



GREEN SYNTHESIS AND ANTIBACTERIAL ACTIVITY OF SILVER NANOPARTICLES SYNTHESIZED USING LEMON EXTRACT

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ABSTRACT

The present research looked into the antibacterial activity AgNPs made environmentally friendly utilizing lemon extract. TEM, EDX, and FTIR were used to characterize the synthesized AgNPs. The outcomes demonstrated that 20–30 nm-sized AgNPs were successfully synthesized. According to the FTIR spectrum, Functional groups in the lemon extract were responsible for the occurrence of the reduction of silver ions to AgNPs. The MICs of the AgNPs were 8 and 16 g/mL, respectively, and them significantly Staphylococcus aureus is gram-positive, and Escherichia coli is gram-negative. bacteria's growth was hindered. AgNPs with potential antibacterial action can be made environmentally friendly and economically by employing lemon extract in the green synthesis process. In our research study we used lemon extract silver nano particles Staphylococcus aureus and Escherichia coli, two dangerous microorganisms, for antibacterial activity. This research study proved that green synthesized silver nano particles showed biological activity like antibacterial activity against pathogenic bacteria, these Nano particles can use in Nano medicine as pharm applications.

Key words: Lemon: Nano; Silver Particles; Pathogenic Bacteria

INTRODCUTION

Due to their distinct physical, chemical, and biological features, silver nanoparticles (AgNPs) in the field of nanotechnology, a lot of interest is being generated. They have a wide range of uses in many different industries, including electronics, photonics, catalysis, and biology. Toxic chemicals and solvents are used in the traditional procedures for synthesizing AgNPs, which raises questions concerning environmental contamination and toxicity. Therefore, there is a need for AgNP synthesis techniques that are both economical and environmentally beneficial. A green synthesis of AgNPs has employed plant extracts. i.e both more economical and environmentally

benign than traditional processes. Plant extracts contain a variety of phytochemicals that work as lowering and capping agents for the creation of AgNPs, including flavonoids, terpenoids, and phenolic compounds. One such plant extract that has the ability to convert silver ions into AgNPs is lemon extract, which also contains other phytochemicals including citric acid. The environmentally benign and economically advantageous Plant extracts have replaced conventional methods for the "green" production of AgNPs. Numerous phytochemicals found in plant extracts, including flavonoids, terpenoids, and phenolic compounds, serve as reducing and capping agents for the synthesis of AgNPs. One such plant extract that includes citric acid and other phytochemicals that may decrease silver ions to AgNPs is lemon extract. AgNPs have also been widely researched for one more essential quality is their antimicrobial action. AgNPs are attractive candidates for antimicrobial applications since they have been shown to demonstrate substantial antimicrobial action against certain gram-positive and gram-negative bacteria. In this study, we wanted to synthesize AgNPs using lemon extract and examine their *Staphylococcus aureus* and *Escherichia coli* are examples of gram-positive and gram-negative bacteria that can be effectively treated with an antibiotic. AgNPs lemon extract can be used to create new antibacterial compounds that have a variety of applications in an eco-friendly and economical manner. Gold nanoparticles (AuNPs) are among the most alluring metal nanoparticles (NPs) because of their distinctive physical and chemical properties, good biocompatibility, high dispersibility, high stability, and low toxicity (Siddiqi *et al.*, 2017, Tahernejad-Javazmi *et al.*, 2018, Jahandari *et al.*, 2019). Microorganisms that cause diseases in humans are known as pathogenic bacteria. Humans survive on food, water, and clothing. To treat some disorders caused by bacteria, medical treatments—mostly drugs—are utilised and created. However, today, instead of treating diseases, People prefer to use antibacterial chemicals in items like food containers, cosmetics, clothing, and surgical supplies to prevent the bacteria from spreading (Abbasi *et al.*, 2016). The creation of antibacterial agents is now receiving some attention for this reason. At current time, metal nanoparticles' exceptional qualities can be developed and modified through the use of nanotechnology to achieve biomedical application objectives (Franci *et al.*, 2015). The study of the production, application, and manipulation of materials at the nanoscale is known as nanotechnology. one and one hundred nanometers in size (Iravani, 2011; Moodley *et al.*, 2018). The transformation of metal ions into metal nanoparticles is accomplished using phytochemicals and a few natural substances (Azam *et al.*, 2012). The scientific community is

attracted by the special features of nanoparticles and is constantly looking for new ways to use them in research and engineering (Vilchis-Nestor *et al.*, 2008). The creation of nanoparticles in a variety of sizes and forms is the subject of nanotechnology (Oves, *et al.*, 2013; Sau & Rogach, 2010). Recent advances in Nano sciences have sparked an increase in the number of scientific research areas that focus on studying nanoparticles and using plants to create nanoparticles in a greener manner (Kuppusamy, 2011).

MATERIALS AND METHODS

In our research, we used several chemicals and strains to carry out the experiments and we purchased the chemicals, namely, Methanol, acetone, and distilled water, silver nitrate from the SU Chemicals Pvt. Ltd from Chennai, Tamilnadu.

Preparation of Lemon fruit extract

In a local Chennai market, lemons were purchased. With distilled water, the fruits were cleaned and then diced. Separately, 80 ml of distilled water and 8 g of fruit were added, and both were heated for 2 to 5 minutes. The resulting lemon Using filter paper, extract was filtered. Every filter was then collected separately and for future use, maintained at 4°C.

Preparation of Silver Nanoparticles

Three milliliters of each fruit extract were added to 40 millilitres of a 1 mM aqueous AgNO₃ solution in a conical flask. After that, the combination was kept at 37°C for five hours in total darkness. Colloidal suspension is hue changed from colorless to reddish brown, indicating the biogenesis of silver nanoparticles



Lemon fruit

Before the formation
of AgNps

After the formation
of AgNps

Characterization

UV-Vis Absorption study

A Labomed double-beam UV-visible spectrophotometer with a 1.0 nm of spectral bandwidth was used to record the L-AgNPs' UV-Vis absorption spectra, which were created. at various time

intervals. To verify the stability of L-AgNPs samples, absorption spectra were also captured.

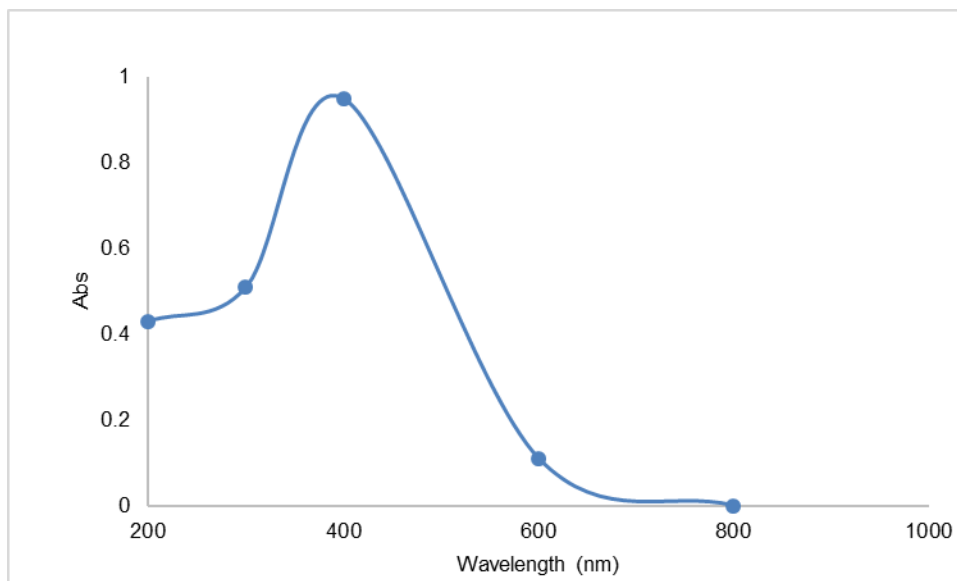


Fig. 1 X-ray diffraction

The JEOL JDX Powder X-Ray diffractometer at 2 with 0 to 100° range available the L-AgNPs XRD pattern. The synthesized L-AgNPs were compared to the typical Ag XRD pattern. CuK radiation with a λ of 1.5406 was used to record the pattern at a voltage of 40 kV, a current of 15 mA, and a scan rate of 10°/min.

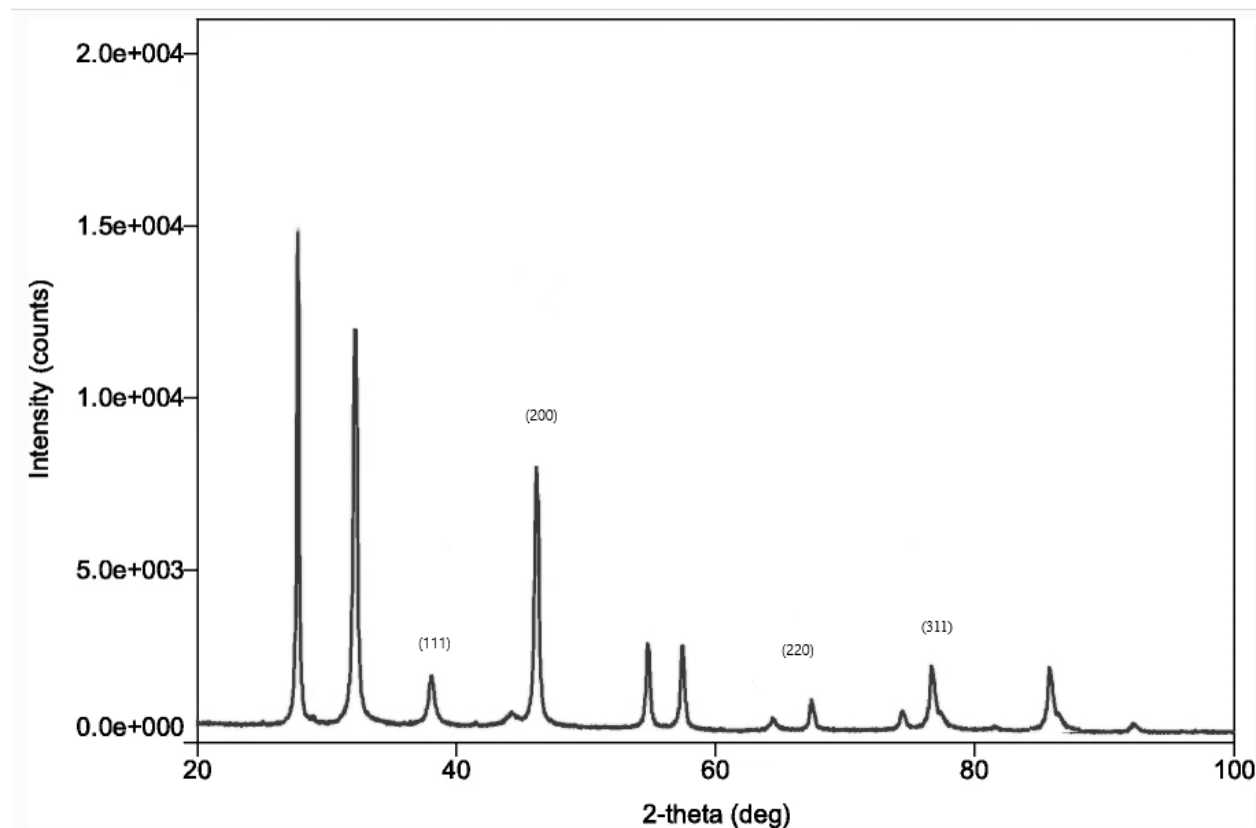


Fig. 2 XRD analysis

XRD ANALYSIS

The XRD pattern of silver nanoparticles which confirmed the crystalline nature of L-AgNPs - Ray diffractometer at 2θ with a range of 0 to 100° . The four distinct diffraction peaks at 2θ values of 32.6° , 43.78° , 65.32° , and 78.11° is index able to the Ag reflection planes (1 1 1), (2 0 0), (2 2 0), and (3 1 1). Presence of additional peaks was due to presence of organic compounds which are responsible for stabilization of silver nanoparticles.

TEM study

Using a JEOL (JEM2100) HR TEM transmission electron microscope, morphological investigations of silver nanoparticles were examined. AgNPs samples were prepared for TEM imaging by applying 1.0 l of an AgNPs colloidal solution to a 400-mesh copper grid that had been coated with carbon, letting the film sit for letting the grid dry for the following two hours before taking measurements.

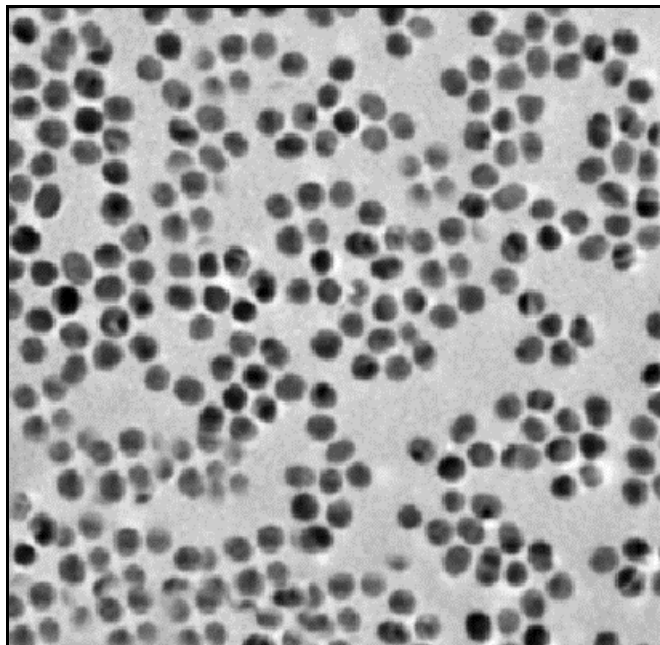


Fig. 3 TEM

TEM Small particles with a diameter ranging from 20 to 50 nm were visible, as shown by photographs.

Microbial culture

For this antibacterial study, the microbes It employed *Escherichia coli* and *Staphylococcus aureus*. The bacterial strains were cultivated in nutrient broth at 37°C and then stored at 4°C before being cultured in the nutrient agar medium.

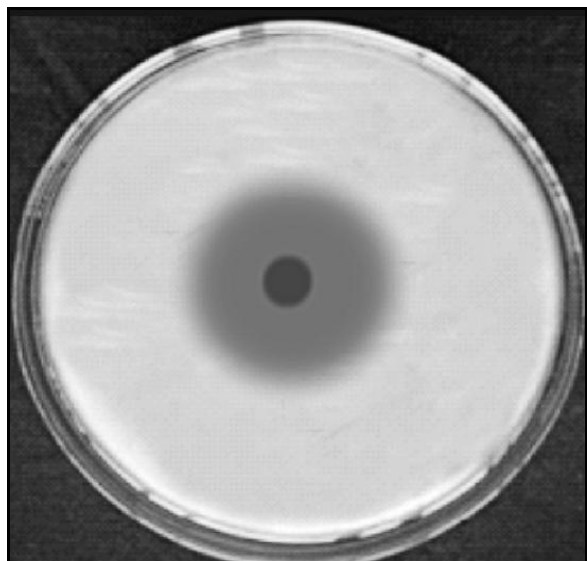
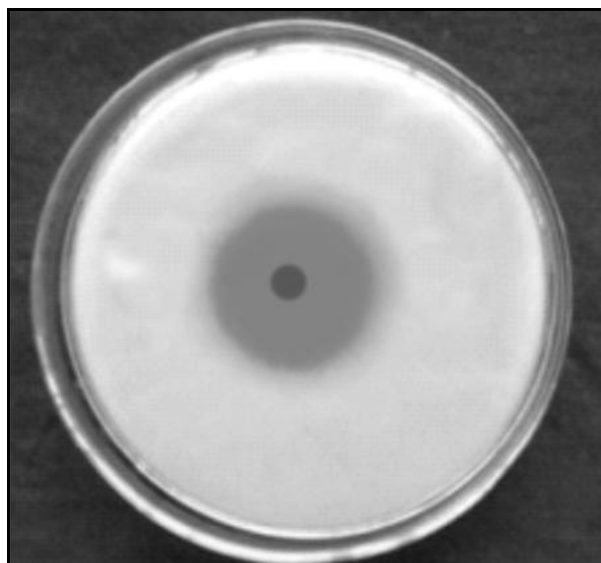
Antibacterial Activity:

Antibacterial activity of L-AgNPs synthesised were determined using a agar well-diffusion method. About On glass petri dishes, 15-20 ml of nutritional agar was placed, and to settle for few minutes. The bacterial strain was cultured using pour plate method and well was made in the agar plate. Different dilution of (20, 40, 60, 80, and 100 µl) test nanoparticle-filled liquid was added between these holes. 48 hours were spent incubating the plates at 37°C. The zone of inhibition was then calculated.

Statistic evaluation

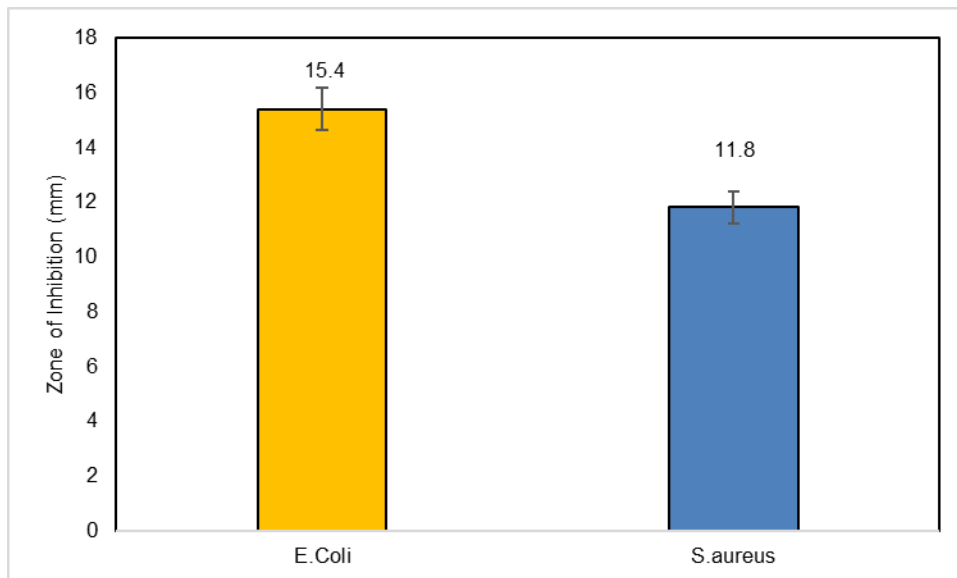
For multiple comparisons in the data's statistical analysis, one-way analysis of variance (ANOVA) was utilised, and the LSD test was used for post hoc analysis. The threshold for statistical significance was P 0.05. The analysis of the data was done with SPSS (22.0).

Results of agar diffusion assay



Escherichia coli

Staphylococcus aureus



Results shown as Mean SD (n=3).

Results and Discussion

This study demonstrated that *Staphylococcus aureus* was not the only bacteria that lemon silver nanoparticles could effectively combat. Ling Gao *et al.*, 2023 discussed about the Gold nanoparticles (AuNPs) have been created using both plant and ultrasound extracts, which are both environmentally friendly methods. It is still unknown, though, how the addition of citrus peel extract (CPE) and ultrasound would influence the bioactivity and structural properties of AuNPs. In this study, The particle size, stability, yield, effectiveness of phenolic encapsulation, and anti-inflammatory activity of AuNPs were all examined in relation to the ultrasonic environment. The findings demonstrated a positive correlation between temperature and the particle size and anti-inflammatory potency of synthetic AuNPs. increased when the power intensity rose, the reaction mixture's total phenolic content (TPC) increased while the particle size was significantly reduced. An increase in TPC was the cause of AuNPs' improved anti-inflammatory activity. The AuNPs generated with or without ultrasonic treatment were characterised by us. In his works, Haroon Mahmood *et al.* (2022) discussed the creation of nanoparticles and their potential applications. Using citrus limon fruit extract and an AuCl₃ solution, gold nanoparticles were effectively created. The results of phytochemical analysis and functional group analysis were similarly positive. The gold nanoparticles were located using UV spectroscopy and FT-IR analysis, and they were verified by XRD studies, SEM, and EDX

analysis. The gold nanoparticles were measured to be 30.6 nm in size. The antibacterial activity of synthesised gold nanoparticles against *Klebsiella pneumonia* and *Listeria monocytogenes* shown significant antimicrobial activity against these two pathogens. In this study, the potency of AuNPs produced through green synthesis was established. Using an aqueous citrus limon zest extract, Yasmina Khane et al. (2013) reviewed and addressed the green synthesis of (AgNPs), optimising the many experimental parameters required for production and stability of AgNPs. By using UV-Visible analysis to find the 535.5 nm band for surface plasmon resonance, the formation of Evidence a collection of nanoparticles. This claim was supported by the observation that the colour of the colloidal suspension of silver nitrate changed from yellow to a reddish-brown hue after the plant extract was added. 1 mM silver nitrate, a mixed extract at a ratio of 1:9, and an incubation time of 4 hours were found to be the ideal conditions. The phytochemical components found in Citrus limon zest extract were found, according to the Fourier transform infrared spectroscopy spectrum. Ghalia Batool Alvi *et al.* Explained that the nanotechnology is concerned with creating objects and materials at the 1-100 nanometer scale. In terms of simplicity and dependability, biological production of nanoparticles utilizing microbes and plants is the most effective way. Selenium metal salt from citrus fruit extracts was used to synthesise metallic nanoparticles, and their characterization and assessment for antibacterial properties against pathogenic microbes were key goals of this study. Citrus fruit extracts from Lemon and Grapefruit were used as precursors in the approach of the simple green method, which was used to create nanoparticles. The solution's bright red hue suggested that selenium nanoparticles (SeNPs) were being synthesised. UV-Vis Spectrophotometry was initially employed to describe the nanoparticles, and then FTIR analyses and DLS graphs were acquired using Zetasizer.

CONCLUSION

A strategy that has the potential to produce nanoparticles with potential antibacterial action lemon scent is used in the environmentally friendly synthesis of silver nanoparticles. As reducing and capping agents, plant extracts provide a secure, economical, and environmentally friendly alternative to traditional techniques. The potential for usage as antimicrobial agents is suggested by the synthesized AgNPs' notable antibacterial activity against both gram-positive and gram-negative bacteria. The mechanisms underpinning AgNPs' antibacterial effectiveness as well as any possible harm to human cells need to be further

understood, according to more research. In order to produce AgNPs on a wide scale, the green synthesis method's scalability must also be evaluated. Despite this, this study offers useful information on the production of nanoparticles with potential applications in biomedicine utilising plant extracts. In our research study we used lemon extract to silver nanoparticle synthesis and evaluation of pathogenic bacterium antibacterial efficacy. Based on this research showed that ecofriendly green synthesized particles showed pharma activity in nanomedicine.

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