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Phyto mediated Biogenic Synthesis of Silver Nano Particles using *Syzygium cumini* bark extracts and its bio efficacy on antimicrobial and hepatoprotective activity

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ABSTRACT

The biosynthesis of silver nanoparticles, or AgNPs, has received increased attention recently because to their substantial potential in drug administration, catalysis, imaging, nano-device manufacturing, and medicine. We suggest creating silver nanoparticles by synthesizing plant extract of *Syzygium cumini* and assessing their antibacterial and chemocatalytic properties. Silver nitrate in aqueous solution is used to create AgNPs. The produced silver nanoparticles (AgNPs) were examined using Fourier transform infrared (FTIR), X-ray diffraction (XRD), transmission electron microscopy (TEM), UV-Vis spectroscopy, and EDX analysis. The UV-Vis absorption spectra of the generated AgNPs showed a large absorption peak between 375 and 390 nm. TEM analysis of AgNPs showed that they had a hexagonal matrix shape and an average particle size of about 30 nm. AgNPs. An XRD examination reveals the crystalline structure of AgNPs. EDX analysis verified the elemental silver's presence. Protein amide groups are significant reducing agents and are essential for the bio-reduction of Ag⁺ ions to Ag⁰, according to FTIR studies. In this work, the antibacterial and hepatoprotective properties of extracts from the medicinal plant *Syzygium cumini* are studied in relation to manufactured AgNPs. The biosynthesized AgNPs show strong antibacterial and hepatoprotective efficacy.

Keywords: AgNPs, *Syzygium cumini*, particle size, reducing agents, antibacterial, hepatoprotective.

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1. Introduction

1.1. Nanotechnology

Nanotechnology is currently one of the most active fields of materials science study. The next industrial revolution has been named nanotechnology, based on the development of nanomaterials¹. The compound term nano is derived from the Greek word "nanos," which meaning "dwarf." In mathematics, a nanometer is commonly defined as one billionth of a quantity or phrase². One way to describe this is 1×10^{-9} , or simply 10^{-9} . It is the science of little things, encompassing almost all academic disciplines that are involved in comprehending the cosmos and functioning with materials at the atomic and molecular levels³. In his famous address at the California Institute of Technology on December 29, 1959, Nobel winner Richard Feynman first presented the idea of nanotechnology. Richard Feynman discussed the idea of nanoparticles in his 1960 book "There is plenty of room at the bottom." He pointed out that if all that was needed to hold a bit of information was just 100 atoms, then every book ever written could fit inside a cube with sides measuring just 0.02 inches. A Japanese researcher at the University of Tokyo named Norio Taniguchi coined the word "nanotechnology" in 1970. Taniguchi was primarily involved in the development of materials at the nanoscale level. Another technologist, K. Eric Drexler, highlighted the importance of nanotechnology in 1980. Solid colloidal particles with a size range of 10 to 1000 nm (1.0 μm) are known as nanoparticles. These include of physiologically active substances or active medications that have been dissolved, imprisoned, or on which the active ingredient has been adsorbed or bound. Nanotechnology and medicine share the same objective of using targeted and controlled drug delivery to treat patients as effectively and without side effects as possible and to diagnose as early and accurately as possible.⁴ Nanoparticles are solid colloidal particles that range in size from 10 to 1000 nm (1.0 μm). Physiologically active compounds or active pharmaceuticals that have been dissolved, trapped, or on which the active component has been bonded or adsorbed are among them. Targeted and regulated drug delivery is used in both medicine and nanotechnology to treat patients as effectively and without side effects as possible and to identify them as early and correctly as possible. The characteristics of nanoscale particles are highly significant since they differ greatly from those of bulk scale particles. Large molecules, micelles, colloids, aggregation molecules, and polymer molecules are all related to the size range in chemistry. The most common links between electrical engineering, physics, and nanoscience are related to quantum and electron behaviour in small structures. As biological molecules like DNA, RNA, and subcellular organelles can be thought of as nanostructures, biology and biochemistry have also been closely linked to nanoscience⁵. The use of nanoscale structures in medication administration, gene sequencing, and diagnostics is creating the

nanotechnology-biomedical engineering interface. The Royal Society and Royal Academy of Engineering define "nanotechnology" as "the design, characterisation, production, and application of structures, devices, and systems by controlling shape and size at nanometre scale," or "the study of phenomena and manipulation⁶ of materials at atomic, molecular, and macromolecular scales, where properties differ significantly from those at a larger scale." Through meticulous atomic and molecular manipulation of matter, distinct particles can be created. By carefully modifying matter at the atomic and molecular level to create particles with unique physico-chemical properties that enhance conductivity, strength, durability, reactivity, or other properties of products and applications, nanotechnology⁷ is used to create new materials and products at the nanoscale. Nanotechnology⁸ has the potential to significantly minimize environmental impact and save energy. Currently, there are over 800 consumer products on the market that include nanotechnology in some way⁹. According to estimations, the nanotechnology industry, currently valued in the billions of dollars, is expected to reach \$1 trillion by 2015¹⁰ and \$3 trillion by 2018¹¹. Currently, the globe produces millions of tonnes of nanomaterials each year, and in the near future, there will probably see a big rise in output.

1.2. Nanomaterial types

Both inorganic and carbon-based materials, such as fullerenes and nanotubes, can be used to create nanomaterials. Metals (iron, zinc oxide, cerium oxide, gold, and silver), metal oxides (titanium dioxide, iron oxide, zinc oxide, and cerium oxide), and quantum dots (cadmium sulfide and selenium) are some examples of these.¹²

1.2.1. Natural nanoparticles

Naturally occurring nanoparticles (1 to 100 nm) as suspended particles smaller than 25 nm are seen in aquatic surface and ground water^{13, 14, 15}. Viruses, polysaccharides, and bacterial exudates are a few examples of complex nanostructured proteins that are present in biological systems and govern a range of biological activities. In addition to sea spray, volcanoes, fires, and rock weathering¹⁶, other sources of nano-sized components include microbial activity and chemical hydrolysis¹⁷.

1.2.2. Anthropogenic nanoparticles

Anthropogenic nanoparticles, sometimes referred to as accidental or adventitious, have existed since the beginning of human civilization. However, because of inorganic nanomaterial emissions from human activity, the amount of naturally occurring nanoparticles in the atmosphere has increased by almost twofold¹⁸. According to a recent definition, nanomaterials include colloids found in soil, which include clays, organic matter, iron oxides, and other minerals crucial for biogeochemical processes; ultrafine particles found in the air, which include nanomaterials from stationary combustion sources and diesel and gasoline-

fueled vehicles; and colloids found in water, which include macromolecules, humic and fulvic acids, proteins and peptides, and hydrous iron and manganese oxide¹⁹.

1.2.3. Carbon based nanomaterials

Applications for carbon-based nanomaterials include biology, electronics, and optics.

1.2.3.1. Fullerenes

Fullerenes are carbon compounds consisting of sixty atoms. The poor solubility of fullerenes and their derivatives in biofluids²⁰ restricts their use in the field of medicine. Nevertheless, they have drawn the interest of numerous scientists because to their many fascinating applications in this field, and early research has examined their effects on apoptosis, DNA photocleavage, neuroprotection, HIV protease inhibition, and other biological aspects²¹.

1.2.3.2. Carbon nanotubes

The cylindrical carbon particles known as carbon nanotubes, or CNTs, have a diameter of one to ten nanometers and a length of several micrometres. They can have many walls or just one wall (SWCNT or MWCNT). They have been employed as microelectrodes in electrochemical processes, supports for heterogeneous catalysis, nanoprobe in atomic force microscopy, electron field emitters, and are presently being investigated as potential hydrogen storage devices²². They have applications outside of the manufacturing sector and are strong yet adaptable (i.e., aircrafts, sports equipment, etc.).

1.2.4. Metal oxide nanoparticles

The natural environment contains significant concentrations of iron oxide nanoparticles²³. In order to facilitate successful transport of the desired gene and release inside the cell, gene therapy, clinical diagnostics, drug delivery, magnetic resonance imaging, and other uses, they have already been employed as contrast agents^{24,25,27,28}. For nanobiotechnological applications, their super magnetic characteristics and relatively low toxicity make them perfect²⁹. Zinc oxide nanoparticles are conventional band gap semiconductors, meaning that they have an energy range in a solid where there are no electron states. These nanoparticles have drawn attention due to their potential applications in sunscreens, chemical sensors, solar cells, water remediation technologies, electronic devices, antimicrobials, and cosmetics³⁰. Their capacity to block UV-A and UV-B radiation while maintaining optical transparency has also drawn research attention^{31,32}. Titanium dioxide has attracted a lot of interest^{33,34,35} for application in photocatalytic activity³⁶ and photocells³⁷ because of its stability and affordability. TiO₂ particles can also be found in other products energy storage devices³⁸, paints and coatings^{39,40} and sunscreens. Three crystalline phases—rutile, brookite and anatase can be produced from TiO₂^{41,42}.

1.2.5. Metal nanoparticles

1.2.5.1. Zero-valent iron nanoparticles

Usually employed in bioremediation processes, zero-valent iron nanoparticles have a size-dependent ability to partially break down and adsorb pollutants^{43, 44}. Their enormous volume to surface area boosts their efficacy and raises the quantity of electron transfer required to eliminate dangerous atoms.

1.2.5.2. Silver nanoparticles

Because of its extraordinary and distinctive qualities, including as conductivity, chemical stability, catalytic activity, nonlinear optical behaviour, and antibacterial activity⁴⁵, silver nanoparticles have been the subject of a significant lot of research. Their characteristics enable their employment in a multitude of applications, such as bacterial disinfectants for use in infusion systems, catheters, etc.⁴⁶ microelectronics⁴⁷, medical textiles⁴⁸, and inks. Since consumers value antibacterial qualities, adding silver nanoparticles to products like plastics, apparel, lotions, and soaps boosts their commercial value. The cost of producing silver nanoparticles is likewise reasonable. Currently, more consumer goods include silver than any other nanomaterial⁴⁹. Because of their ability to be implanted in a variety of textiles and their antibacterial qualities, silver nanoparticles have multiple applications in hospitals. These include medical bandages, burn treatment dressings, and surgical dressings⁵⁰. Silver nanoparticles are being used in an increasing number of domestic appliances, such as refrigerators, washing machines, and food containers, to prevent surface mold growth⁵¹. It has been demonstrated that washing socks containing embedded silver nanoparticles reduces the quantity of silver released into the atmosphere^{52, 53}. In a similar vein, water filtering anti-fouling membranes may leak silver⁵⁴. Due to their optical responses to environmental pollutants, such as the potential for silver nanoparticles to be utilized as herbicide detectors, gold and silver have demonstrated promise as environmental sensors⁵⁵. Zero-valent iron can be utilized to build subterranean reactive barriers, which can be used to treat contaminated groundwater⁵⁶. Ohio State University researchers are utilizing polymer semiconductors, which are solar energy absorbers with the capacity to produce electricity. They have found that the plastic's ability to generate electrical current is increased when small amounts of silver are added⁵⁷.

1.2.5.2.1 Importance of silver nanoparticles

- 1) Because of their special chemical and physical characteristics, silver nanoparticles are used in military, medicinal, and cosmetic applications. The characteristics of nanoscale particles are often marginally different from those of their bulk counterparts⁵⁸.
- 2) It is used in medication delivery systems, biosensing, imaging, and air filtration and quality control.

3) There are many applications for organically produced silver nanoparticles, including antimicrobials, optical receptors, chemical reaction catalysts, solar energy absorption coatings, and biolabeling. They are also utilized in electrical batteries as an intercalation material⁵⁹.

4) Silver nanoparticles are helpful in several areas, such as microelectronics, antimicrobials and therapies, high sensitivity biomolecular detection, and catalysis, despite their cytotoxicity.

5) A number of well-known producers of consumer goods are already creating household products that take advantage of silver nanoparticles' antibacterial qualities. These goods include air conditioners, washing machines, and refrigerators with nano silver inside⁶⁰.

6) Because silver nanoparticles are antibacterial, a number of new items have been developed, including antibacterial dryers, toothpaste, and soap, sheets that lessen the smell of textiles (especially socks), and apparel that has been infused with nanoparticles.⁶¹

The metabolism and detoxification of toxins that enter the body and have the potential to damage the liver and cause catastrophic diseases are two of the liver's many essential tasks. As a result, important toxicological problems associated with some illnesses have centered on how they

2. Materials and Methods

2.1 Collection, authentication and processing of plant materials

The plant materials will be either collected from the field or purchased from the market. The herbarium will be prepared and preserved for the herbs collected from the field and the material will be authenticated by a taxonomist. For the samples procured from the market, authentication will be done by performing morphological and microscopical studies and comparing the observations with those reported in the literature. The plant material will be dried at a temperature not exceeding 50°C and then coarsely powdered⁶². The material will be stored in an airtight container protected from light at room temperature.

2.2 Preparation of extracts of plant materials

The different extracts of the drug will be prepared by different extraction techniques such as soxhlet extraction, hot maceration etc. The extracts will be separated from the marc by filtration, concentrated to dryness and stored at -20°C until further use⁶³.

2.3 Synthesis of silver nanoparticles by using plant extract⁶⁴

Plant extract (0.1 g) will be vigorously stirred for 4 hours after being introduced to 20 mL of distillation deionized water. Following that, forty milliliters of AgNO₃ (1 × 10⁻³ M) will be added and stirred for 24 hours at room temperature (25°C). Ag-NPs will be acquired gradually throughout the course of the incubation time. Throughout the reduction process, the

solution will be kept at room temperature and in the dark to avoid any photochemical reactions. Before being used, nitrogen gas is used to cleanse the solution component. After that, reduction took place with nitrogen present in order to remove oxygen⁶⁵. Following a 20-minute centrifugation at 15,000 rpm, the resulting colloidal suspensions of Ag/plant extract will be rinsed four times to eliminate any remaining silver ion residue. The Ag/plant extract⁶⁶ will then be obtained by vacuum-drying the precipitate nanoparticles for an entire night at 30°C.

2.4 Characterisation of plant extract mediated silver nanoparticles⁶⁷

2.4.1 UV-Vis Spectroscopy

In this study, the absorption of nanoparticles in the UV-visible spectral region will be determined using ultraviolet-visible spectrophotometer (UV-Vis -Model Lab India, UV3200). In other words, it utilizes light ranging from visible to near-UV and near-infrared (NIR) spectrum. Absorption in the visible range directly affects the compounds' perceived color. Molecules change electrically in this area of the electromagnetic spectrum.

2.4.2 Scanning Electron Microscope (SEM) Analysis

Scanning Electron microscope (SEM) Scanning Electron Microscope (SEM) analysis will be done using SEM machine. The SEM equipment will be used for analysis utilizing a scanning electron microscope (SEM). To make thin films, a small portion of the sample will be deposited onto a copper grid covered in carbon. After using blotting paper to remove any remaining solution, the film on the SEM grid will be exposed to a mercury light for five minutes to dry⁶⁸. The SEM analysis was done using HITACHI, S3700 N Model.

2.4.3 Fourier Transmission Infra-Red Spectroscopy (FTIR)

FTIR is a chemical analysis technique that calculates the infrared intensity of light in relation to its wavelength or wave number. It will be applied to the analysis of the biomolecule and the bonding interactions among the silver nanoparticle molecules. IR spectroscopy is used to determine the vibrational characteristics of the chemical functional groups in the sample. When infrared light interacts with matter, chemical bonds will display bending, stretching, and contracting behaviors. This chemical functional group has a tendency to absorb infrared light within a particular wave number range of the remaining molecule's structure⁶⁹. FTIR analysis was performed using 2-3 drops of colloidal solution synthesized silver nanoparticles; it was mixed with KBr powder for moisture absorption in a clean mortar and pestle. This method was also used for the plant extract. FTIR analysis was carried out using Shimadzu, 8400 S spectrum model.

2.4.4 X-Ray Diffraction (XRD) Measurements

The phase formation of bio-reduced silver nanoparticles will be investigated using XRD. The diffraction data of fully dried nanoparticle thin films on glass slides will be captured with an

X-ray diffractometer⁷⁰. XRD analysis was performed using colloidal solution of synthesized AgNPs. The diffraction pattern of XRD (XRD – Bruker D8 model) was recorded on the angle range of 20° to 80°. XRD method was used for the determination of crystalline structures and the positions as well as intensities of diffraction pattern.

2.4.5 Transmission electron microscopy (TEM)

Transmission electron microscopy (TEM) was used to confirm the morphology of the produced AgNPs. The sample was prepared using *Syzygium cumini* extract and AgNPs (Phytosynthesized). The TEM analysis was performed using (JOEL, JEM-F200 model) a drop of synthesized silver nanoparticles colloidal solution, it was exposed to infrared light (30 min) up to the sample evaporation.

2.4.6 Zeta potential analysis

Zeta potential was measured with the Malvern Zetasizer instrument for the zeta potential analysis sample was prepared using mixer of AgNPs colloid solution in zeta dip cell. Using this type of analysis the potential stability of the colloidal system and noble properties was confirmed.

2.5 Evaluation of Antibacterial activity

The antibacterial activity of the plant extracts as well as synthesized AgNPs will be assessed using agar diffusion method. By spreading the bacterial inoculums on the media⁷¹, nutrient agar will be inoculated with the specified microorganisms, including *Staphylococcus aureus*, *Escherichia coli*, and *Aspergillus niger*, *Aspergillus flavus*. Using a sterile cork borer, wells will be punched into the agar and then filled with plant extracts (10 mcg/disc) a common reference antibiotic, will be utilized as controls for the examined bacteria. Following a 24-hour incubation period at 37° C, the diameter of the zone of inhibition will be utilized to measure the antibacterial activity of the plates. By comparing the zones of inhibition of the various extracts, the antibacterial capability of each will be assessed.

3. Result and Discussion

3.1. Preparation of Extracts

The Plant material for present analysis was collected from local area of Baripada. *Syzygium cumini* barks are washed for 15-minute rinse under running water, they were allowed to air dry for two days under shade. Using a hand grinder, dry barks were pounded into fine powder⁷². Afterwards, the bark powder was placed in a 500 ml Erlenmeyer flask, 100 ml of double-distilled water was added, and a magnetic stirrer was used to continually stirred the mixture for ten minutes⁷³. First, the extract was run through Whatman filter paper No. 1. (Fig.1) After being collected, the filtrate was kept at 4°C and used in all subsequent experiments⁷⁴.



Figure.1. (a) Powdered bark of *Syzium cumini* (b) powder bark material and distilled water mixed with the help of magnetic stirrer (c) plant extract

3.2. Synthesis of Silver nanoparticles

10 mL of the bark extract was combined with 50 mL of aqueous silver nitrate (1 mM) while being constantly stirred. A visible shift in colour from colourless to yellowish-brown(**Fig.2**) indicates the formation of AgNPs⁷⁵.

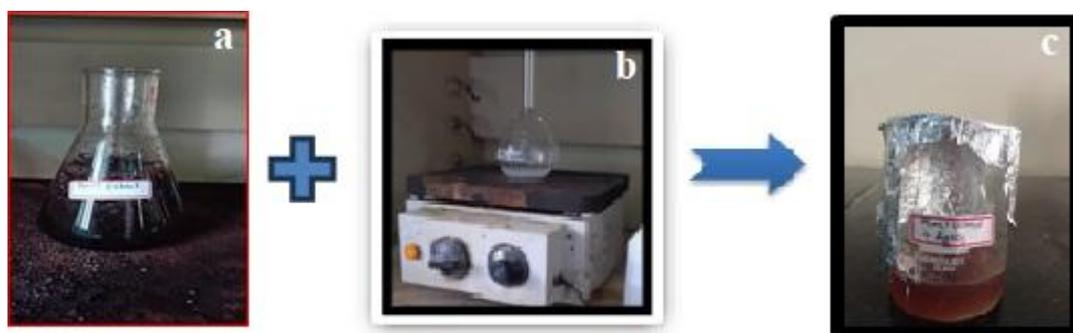


Figure.2. (a) Plant extracts (b) 1mM AgNO₃ (c) silver nanoparticle formation

3.3 UV-Visible spectroscopy

To track the optical properties and electrical structure of the generated nanoparticles and learn more about how AgNPs are made, UV-visible spectroscopy is a helpful tool. A nanoparticle's electron cloud may vibrate on its surface when it absorbs electromagnetic waves at a particular frequency⁷⁶. Surface plasmon resonance, or SPR, is the name given to this phenomenon, which the UV-Vis spectrophotometer records as electromagnetic wavelengths. Fig. 3 shows the optimization of incubation duration during the synthesis of AgNPs. UV wavelengths of SGC-AgNP-B (AgNPs from Bark extract) were measured from 15 minutes to 24 hours of incubation based on the spectra. As the incubation period increased, the peaks became more intense. The wavelength absorbance increases in intensity due to the increasing amount of nanoparticles created by the reduction of silver ions and biomolecules in the aqueous plant extract solution. With extract concentrations of 2%, 4%, 6%, 8%, and 10% (v/v), the analysis for extract concentration was carried out for 24 hours at 25°C. The produced SGC-AgNP-B peaks were detected at wavelengths ranging from 375 nm

to 390 nm. The peak's sharpness suggests that the synthesized AgNPs have formed a spherical shape and are distributed uniformly⁷⁷.

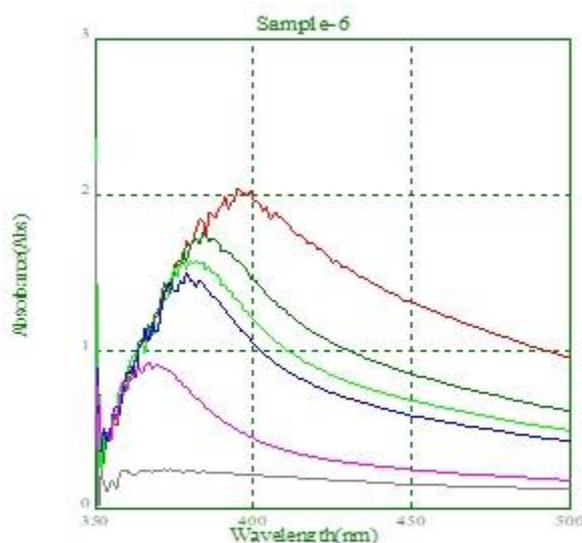


Figure.3. UV-Visible spectra of silver nanoparticles of aqueous bark extract of *Syzygium cumini*

3.4. FT-IR Spectroscopy SGC-Plant extract and AgNPs

FT-IR spectrum investigation verified the SGC interaction on the SGC-AgNPs⁷⁸ surface. The IR spectrum of SGC showed intense bands at 3329, 2130, 1636, 620 cm^{-1} (Fig. 4) and that of SGC-AgNPs showed intense bands at 3325, 2139, 1636, 592 and 577 cm^{-1} (Fig.4). The significant difference observed between the of FT-IR bands of SGC and SGC -AgNPs was due to the reduction process. The stretching vibration mode of -OH groups in SGC was identified by a strong and intense band centered at 3325 cm^{-1} (Fig. 4a & 4b), while the stretching vibration of C-H groups was identified by another band at 2139 cm^{-1} .

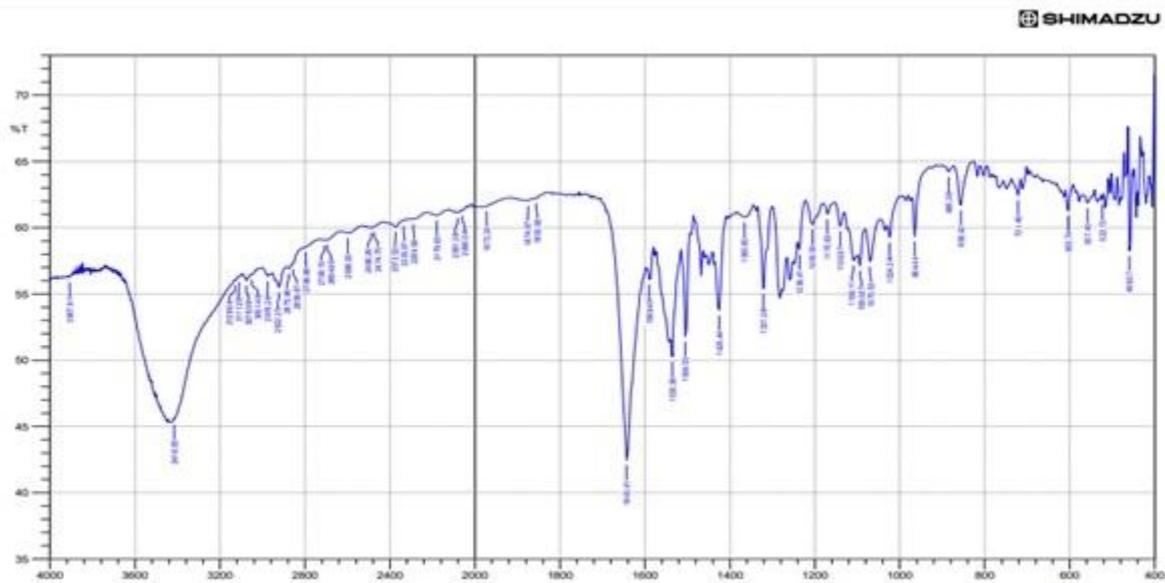


Figure 4a. FT-IR Spectroscopy SGC plant Extract

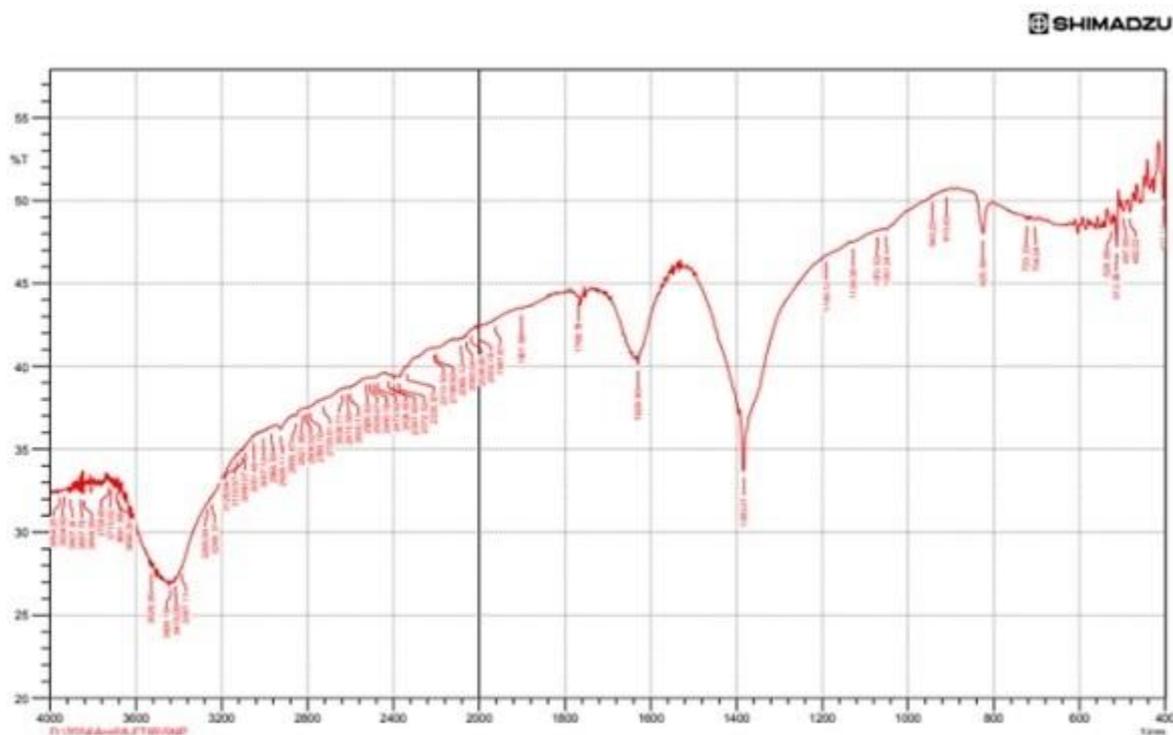


Figure 4b. FT-IR Spectroscopy SGC- AgNPs

3.5. X-Ray Diffraction (XRD)

Various Bragg reflections clearly showed the presence of (13), (18.9), (20.1), and (24.9) sets of lattice planes that may be indexed to a face-centered-cubic (fcc) structure for silver in the XRD pattern of the SGC-AgNPs, as shown in Fig. 5. Therefore, it is evident from the XRD pattern that the SGC-AgNPs that were produced were primarily crystalline in character. Apart from the Bragg peaks that are indicative of fcc AgNPs, there were also unassigned peaks that were found, which indicated the formation of a bioorganic phase crystal on the surface of SGC-AgNP.

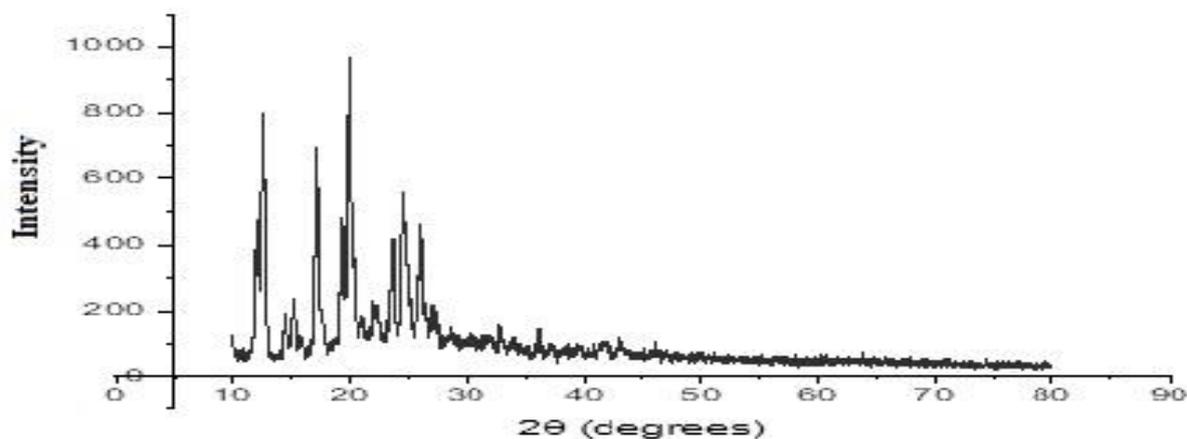


Fig. Figure 5. X-ray diffraction of AgNPs in *S. cumini*

3.6. SEM Analysis SGC-AgNPs⁷⁹

The material becomes colloidal with the electrons inside it that are knocked out of their orbit when it is exposed to an electron beam inside a scanning electron microscope (SEM), according to SEM analysis. SEM morphology obtained from the present analysis Nano crystals was identified (**Fig. 6**).

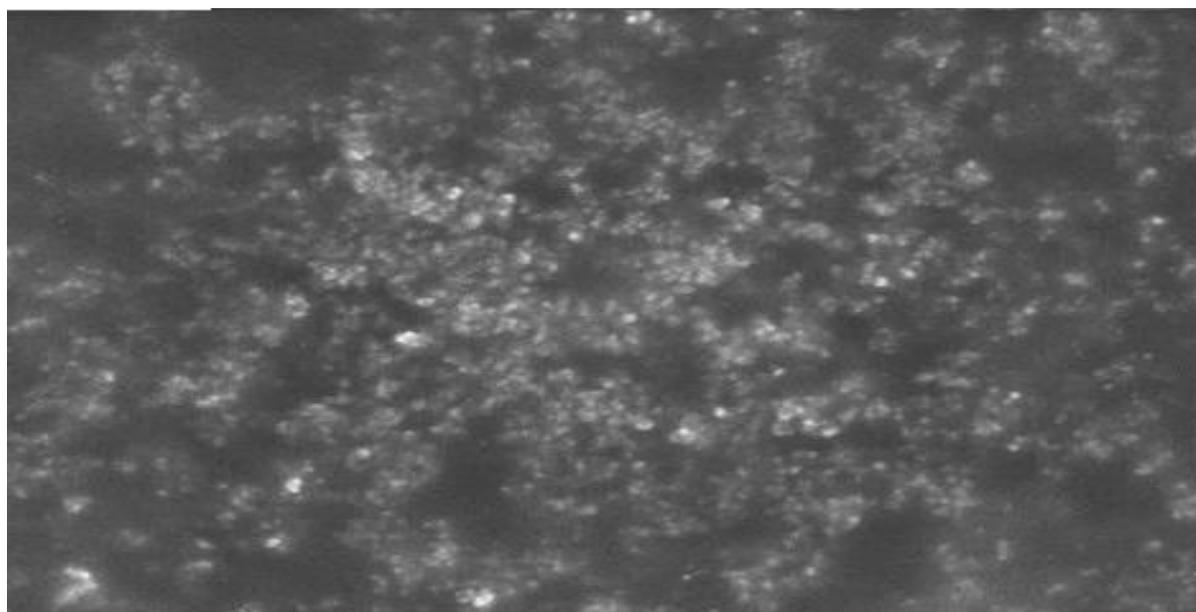


Figure 6. SEM Analysis SGC-AgNPs

3.7. DLS (Particle size) and zeta potential of SGC-AgNPs

The data of Dynamic light scattering studies⁷⁹ indicated uniformity in the distribution of particles and the average size of the DLS-AgNPs (**Fig. 7a**) to be 30 nm. The PDI is 0.507 The stability of the SGC-AgNPs was inferred from zeta potentiometer measurements. A zeta

potential value of -18.1 mV (**Fig. 7b**) essentially indicated avoiding of aggregation of nanoparticles in suspension.

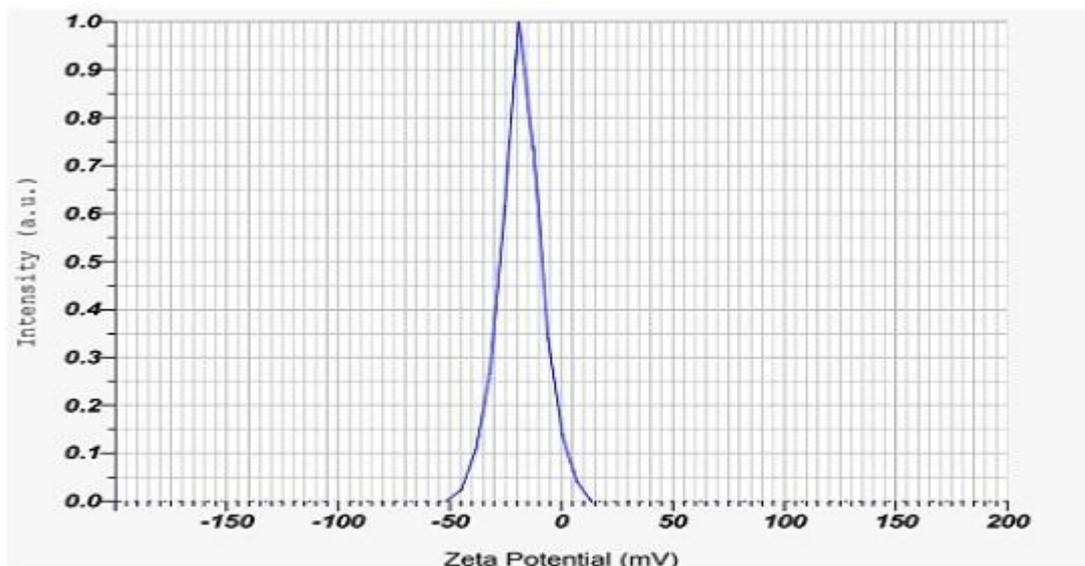


Figure 7a. Zeta Potential of SGC-AgNPs

Z-Average
PI

: 30.0 nm
: 0.507

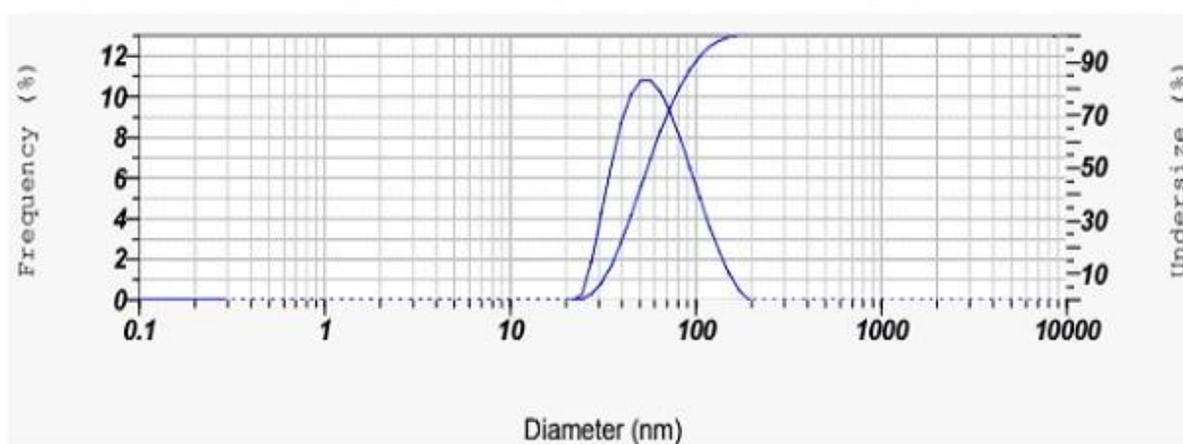


Figure 7b. Particle size of SGC-AgNPs

3.8. TEM images of SGC-AgNPs

Using TEM analysis the shape, size and morphology was confirmed. The TEM investigation verifies that the produced AgNPs were spherical and ranged in size from 20 to 200 nm. The TEM images⁸⁰ of SGC-AgNPs are shown in **Fig.9**. From the 100 nm magnification of nanoparticles the morphology of SGC-AgNPs was found to be spherical in shape. The DLS-AgNPs size distribution histogram showed a broad particle size distribution with nanoparticle sizes of 30 nm (Fig. 8).

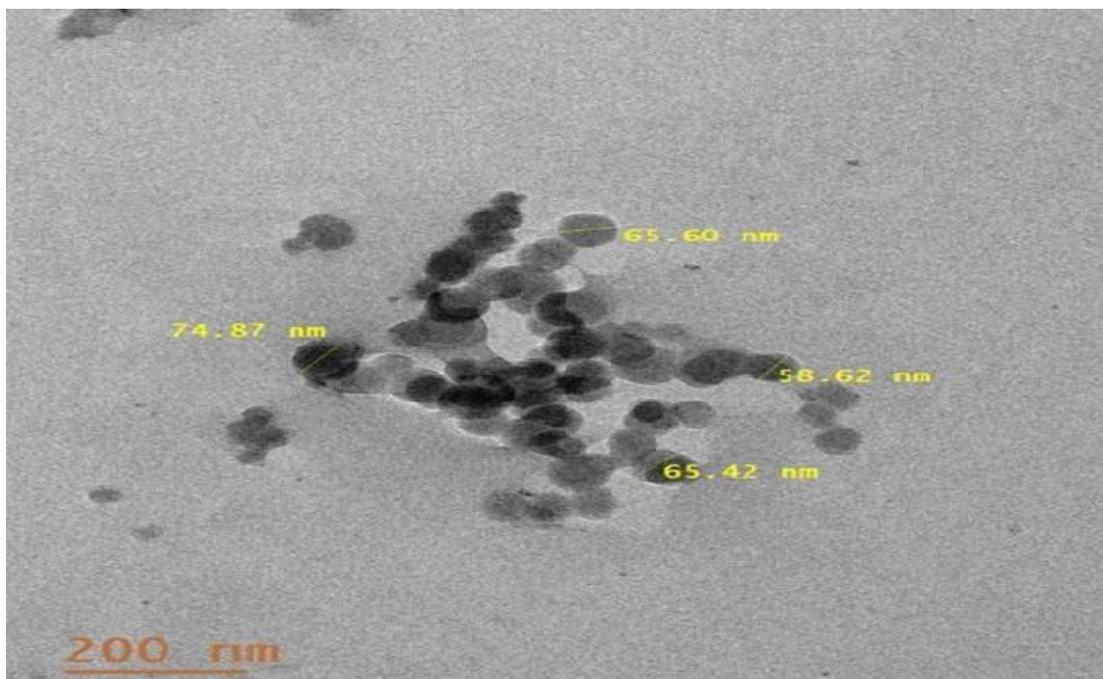


Figure 8. TEM Image of SGC-AgNPs

4.0 Antimicrobial activity of SGC-AgNPs

Table 1 and Figure 9 report the findings of an Agar well diffusion method test of antibacterial activity⁸¹ against four selected pathogens *S. aureus*, *E. coli*, *A. niger*, and *A. flavus*. The results showed that the zone of inhibition of SGC-AgNPs was often higher than that of SGC and standard. Maximum zone of inhibition of 22 ± 0.9 mm was observed against *S. aureus* at $100 \mu\text{g/mL}$ concentration of SGC-AgNPs, while standard Ampicillin showed only 8.1 ± 0.1 mm (Table 1 & Fig. 9). The zones of inhibitions of SGC-AgNPs against other microorganisms are *E. coli* (16 ± 0.6 mm), the fungal strain showed zone of inhibition *A. niger* (14 ± 0.3 mm), and *A. flavus* (13 ± 0.4 mm) while standard Ketoconazole showed only 7.9 ± 0.9 mm (Table 1). The zone of inhibition was nearly two fold in the case of tested microorganism for SGC-AgNPs when compared to standard gentamicin at the same concentration. Although the precise mechanism underlying SGC-AgNPs' bactericidal impact is unknown, it has been hypothesized that the particles may adhere to the pathogen's cell membrane and interfere with vital processes like respiration and permeability. The surface area available for interaction determines how well SGC-AgNPs bind to bacteria; smaller particles with greater surface area available for interaction will have greater antibacterial and antioxidant effects than bigger particles^{82,83}. The manufactured nanoparticles' comparatively smaller size allowed them to adhere to the bacterium's cell wall more easily, creating disruption that ultimately resulted in cell death. Furthermore, it has previously been documented that the size of the particles may have an impact on the antibacterial properties of silver nanoparticles.

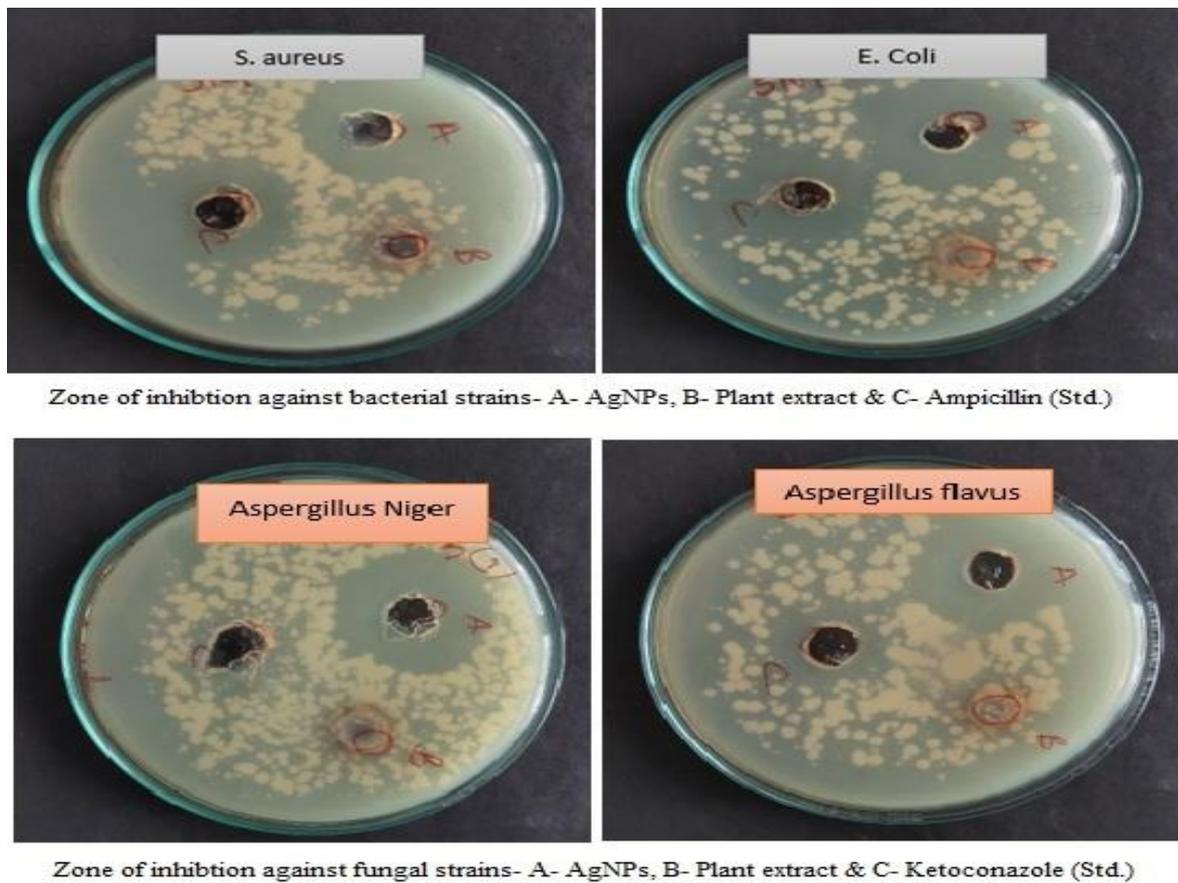
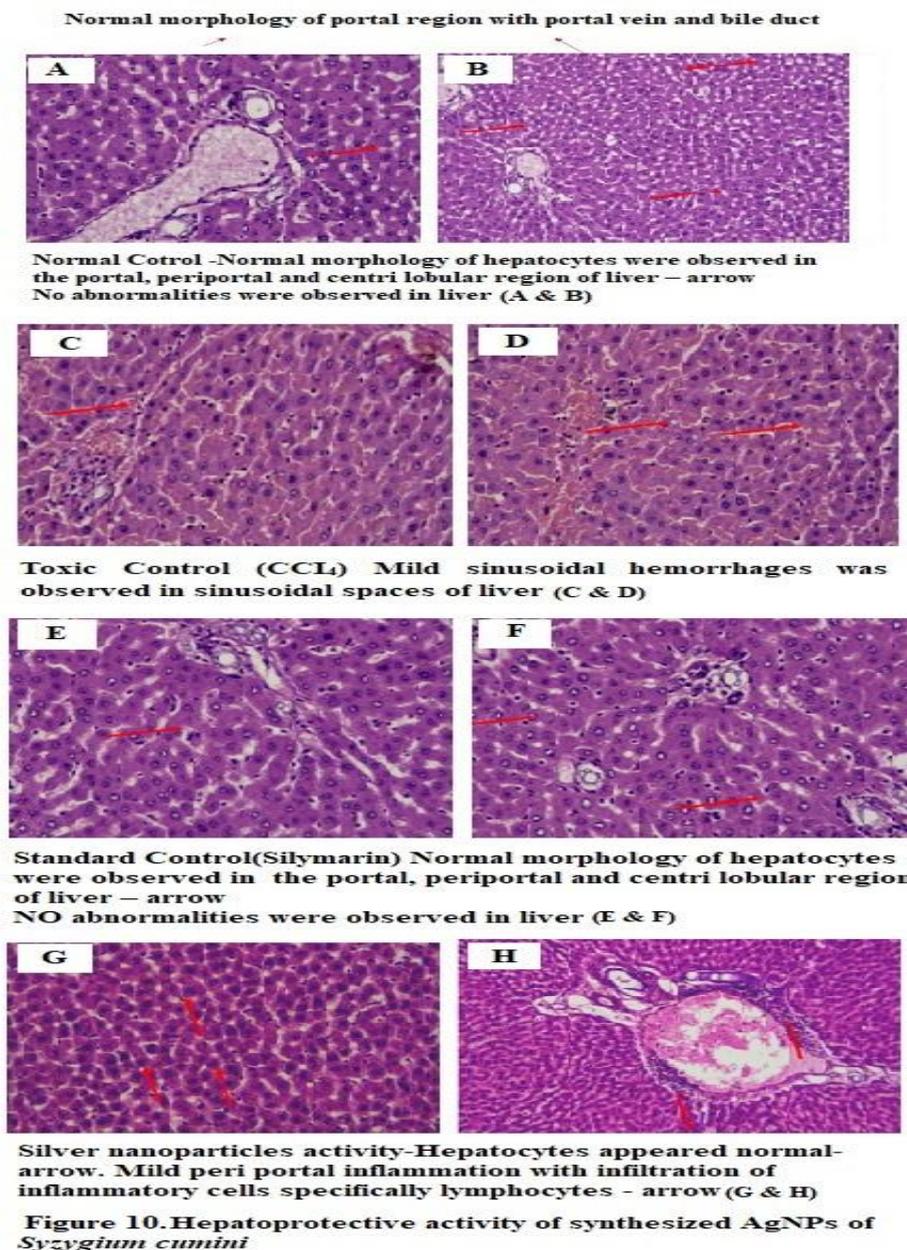


Figure 9. Antimicrobial activity of SGC-AgNPs

5. Hepatoprotective activity

Normal control animals' livers' histopathological sections (**Figs. 10.A & B**) showed a typical hepatic architecture with discrete hepatocytes and sinusoidal spaces. The hepatic architecture was disturbed in the CCl₄-intoxicated group, suggesting an anomaly inside the hepatic cells. (Fig. 10C & D). Mild sinusoidal hemorrhages were observed in sinusoidal spaces of liver. In the liver's portal, periportal, and centrilobular regions, hepatocytes had normal morphology (arrow). After receiving regular treatment, the liver showed no abnormalities at all (Fig. 10E & F). Hepatocytes are appeared normal and normal hepatic architecture was restored after treatment with extract of silver nanoparticles (Fig. 10G & H).



CONCLUSION

Using *S. cumini* aqueous bark extracts as a unique reducing and stabilizing agent of silver salts, the present work offers a straightforward, innovative, and effective synthesis of silver nanoparticles. Ag-NPs that were biosynthesized showed surface plasmon resonance, as demonstrated by UV-Vis spectral analysis. The produced Ag-NPs' size is less than 30 nm, according to the DLS and TEM image investigations. For biosynthesized Ag-NPs, the zeta potential value was (-18.1 mV), showing the nanoparticles' stability. Based on FT-IR spectra, it was discovered that the heterocyclic water-soluble chemicals detected in the *S. cumini* aqueous extracts were the biomolecules that capped and stabilized Ag-NPs. The zone of inhibition was nearly two-fold in the case of tested microorganism for SGC-AgNPs when compared to standard gentamicin at the same concentration. Present studies will help us to access the antibacterial and hepatoprotective activities of the synthesized Ag-NPs. Results

obtained from present analysis it is concluded that this method is quick cheap and economic. *S. Cumini* extracts has great medicinal value which were further focused on the biomedical and pharmaceutical applications of the synthesized Ag-NPs. The biomedical and pharmaceutical sectors will benefit from the easy promotion of medication delivery by the synthesis of phyto-medicated Ag-NPs from medicinal plants utilizing a biological technique at a marketable level. Researchers studying nanobiotechnology will gain more understanding from these kinds of investigations.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The study was approved by the Ethical Review Board of Jeeva life Sciences with approval no. CPCSEA/IAEC/JLC/20/11/23/067

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AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

CONFLICT OF INTERESTS: Declared none.

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