 100 mL of water. The aqueous solution cooled and filtered through the Whatman No. 1 filter paper.

Synthesis of AgNPs

The aqueous leaf extract of *L. crispa* (5 mL) was added to a 250-mL Erlenmeyer's flask containing 1 mM of 95 mL silver nitrate ($AgNO_3$) solution (final volume adjusted to 100 mL). The reaction mixture changed color from pale yellow to dark brown after 15 to 20 min, indicating the phytoreduction of silver ions and the formation of AgNPs. This manifested that the phytochemicals in the leaf extract acted as a reducing agent to generate AgNPs.

Characterization of Synthesized AgNPs

The synthesized AgNPs were characterized using spectral and microscopic methods. The absorption spectra of the sample were taken from 200 to 800 nm, using UV-visible spectroscopy (Jasco Corporation, Tokyo, Japan). The sample was measured using Fourier transform infrared (FTIR) analysis (FTIR System, Bruker, Germany) in the range of 4000 to 500 cm^{-1} to determine the functional groups utilized during the synthesis. X-ray diffraction (XRD) analysis was used to determine the crystalline nature, phase composition, and size of the AgNPs (Rigaku Miniflex 600 Japan). The sample of AgNPs was measured at angle 2θ ranging from 30° to 80°. The surface morphology was determined using AFM (Nano Surf Flex AFM). The morphological features of AgNPs mainly the size and shape of nanoparticles were analyzed using high-resolution-transmission electron microscopy (HR-TEM) analysis (Hitachi, Model: S-3400N).

Antituberculosis Activity of Synthesized AgNPs

The antituberculosis (TB) activity of AgNPs was evaluated using the blue dye alamar micro-plate method against *Mycobacterium tuberculosis* H37 Rv strain (ATCC No-27294) (Radhakrishnan *et al.* 2014). This approach is safe, uses thermally stable reagents, and has a high correlation with the BACTEC radiometric method. To prevent evaporation of medium in the test wells during incubation, about 200L of sterile deionized water was transferred to all outside perimeter wells of sterile 96 well plates. The 96 well plates were filled with 100 µL of Middlebrook (7H9 broth) and a serial dilution of AgNPs was performed on the plates in real time. The amounts of biogenic AgNPs examined ranged from 100 to 0.2 µg/mL. The pyrazinamide, ciprofloxacin, and streptomycin were 3.125, 3.125, and 6.25 µg/mL, respectively, used as standard. All the examined plates were sealed with semi-transparent parafilm and incubated at 37 °C for 5 days. After 24 h, 25L of freshly prepared alamar blue reagent (1:1) and polysorbate 80 (Tween-80) (10%) were added to the plate and incubated for another 24 h. The absence of blue color indicates the absence of bacterial growth, whereas the presence of pink color indicates the presence of bacterial growth. The smallest MIC dose prevents the coloration from shifting from blue to pink.

Anti-cancer Activity of Synthesized AgNPs

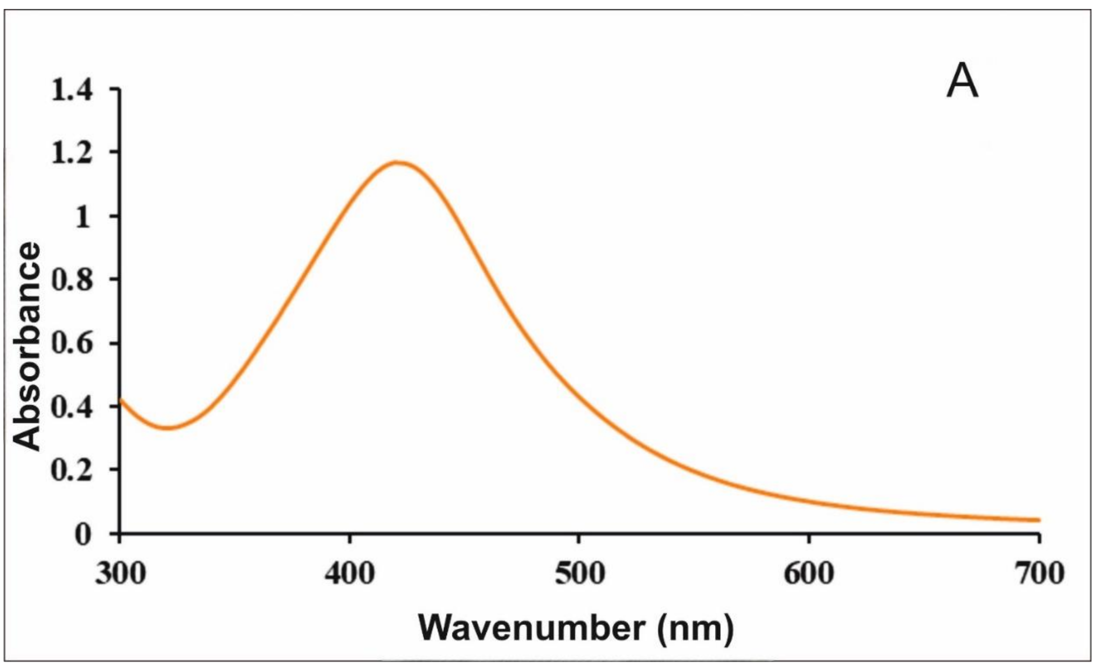
The MTT assay was used to determine the effect of AgNPs on cell augmentation. MCF-cell line was obtained from the NCCS in Pune, India. Subculture cells were held overnight at 37 °C, 95% humidity, and 5% CO_2. Control medium (cell-free medium). The MCF-7 cells were then plated in 96-well flat-bottom microplates (Cat No-15240062) for 24 h to grow. The cells were then treated for 24 h with biogenic AgNPs at doses of 12.5, 25, 50, 100, 200, and 400 g/mL. The cell-containing wells were washed twice with phosphate buffer solution before inserting approximately 20 µL (5mg/mL in phosphate buffer solution) MTT staining solution into each well. The plate was incubated at 37 °C for 4 h and the absorbance was measured with a 570 nm microplate reader.

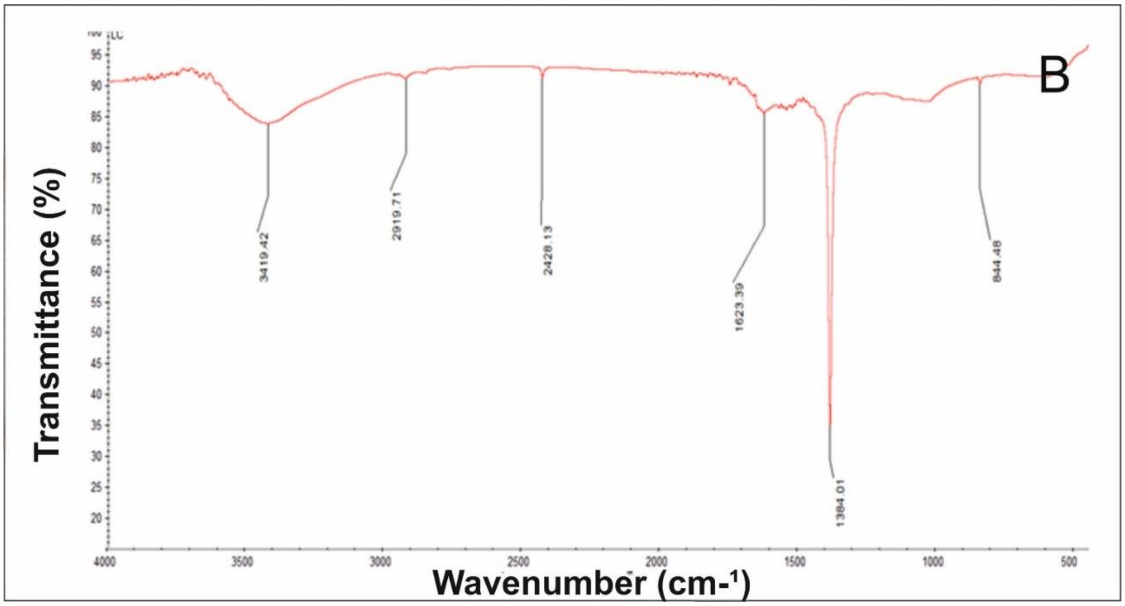
RESULTS

Characterization of Synthesized Silver Nanoparticles

Synthesis of nanoparticles can be observed based on the characteristic change in the color of the solution when metal ions interact with the plant extract containing various phytochemicals. The phytoconstituents like flavonoids and terpenoids *etc.* are involved in the capping and stabilizing of nanoparticles (Sen *et al.* 2012). The color changed from bright yellow to dark brown, suggesting that silver ions were reduced and AgNPs were formed. UV-visible spectroscopy was used to perform preliminary characterization of isolated AgNPs from *L. crispa* leaf extract. A characteristic peak was observed at 417 nm (Fig. 1A).

The FTIR results of leaf extract synthesized from *L. crispa* showed different wavelengths (Fig. 1B). The absorbance peaks were at 3419.42, 2919.71, 2428.13, 1623.39, and 1384.01 cm^{-1}. The absorbance peak 3419.42 cm^{-1} is assigned to the O-H stretching and vibration of phenol or alcoholic compounds. The crystallinity of the biogenic AgNPs synthesized from *L. crispa* leaf extract was observed by XRD analysis (Fig.1C). The major diffraction peaks were detected at (111), (200), (220), and (311), with strong peaks indexed as 38.45°, 54.12°, 64.26°, and 78.48° planes of face centered cubic (FCC) lattice of AgNPs (Fig. 1C)





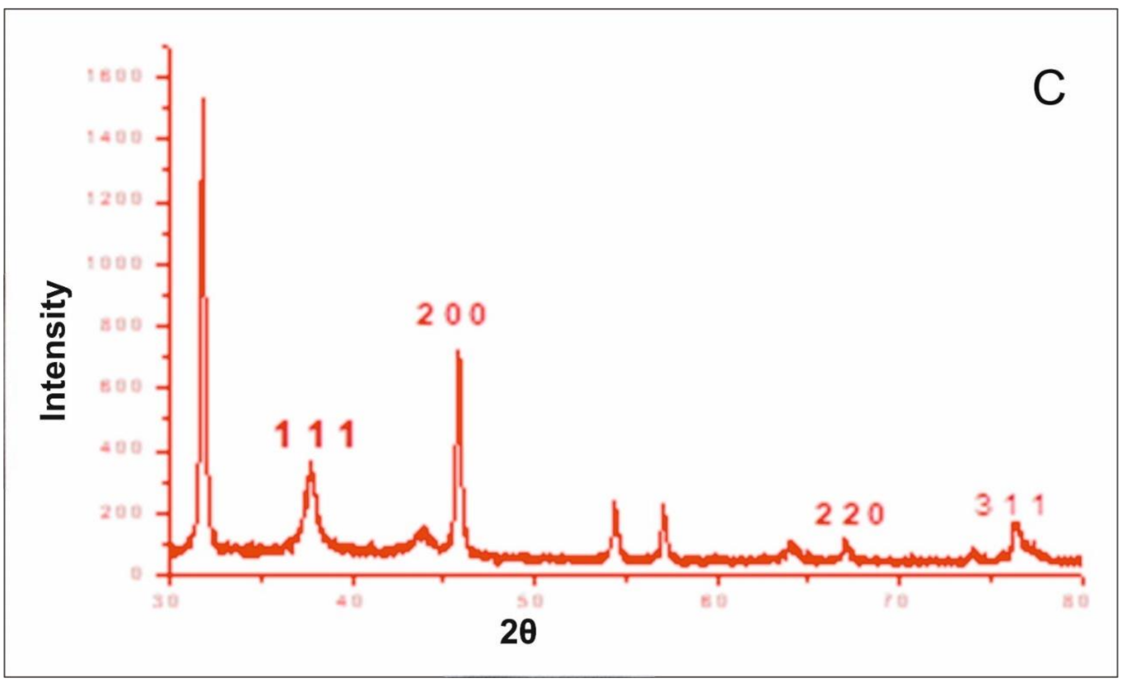


Fig. 1. (A) UV-Visible spectroscopy analysis; (B) FTIR spectra, and (C) XRD spectrum showing the peaks of biogenic AgNPs from leaf extract of *L. crispa*

The AFM micrograph shows the surface roughness and morphology of AgNPs (Fig. 2). The synthesized AgNPs were spherical, homogeneous, and anisotropic in shape. The form of AgNPs was comparable to that of the authors' previous investigations. The crystalline size of AgNPs was found to range between 32 and 50 nm. The HR-TEM examination of AgNPs from *L. crispa* leaf extract reveals the nanoparticles' crystalline structure and surface shape. The particle exhibited lattice fringes and had an size range of 15 to 85 nm (Fig. 3). Finally, the AFM and TEM tests were found consistent with earlier observations.

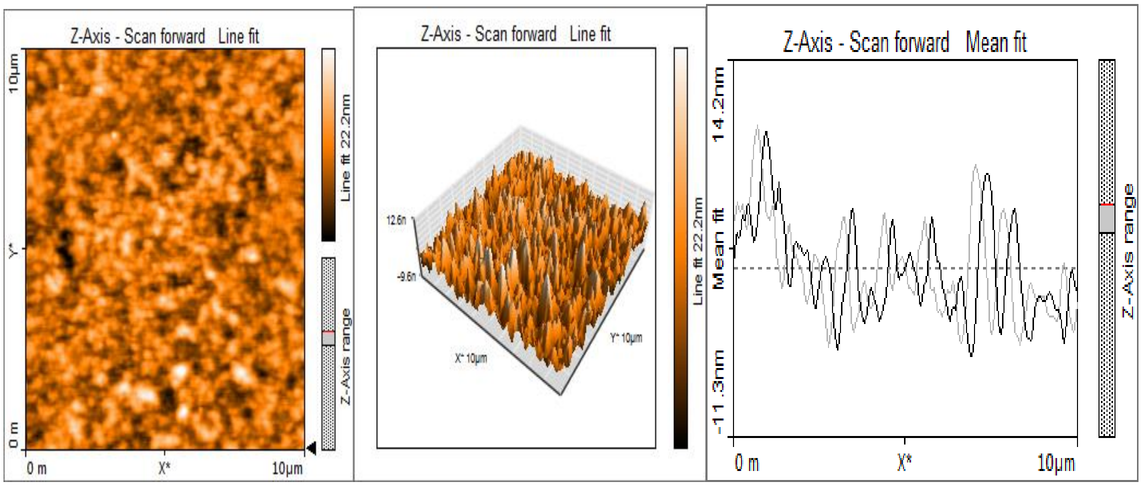


Fig. 2. The AFM analysis of AgNPs from leaves extract of *L. crispa*

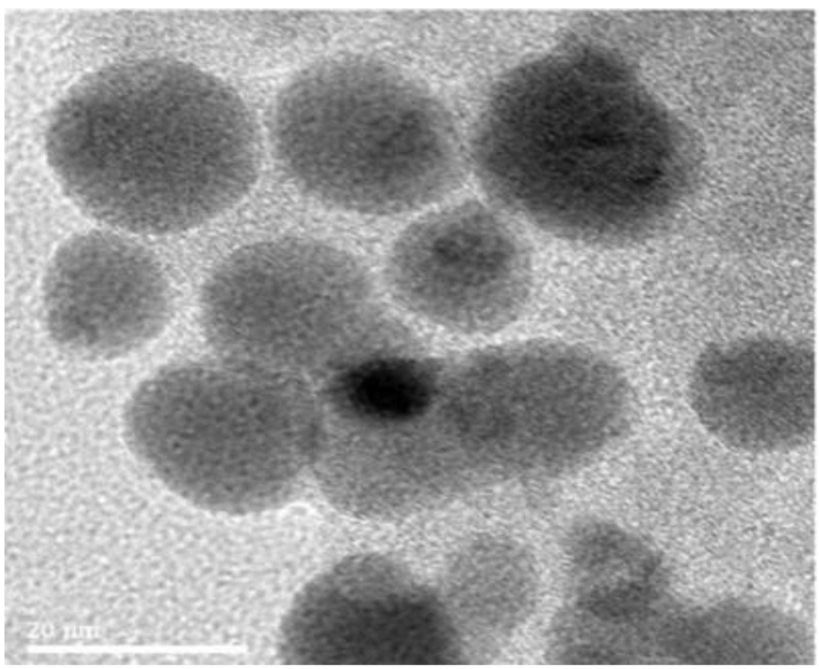


Fig. 3. The HR-TEM images of AgNPs from leaves extract of *L. crispa*

Anti-tuberculosis Activity of AgNPs

This study demonstrated the biogenic AgNPs derived from *L. crispa* leaf extract and their anti-tuberculosis effectiveness. First-line medications for active tuberculosis include pyrazinamide, rifampicin, polyethylene glycol, and streptomycin (Fig. 4 and Table 1). In line with earlier reports, the biogenic AgNPs from leaves extract of *L. crispa* showed inhibition activity against clinical isolates of *M. tuberculosis* H37Rv strain. The bacteria treated with various concentration of AgNPs solution *viz.* 0.2 0.8, 1.6, 3.12, 6.25, 12.5, 25, 50, and 100 µg/mL (Table 1). Bacterial sensitivity increased as AgNP concentration increased. After incubation, it appears blue in the well, indicating no bacterial growth, and pink in the well, indicating growth. Bacterial growth was inhibited by AgNPs concentrations ranging from 6.25 to 100 µg/mL. Antibiotics, such as pyrazinamide, ciprofloxin, and streptomycin, showed the least inhibitory impact at 3.12 µg/mL concentrations of AgNPs solution, with streptomycin showing at 6.25 µg/mL concentrations.

Table 1. Anti-tuberculosis Activity of AgNPs

Samples	Concentrations (µg/mL)								
	100	**50**	**25**	**13**	**6.25**	**3.12**	**1.6**	**0.8**	**0.2**
AgNPs	S	S	S	S	S	R	R	R	R
$AgNO_3$	R	R	R	R	R	R	R	R	R
Leaf extract	S	R	R	R	R	R	R	R	R
Pyrazinamide	S	S	S	S	S	S	R	R	R
Ciprofloxin	S	S	S	S	S	S	R	R	R
Streptomycin	S	S	S	S	S	R	R	R	R

Note: S- Sensative; R- Resistance

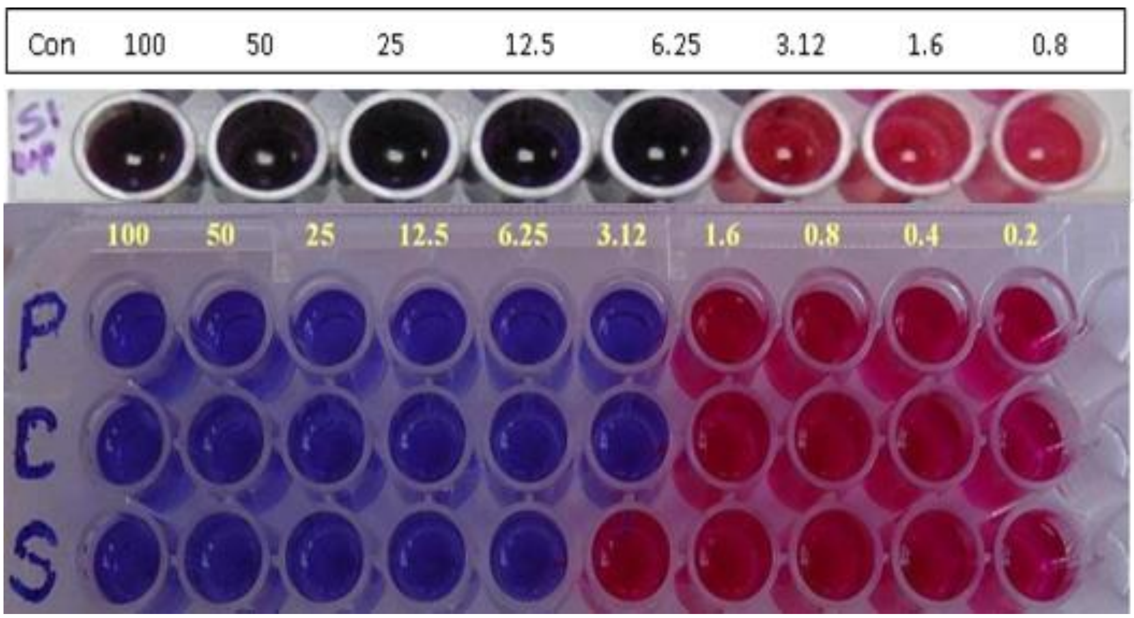


Fig. 4. Photographs showing Anti-tuberculosis activity of AgNPs (P- Pyrazinamide, C- Ciprofloxin, S- Streptomycin)

Cytotoxic Activity

In the present investigation, a human breast cancer line was selected because of the excessive prevalence in India. Therefore, *L. crispa* leaf mediated synthesis of AgNPs were used against MCF-7 cell line (Fig. 5). Cell viability percentages of MCF-7 cell line treated with various concentrations of fabricated AgNPs *viz.* 12.5, 25, 50, 100, 200, and 400 µg/mL of AgNPs were 98.99, 89.07, 79.21, 28.25, 20.01, and, 15.14% respectively (Table 2). Biogenic AgNPs (12.5 µg/mL) at low concentrations showed the highest percentage (98.09%) of cytotoxic activity, when compared with leaf extract and control. The biogenic AgNPs showed concentration-dependent cytotoxic effects on *in-vitro* human breast cancer cell lines. Studies on *Leea crispa*-mediated ZnO nanoparticles showed similar activity against breast cancer cell lines using MTT assay, where 87% reduction was seen when concentraction and time increases at low concentrration (5 µg/mL) when compared to 23%

(20 µg/mL) (Ali *et al.* 2021). The experimental values of IC_{50} were determined as 298.96, 251.60, and 80.71 µg/mL for paclitaxel, silver nitrate, and AgNPs, respectively. The biogenic AgNPs showed exceptional anticancer activity compared with standard paclitaxel.

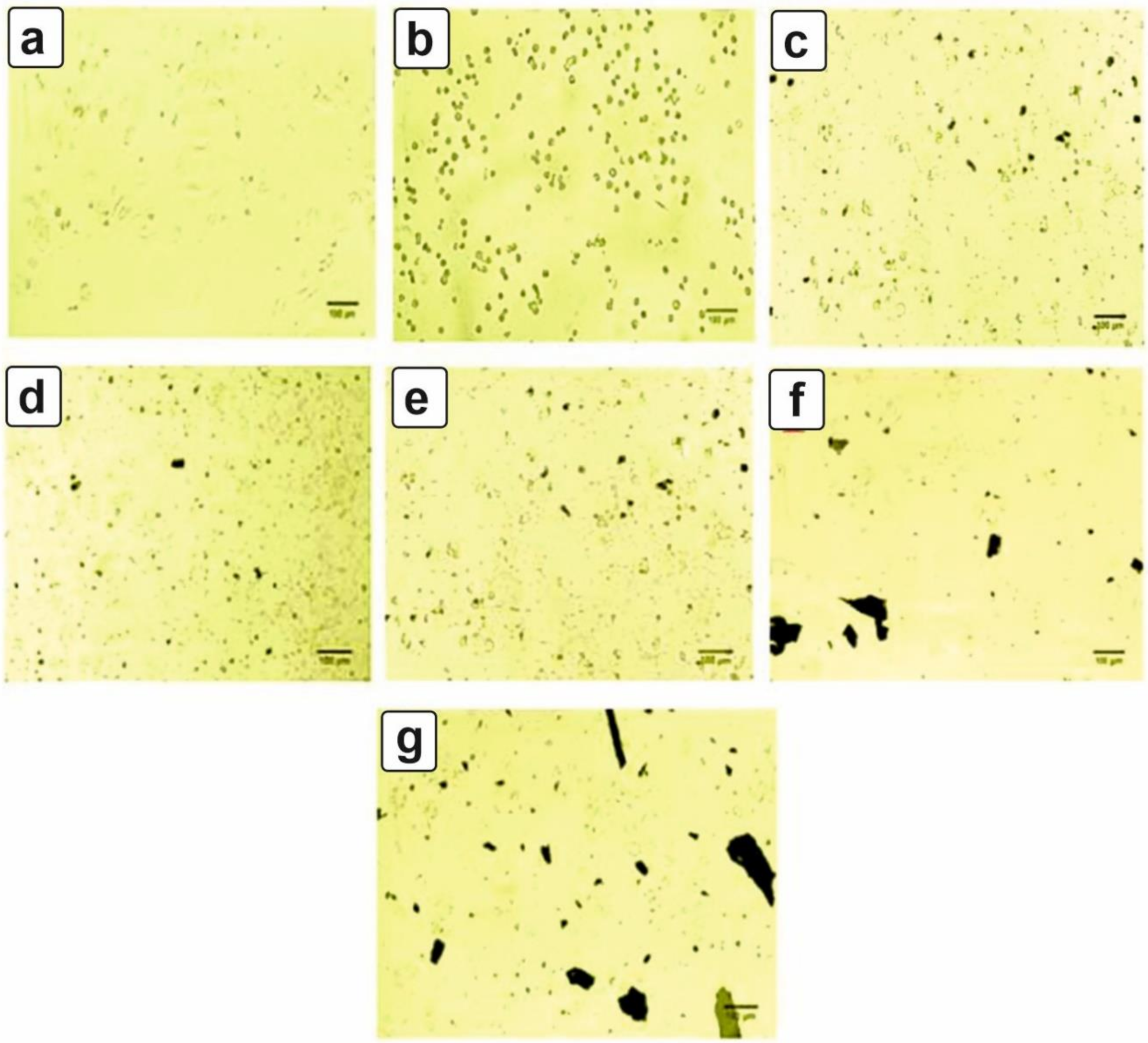


Fig. 5. Morphological observation of MCF-7 cells after incubation of AgNPs: a) Medium with untreated cells,b) Paclitaxel (standard), c) Silver nitrate, (d through g) treated with different concentrations (12.5 to 400 µg/mL) of AgNPs (Scale bar - 100µm)

Table 2. Cytotoxic Activity of AgNPs Fabricated from *L. crispa* Leaf Extract

Concentration of AgNPs (µg/mL)	Leaf Extract	AgNPs
400	42.48	15.14
200	55.35	20.01
100	71.11	28.95
50	83.33	79.21
25	90.24	89.07
12.5	97.00	98.09
Control	100 Cells (MCF-7)	

DISCUSSION

Temperature is an important component in the production of nanoparticles. The pH of the aqueous solution was changed to 8, 9, and 10 levels. The specimen prepared at pH 9 showed a large absorbance peak at 418 nm, which can be attributed mostly to the stimulation of AgNPs' Surface Plasmon Resonance (SPR) vibrations (Subbaiya *et al.* 2017; Raj *et al.* 2020). Furthermore, when compared to larger absorbance peaks, the narrow absorbance peak of AgNPs demonstrates nano-sized particles. The FTIR results showed noticeable peaks, suggested the synthesis of nanoparticles. Similar absorbance peaks were observed in AgNPs synthesized by *Amorphophallus paeoniifolius* tuber extract (Nayem *et al.* 2020). The short peak at 2919.7 cm^{-1} likely corresponds to asymmetric and symmetric (C-H) vibrations of a methylene group ($-CH_2$) of aliphatic compounds. The prominent band 2428.1 cm^{-1} is ascribed to the stretching of the phosphine (P-H) functional group (Nayaka *et al.* 2020). The intense peak at 1623.4 cm^{-1} indicates the presence of an amide group for the formation AgNPs (Muslim and Owaid 2019). The band 1384.0 cm^{-1} can be ascribed to the C-H bending vibration of AgNPs. From the results, it is evident that the identified phytochemicals such as proteins, lipids, phenolic compounds, flavonoids, alkaloids, and tannins are likely involved in the capping, reduction, and stabilization of AgNPs.

The XRD analysis revealed noticeable peaks, indicating that the nanoparticles were crystalline. The approximate sizes of produced AgNPs were estimated as 23, 18, 13, and 10 (Debye-Scherrer equation) (Table 1). The obtained findings corresponded to JCPDS card number 04-0783. The current findings corroborate prior findings, utilizing tea leaf extract. The zeta potential of -41.6 mV of the synthesized nanoparticles indicated its stability (Tengku Sallehudin *et al.* 2018). The high negative values ranged between -40 and -50 mV, illustrating the repulsion of the particles and giving tremendous stability to the AgNPs (Ajitha *et al.* 2015; Nadzir *et al.* 2019). The results reveal that negatively charged nanoparticles reduce nanoparticle aggregation and also exhibit long-term stability (Elamawi *et al.* 2018). The negatively charged nature of the particles may be due to the capping of biochemicals present in the leaf extract of *L. crispa.*

Metallic nanoparticles have distinct antiviral, antibacterial, and antiparasitic properties, making them promising candidates for future use in infectious disease treatment. AgNPs can disrupt metabolic processes, hence inhibiting mycobacterial growth (Chopade *et al*. 2016). Patil and Taranath (2016) reported using *Limonia acidissima* leaf-mediated synthesis of ZnO nanoparticles and showed growth inhibition activity of *M. tuberculosis*. The current study demonstrated the minimum dosage (80.7 μg/mL) of AgNPs needed to trigger the breast cancer cell line. In contrast, biogenic AgNPs may stimulate reactive oxygen species (ROS) and damage the cellular components, which leads to necrosis or death (Vivek *et al.* 2012). The intercellular ROS production increased, and antioxidant status was decreased in MCF-7 cancer cells (Erdogan *et al.* 2019). The cytotoxic property of AgNPs is due to the release of silver cations leading to DNA damage by disruption of intercellular macromolecules (Wang and Kirsch 2006). The green-mediated AgNPs from *Syzygium aromaticum* crude extract showed efficient anticancer activities against MCF-7 breast and A549 lung cancer cell lines (Venugopal *et al.* 2017). In addition, biogenic AgNPs are cost-effective, produce safer products, and cannot damage normal cells at higher concentrations. The bulk AgNPs show minimum inhibitory effect, but the tiny-sized nanoparticles act like a magic bullet that only treats the infected site. Hence, care should be taken for their use in biomedical applications.

CONCLUSIONS

1. The present investigation suggests that green synthesized silver nanoparticles (AgNPs) are a potential source for a cost-effective, non-toxic, and environmentally friendly product that can be used in various biomedical applications.
2. The biogenic AgNPs were spherical, and their size ranged from 10 to 96 nm. These nanoparticles showed potential anti-tuberculosis activity in the minimum concentration of AgNPs solution, suggesting their potential to develop into a novel and cost-effective drug.
3. The study also highlights the novelty of *Leea crispa* mediated green synthesized nanoparticles to be used as a potential source for various biomedical applications which can help researchers develop cost-effective and site-specific targeted nanoparticles.

DECLARATIONS

Data Availability Statement

The data presented in this study are available on request from the corresponding authors.

Conflicts of Interest

The authors hereby declare that they have no conflict of interest and have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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REFERENCES CITED

Abdel Hafez, O. H., Ali, T. F. S., Fahim, J. R., Desoukey, S. Y., Ahmed, S., Behery, F. A., Kamel, M. S., Gulder, T. A., and Abdelmohsen, U. R. (2020). "Anti-inflammatory potential of green synthesized silver nanoparticles of the soft coral *Nephthea* sp. supported by metabolomics analysis and docking studies," *International Journal of Nanomedicine* 15, 5345-5360. DOI: 10.2147/IJN.S239513

Abdussalam-Mohammed, W., Mohamed, L., Abraheem, M. S., Mansour, M. M. A., and Sherif, A. M. (2023). "Biofabrication of silver nanoparticles using *Teucrium apollinis* extract: Characterization, stability, and their antibacterial activities," *Chemistry* 5(1), 54-64. DOI: 10.3390/chemistry5010005

Ajitha, B., Ashok Kumar Reddy, Y., and Sreedhara Reddy, P. (2015). "Green synthesis and characterization of silver nanoparticles using *Lantana camara* leaf extract," *Materials Science and Engineering: C* 49, 373-381. DOI: 10.1016/j.msec.2015.01.035

Ali, S., Sudha, K. G., Karunakaran, G., Kowsalya, M., Kolesnikov, E., and Rajeshkumar, M. P. (2021). "Green synthesis of stable antioxidant, anticancer and photocatalytic activity of zinc oxide nanorods from *Leea asiatica* leaf," *Journal of Biotechnology* 329, 65-79. DOI: 10.1016/j.jbiotec.2021.01.022

Aswathi, V. P., Meera, S., Maria, C. G. A., and Nidhin, M. (2023). "Green synthesis of nanoparticles from biodegradable waste extracts and their applications: A critical review," *Nanotechnology for Environmental Engineering* 8(2), 377-397. DOI: 10.1007/s41204-022-00276-8

Balaraman, P., Balasubramanian, B., Kaliannan, D., Durai, M., Kamyab, K., Park, S., Chelliapan, S., Lee, C-T., Maluventhen, V., and Maruthupandian, A. (2020). "Phyco-synthesis of silver nanoparticles mediated from marine algae *Sargassum myriocystum* and its potential biological and environmental applications," *Waste and Biomass Valorization* 11, 5255-5271. DOI: 10.1007/s12649-020-01083-5

del Carmen, Travieso Novelles., María, *et al.* (2021). "Biosynthesis of fluorescent silver nanoparticles from *Leea coccinea* leaves and their antibacterial potentialities against *Xanthomonas phaseoli* pv *phaseoli*," *Bioresources and Bioprocessing* 8, 1-11. DOI: 10.1186/s40643-020-00354-2

Chopade, B. A., Singh, R., Nawale, L., Arkile, M., Wadhwani, S., Shedbalkar, U., Chopade, S., and Sarkar, D. (2016). "Phytogenic silver, gold, and bimetallic nanoparticles as novel antitubercular agents," *International Journal of Nanomedicine* 11, 1889-1897. DOI: 10.2147/IJN.S102488

Elamawi, R. M., Al-Harbi, R. E., and Hendi, A. A. (2018). "Biosynthesis and characterization of silver nanoparticles using *Trichoderma longibrachiatum* and their effect on phytopathogenic fungi," *Egyptian Journal of Biological Pest Control* 28(1), article 28. DOI: 10.1186/s41938-018-0028-1

Erdogan, O., Abbak, M., Demirbolat, G. M., Birtekocak, F., Aksel, M., Pasa, S., and Cevik, O. (2019). "Green synthesis of silver nanoparticles *via Cynara scolymus* leaf extracts: The characterization, anticancer potential with photodynamic therapy in MCF7 cells," *PLOS ONE* 14(6), article ID e0216496. DOI: 10.1371/journal.pone.0216496

Hossain, F., Mostofa, Md. G., and Alam, A. K. (2021). "Traditional uses and pharmacological activities of the genus *Leea* and its phytochemicals: A review," *Heliyon* 7(2), article ID e06222. DOI: 10.1016/j.heliyon.2021.e06222

Huq, Md. A., Ashrafudoulla, Md., Rahman, M. M., Balusamy, S. R., and Akter, S. (2022). "Green synthesis and potential antibacterial applications of bioactive silver nanoparticles: A review," *Polymers* 14(4), article 742. DOI: 10.3390/polym14040742

Jain, N., Jain, P., Rajput, D., and Patil, U. K. (2021). "Green synthesized plant-based silver nanoparticles: Therapeutic prospective for anticancer and antiviral activity," *Micro and Nano Systems Letters* 9(1), article 5. DOI: 10.1186/s40486-021-00131-6

Kurian, J. T., Balasubramanian, B., Meyyazhagan, A., Pappuswamy, M., Alanazi, A. M., Rengasamy, K. R., Arumugam, V. A., Sebastian, J. K., and Chen, J.-T. (2023). "One-pot synthesis of silver nanoparticles from *Garcinia gummi-gutta*: Characterisation, antimicrobial, antioxidant, anti-cancerous and photocatalytic applications," *Frontiers in Bioscience-Landmark* 28(8), article 169. DOI: 10.31083/j.fbl2808169

Muslim, R. F., and Owaid, M. N. (2019). "Synthesis, characterization, and evaluation of the anti-cancer activity of silver nanoparticles by natural organic compounds extracted from *Cyperus sp.* rhizomes," *ACTA Pharmaceutica Sciencia* 57(2), article 129. DOI: 10.23893/1307-2080.APS.05708

Nadzir, M. M., Idris, F. N., and Hat, K. (2019). "Green synthesis of silver nanoparticle using *Gynura procumbens* aqueous extracts," *AIP Conference Proceedings* 2124, article ID 030018. DOI: 10.1063/1.5117140

Nayaka, S., Bhat, M. P., Chakraborty, B., Pallavi, S. S., Airodagi, D., Muthuraj, R., Halaswamy, H. M., Dhanyakumara, S. B., Shashiraj, K. N., and Kupaneshi, C. (2020). "Seed extract-mediated synthesis of silver nanoparticles from *Putranjiva roxburghii* Wall., phytochemical characterization, antibacterial activity and anticancer activity against MCF-7 cell line," *Indian Journal of Pharmaceutical Sciences* 82(2), article 646. DOI: 10.36468/pharmaceutical-sciences.646

Nayem, S. M. A., Sultana, N., Haque, Md. A., Miah, B., Hasan, Md. M., Islam, T., Hasan, Md. M., Awal, A., Uddin, J., Aziz, Md. A., *et al.* (2020). "Green synthesis of gold and silver nanoparticles by using *Amorphophallus paeoniifolius* tuber extract and evaluation of their antibacterial activity," *Molecules* 25(20), article Number 4773. DOI: 10.3390/molecules25204773

Patil, B. N., and Taranath, T. C. (2016). "*Limonia acidissima* L. leaf mediated synthesis of zinc oxide nanoparticles: A potent tool against *Mycobacterium tuberculosis*," *International Journal of Mycobacteriology* 5(2), 197-204. DOI: 10.1016/j.ijmyco.2016.03.004

Raj, S., Singh, H., Trivedi, R., and Soni, V. (2020). "Biogenic synthesis of AgNPs employing *Terminalia arjuna* leaf extract and its efficacy towards catalytic degradation of organic dyes," *Scientific Reports* 10(1), article ID 9616. DOI: 10.1038/s41598-020-66851-8

Saha, J., Begum, A., Mukherjee, A., and Kumar, S. (2017). "A novel green synthesis of silver nanoparticles and their catalytic action in reduction of Methylene blue dye," *Sustainable Environment Research* 27(5), 245-250. DOI: 10.1016/j.serj.2017.04.003

Song, J. Y., and Kim, B. S. (2009). "Rapid biological synthesis of silver nanoparticles using plant leaf extracts," *Bioprocess and Biosystems Engineering* 32(1), 79-84. DOI: 10.1007/s00449-008-0224-6

Sen, S., De, B., Devanna, N., and Chakraborty, R. (2012). "Anthelmintic and *in vitro* antioxidant evaluation of fractions of methanol extract of *Leea asiatica* leaves," *Ancient Science of Life* 31(3), 101-106. DOI: 10.4103/0257-7941.103184

Subbaiya, R., Saravanan, M., Priya, A. R., Shankar, K. R., Selvam, M., Ovais, M., Balajee, R., and Barabadi, H. (2017). "Biomimetic synthesis of silver nanoparticles from *Streptomyces atrovirens* and their potential anticancer activity against human breast cancer cells," *IET Nanobiotechnology* 11(8), 965-972. DOI: 10.1049/iet-nbt.2016.0222

Tengku Sallehudin, T. A., Abu Seman, M. N., and Tuan Chik, S. M. S. (2018). "Preparation and characterization silver nanoparticle embedded polyamide nanofiltration (NF) membrane,"*MATEC Web of Conferences* 150, article ID 02003. DOI: 10.1051/matecconf/201815002003

Venugopal, K., Rather, H. A., Rajagopal, K., Shanthi, M. P., Sheriff, K., Illiyas, M., Rather, R. A., Manikandan, E., Uvarajan, S., Bhaskar, M., *et al.* (2017). "Synthesis of silver nanoparticles (Ag NPs) for anticancer activities (MCF 7 breast and A549 lung cell lines) of the crude extract of *Syzygium aromaticum*," *Journal of Photochemistry and Photobiology B: Biology* 167, 282-289. DOI: 10.1016/j.jphotobiol.2016.12.013

Vivek, R., Thangam, R., Muthuchelian, K., Gunasekaran, P., Kaveri, K., and Kannan, S. (2012). "Green biosynthesis of silver nanoparticles from *Annona squamosa* leaf extract and its*in vitro* cytotoxic effect on MCF-7 cells," *Process Biochemistry* 47(12), 2405-2410. DOI: 10.1016/j.procbio.2012.09.025

Wang, W., and Kirsch, T. (2006). "Annexin V/β5 Integrin interactions regulate apoptosis of growth plate chondrocytes," *Journal of Biological Chemistry* 281(41), 30848-30856. DOI: 10.1074/jbc.M605937200

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