

# Microwave-assisted rapid biosynthesis of silver nanoparticles using *Simarouba glauca* leaf extract and its antimicrobial activity

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## ABSTRACT

We report a straightforward, single-step microwave-enhanced biosynthesis of silver nanoparticles (AgNPs) via an aqueous leaf extract of *Simarouba glauca*. This study represents the first use of *S. glauca* leaf aqueous extract for AgNPs production. The microwave conditions reached their optimal point through ongoing monitoring of the maximum UV absorbance at 454 nm which shows the surface plasmon resonance of AgNPs. The 454 nm absorption peak reached its highest intensity when AgNPs were made through a 70 °C reaction that lasted 3 min which proved the formation of nanoparticles. The three analytical methods of powder dynamic light scattering (DLS), transmission electron microscopy (TEM) and X-ray diffraction (XRD) were used to verify AgNPs formation. TEM images showed spherical silver particles of 5–15 nm synthesized within 3 min via reduction of Ag<sup>+</sup> (from AgNO<sub>3</sub>) by the leaf extract. The XRD results showed that the produced AgNPs contained crystalline structures which SAED confirmed through selected area electron diffraction. The phytochemical screening analysis showed that the extract comprises phenolic compounds which probably function as reducing agents for Ag<sup>+</sup> ions. The antimicrobial testing revealed that the synthesized AgNPs demonstrated effective antibacterial properties compared toward different bacterial strains (eight) which included both Gram-positive and Gram-negative bacteria.

## 1. Introduction

Nanotechnology is currently one of the most active research domains in materials science, encompassing the design, synthesis, characterization, manufacture, and application of structures and devices with controlled shapes and dimensions at the nanometer scale [1,2]. Bio-nanotechnology represents a subfield which combines physical and chemical methods with biological resources including plants and microbes and algae to generate nanomaterials [3,4]. Bio-nanotechnology has become more popular for metallic nanoparticle fabrication because it offers affordable production and environmentally safe

methods and easy-to-follow procedures and large-scale manufacturing capabilities. [5–10]. Among the all metallic nanoparticles like zinc, copper, titanium, magnesium, gold, silver etc., the silver nanoparticles (AgNPs) are attractive due to the wide range of commercial application like surface-enhanced Raman scattering, electronics, catalysis, sensors [11–16]. The AgNPs also used in everyday consumer's life as food storage containers [17]. In ancient time, Silver-based treatments have been used for infection control throughout history and also AgNPs continue to be widely used modern science because they are imposing antimicrobial properties at small doses due to their extensive surface area. [18].

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Many chemical, physical, and biological routes exist for metal nanoparticle synthesis. The use of chemical methods requires hazardous chemicals which endanger human and harmful for the environment, [19], while physical routes demand high-energy equipment and significant power, making them expensive. Biological sources which include plants and microbes and seaweeds offer an attractive alternative to traditional sources. Multiple studies have reported AgNP synthesis through biological resources yet most of these methods require extended periods of time [20,21]. There are also some literatures available employed microwave irradiation combined with chemical reductants, but reports using biological extracts with microwaves are limited [22]. The process of nanoparticle synthesis through microwave assistance is more favorable than traditional heating methods because it shortens reaction duration and decreases energy consumption and chemical waste production [23,24]. The microwave-assistance green synthesis of silver nanoparticle using plant-based extract as a reducing agent has become a major research field but studies show wide variations in reaction time, nanomaterial characteristics and plant material choices [25–29]. Plant-mediated biosynthesis of AgNPs through conventional heating necessitates extended heating periods between 2 and 24 h as shown by *Cucumis prophetarum* aqueous extract needing 3 h at 80 °C [30] and *Azadirachta indica* leaf extract needing 3 h of heat treatment [20]. The efficacy of microwave-irradiated synthesis methods for AgNP production depends on plant origin since *Parkia speciosa* extract requires 4 min to produce AgNPs [31] but *Spondias pinnata* bark extract needs 90–240 s [25] and *Cannabis sativa* extract needs about 30 min [29]. Existing literature primarily employs well-defined plant structures with proven phytochemical compositions, while targeted research evaluates antimicrobial properties against various bacterial species. There is unexplored potential in plants containing different secondary metabolites that have been demonstrated to have biological impacts to synthesis nanoparticles that have numerous applications. This research effort utilizes *S. glauca* leaf extract as a novel plant substance for the synthesis of silver nanoparticles using microwave-assisted biosynthesis. Antimalarial activity from *S. glauca* has been reported in the literature [32]. The selective extraction of glaucarubinone from the *S. glauca* using a deep eutectic solvent has been reported in our previous work [33], but no reports are available on the utilisation of *S. glauca* leaves extract for the synthesis of AgNPs. These finding led to study for the synthesis of silver nanoparticles using *S. glauca* leaves extract.

Herein we report one-pot microwave assisted biosynthesis of silver nanoparticles using *S. glauca* aqueous leaves extract. Further, the synthesised AgNPs has been checked to prove its efficiency as antimicrobial activity against eight different Gram positive and Gram-negative bacterial strains.

## 2. Materials and methods

### 2.1. Materials

*S. glauca* leaves were collected from Mota Layja, Kutch, Gujarat, India (Latitude 22.8852 and longitude 69.2235). *S. glauca* leaves were washed with running water to get rid of extraneous materials followed by dried it and finally powdered it for further use. AR grade silver nitrate was purchased from M/s S. D. Fine Chemicals, Mumbai. 0.1 M AgNO<sub>3</sub> solution was prepared in distilled water at room temperature.

### 2.2. Preparation of plant extract

To prepare a clear aqueous extract (CAE), 5 g of dried leaf powder was added to 50 mL double-distilled water and heated at 70 °C in a water bath for 2 h. The extract was clarified by filtration through Whatman filter paper or by centrifugation at 10,000 rpm for 5 min. The CAE was stored refrigerated until use. The CAE yield, determined after freeze-drying the aqueous extract, was 18.4 ± 0.2 % relative to the dry leaf powder.

### 2.3. Synthesis of silver nanoparticles

To synthesis AgNPs, 1 mL CAE diluted by 20 mL distilled water followed by addition of 0.2–1 mL of 0.1 M AgNO<sub>3</sub> solution in a separate reaction vessel and kept it in the microwave irradiation (Model No.: Start-S; Terminal T260; Line Voltage 230 V; Magnetron S.N. 131528; Frequency 50 Hz) at 60–80 °C for 1–3 min for the optimisation of reaction parameters. Centrifuge the reaction solutions to obtained AgNPs. After the centrifugation, AgNPs were then washed with distilled water followed dry it in a vacuum dryer to obtain dried AgNPs for further characterization and the filtrate of the AgNPs used for the phytochemical analysis.

### 2.4. Phytochemical analysis

#### 2.4.1. Test for alkaloids

50 mg of CAE was mixed with 2 mL of 0.5 % HCl, warmed, and filtered. To 1 mL of the filtrate, Dragendorff's reagent was added; turbidity or precipitate formation indicated the presence of alkaloids.

#### 2.4.2. Test for flavonoids

1 mL of the dilute ammonia solution was added to the portion of the aqueous filtrate of CAE followed by the addition of concentrated H<sub>2</sub>SO<sub>4</sub>. The appearance of the yellow coloration indicated the presence of flavonoids.

#### 2.4.3. Test for terpenoids

1 mL CAE was mixed with 1 mL chloroform followed by careful addition of 2 mL concentrated H<sub>2</sub>SO<sub>4</sub>. A reddish-brown layer at the interface signified terpenoids.

#### 2.4.4. Test for tannins

100 mg CAE was dissolved in 5 mL distilled water and filtered. Addition of 1 % aqueous FeCl<sub>3</sub> produced intense green, purple, blue, or black coloration if tannins were present.

#### 2.4.5. Test for phenols

To 0.5 mL CAE, 1 mL distilled water and a few drops of 10 % aqueous ferric chloride were added. Blue or green color formation indicated phenols.

#### 2.4.6. Test for coumarins

100 mg CAE was treated with 2 mL 1 N NaOH, heated briefly in boiling water, and examined under UV light. Yellow fluorescence suggested presence of coumarins.

#### 2.4.7. Test for saponins

100 mg CAE was shaken vigorously with 3 mL distilled water; stable froth indicated presence of saponins,

#### 2.4.8. Free reducing sugar (Fehling's test)

1 mL CAE was mixed with 3 mL of an equal mixture of Fehling's solutions I and II and boiled for ~2 min. A brick-red precipitate indicated presence reducing sugars.

#### 2.4.9. Test for carbohydrate (Benedict's test)

The test solution was mixed with a few drops of Benedict's reagent and boiled; the appearance of a reddish-brown precipitate signified carbohydrates.

### 2.5. Characterization

AgNP formation was initially assessed by UV–visible spectroscopy using a VarianCary-500 (Shimadzu UV 3600) spectrophotometer. TEM images were obtained on a HR-TEM (JEOL GEM 2100) at 200 kV. Powder XRD data were collected on an Empyrean powder diffractometer

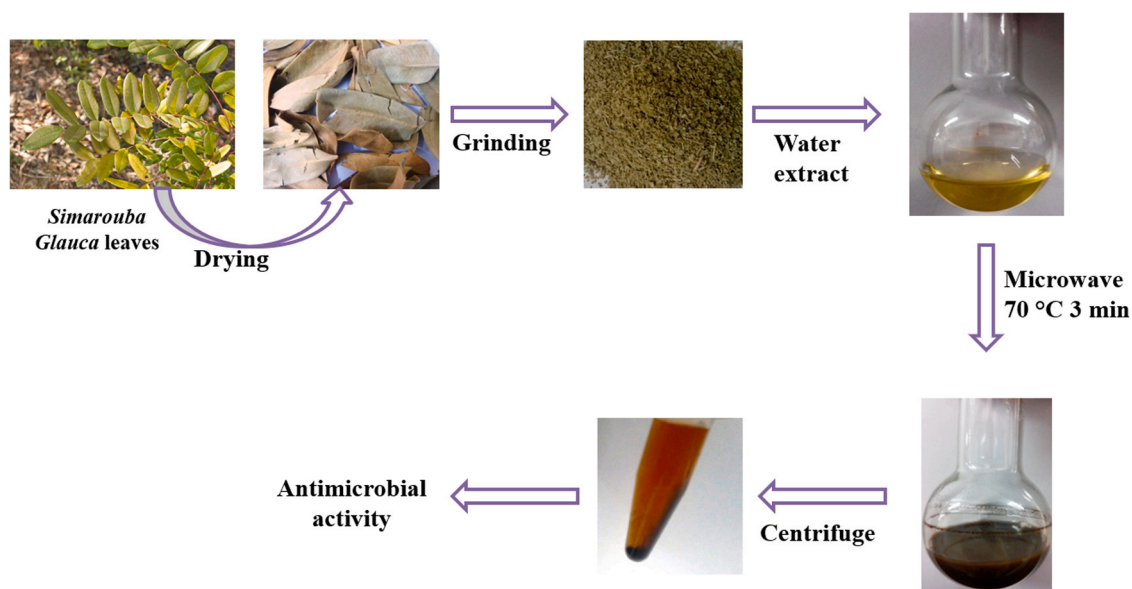


Fig. 1. Synthesis of silver nanoparticles from *S. glauca* leaves.

(Cu-K $\alpha$  radiation) from PAN Analytical. DLS measurements were performed on a Malvern instrument (UK) at a 90° scattering angle using a 488 nm Argon laser GSL 3110.

## 2.6. Bacterial strains

Antibacterial assays were conducted against eight bacterial strains:

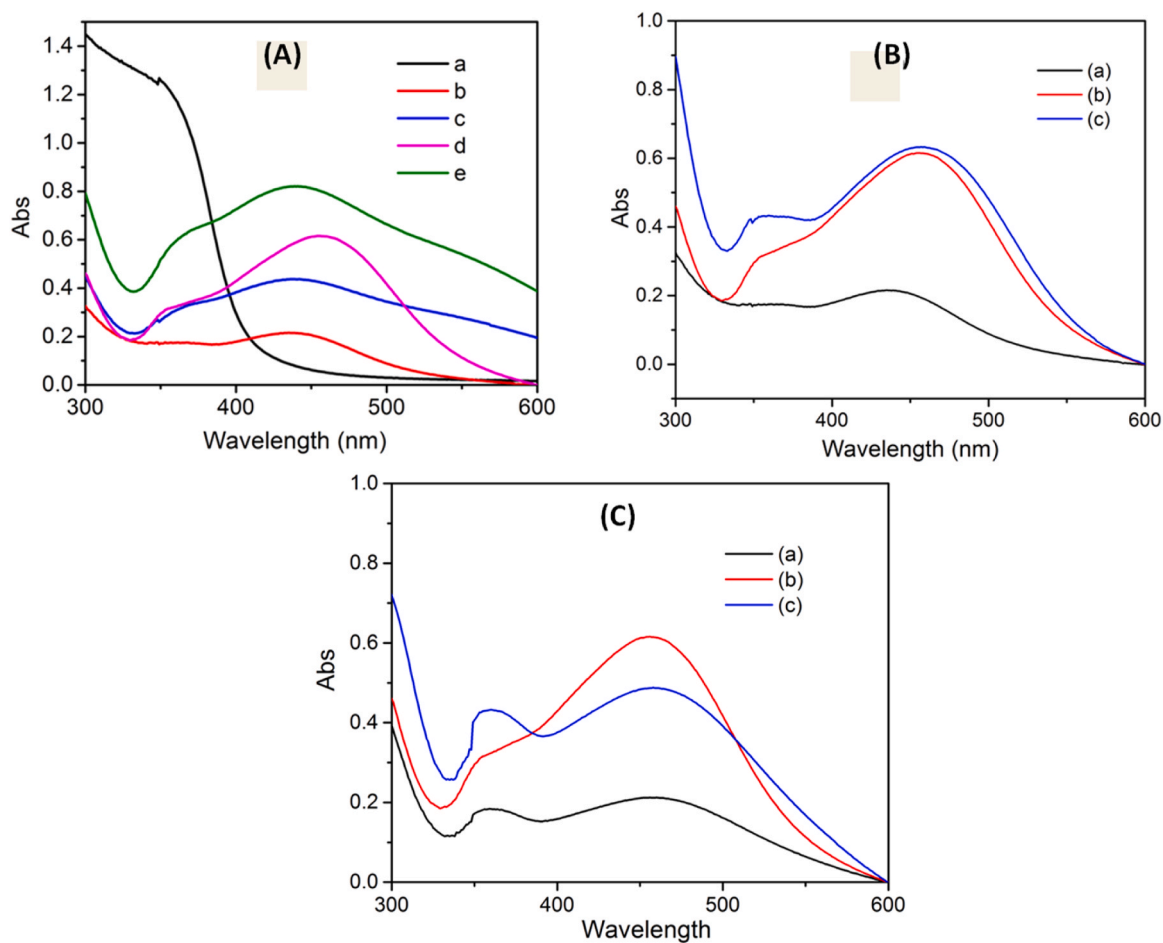


Fig. 2. UV-vis spectra of (A) plant extract with different amount of 0.1 M AgNO<sub>3</sub> (a) 0.0 mL, with (b) 0.2 mL, (c) 0.4 mL, (d) 0.8 mL, and (e) 1 mL solution; (B) dependency of microwave irradiation time (a) 1 min, (b) 2 min, and (c) 3 min; and (C) dependency of microwave irradiation temperature (a) 60 °C, (b) 70 °C, and (c) 80 °C.

Gram-negative *Vibrio cholerae*, *Escherichia coli*, *Shigella flexneri*, *Shigella boydii*, *Salmonella enterica*, *Pseudomonas fluorescens*; and Gram-positive *Bacillus subtilis* and *Bacillus licheniformis*. Strains were stored at  $-80^{\circ}\text{C}$  in Luria Bertani (LB) broth, Miller (HiMedia, India) supplemented with sterile 20 % glycerol.

## 2.7. Evaluation of antibacterial activity of AgNPs

The effect of CAW and AgNPs on the growth of different bacterial strains was checked by broth dilution method [34]. Overnight cultures were prepared by inoculating a single colony into LB broth and incubating at  $37^{\circ}\text{C}$  with shaking at 120 rpm. Cultures were adjusted to an OD600 of 1.0 and diluted 200-fold with fresh LB (corresponding to  $\sim 105$  CFU/mL). Cultures were incubated with or without AgNP stock solutions at  $37^{\circ}\text{C}$ , 120 rpm for 4 h. Concentration-dependent assays were conducted using AgNP concentrations of 4, 8, 12, 16, 20, 24, 28, 32, 36, and  $40\text{ }\mu\text{g/mL}$  in nutrient broth. The Minimum Inhibitory Concentration (MIC) was also determined. After 4 h incubation, aliquots were serially diluted in phosphate-buffered saline and plated on LB-agar. Colonies were counted after overnight incubation at  $37^{\circ}\text{C}$ . The positive control consisted of diluted bacterial culture without AgNPs. All experiments were performed in triplicate and mean values  $\pm$  SD were calculated.

## 2.8. Live and dead bacterial assay

Bacterial cultures treated as above were used to assess viability. For each strain, the IC50 was determined, and that concentration was used in the viability assay. The LIVE/DEAD® BacLight™ Bacterial Viability Kit (Invitrogen Bioservices India Pvt. Ltd.) was used per the manufacturer's instructions. A staining working solution was prepared by adding  $3\text{ }\mu\text{L}$  SYTO® 9 and propidium iodide (PI) to 1 mL filter-sterilized water. After 4 h incubation, a loopful of culture was spread on a glass slide and  $200\text{ }\mu\text{L}$  of staining solution was applied before drying. Slides were covered, incubated in the dark for 15 min at room temperature, and gently rinsed with sterile water to remove excess stain. Observations were made using a fluorescence microscope (Zeiss Axio imager M1) with excitation at 445 nm; emissions were recorded at 525 nm (green) and 605 nm (red).

## 3. Materials and methods

Fig. 1 illustrates the photographs of the reaction mixture of *S. glauca* leaves aqueous extract and silver nitrate ( $0.1\text{ M AgNO}_3$ ) solution.

The extract turned from yellowish to dark brown during 1–3 min of microwave exposure which showed that AgNP formation occurred. The formation of nanoparticles was confirmed through three different analytical methods which included UV–visible spectroscopy and XRD and TEM analyses. The initial confirmation came from UV–Vis spectroscopy (Fig. 2).

The appearance of an absorbance peak at 454 nm in the spectrum indicates that AgNP formation has occurred (Fig. 2). The AgNP synthesis process achieved success through microwave irradiation treatment of *S. glauca* leaf extract. For AgNPs, peaks between  $\sim 400$ – $500\text{ nm}$  are typical and arise from surface plasmon resonance (SPR), caused by collective oscillation of conduction electrons on the metal surface in resonance with incident radiation [35,36].

### 3.1. Characterization

Phytochemical screening of the CAE and the AgNPs filtrate (post-centrifugation) is summarized below. These tests revealed secondary metabolites such as tannins, phenols, coumarins, saponins, reducing sugars, and carbohydrates. Phenolic constituents function as primary antioxidants and are likely responsible for the extract's reducing ability [37]. The reduced amount of phenolic precipitate in the AgNP filtrate

**Table 1**

Phytochemical screenings of plant extract and bio synthesised nanoparticles.

| S. no | Chemical Constituents | Phytochemical Screening                            |                                           |
|-------|-----------------------|----------------------------------------------------|-------------------------------------------|
|       |                       | Plant Extract                                      | AgNP                                      |
| 1     | Alkaloids             | Not Detect                                         | Not Detect                                |
| 2     | Flavonoids            | Not Detect                                         | Not Detect                                |
| 3     | Terpenoid             | Not Detect                                         | Not Detect                                |
| 4     | Tannins               | Present                                            | Present                                   |
| 5     | Phenols               | Present (instance blue color; with high intensity) | Present (Light blue color; Low intensity) |
| 6     | Coumarins             | Present                                            | Present                                   |
| 7     | Saponin               | Present                                            | Present                                   |
| 8     | Glycosides            | Present                                            | Present                                   |
| 9     | Carbohydrates         | Present                                            | Present                                   |

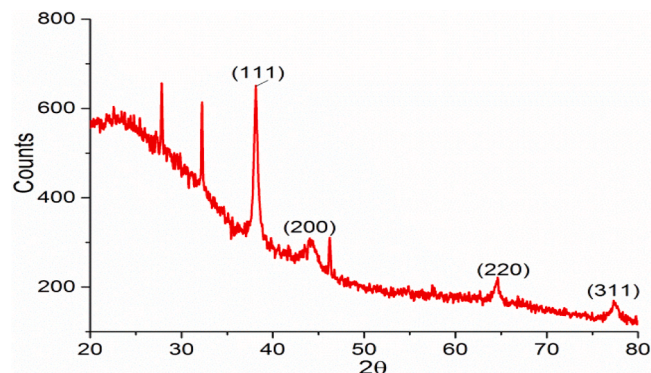


**Fig. 3.** XRD pattern of AgNPs synthesised by microwave irradiation using plant extract.

compared to the CAE suggests phenolics played a role in reducing  $\text{Ag}^+$  to  $\text{Ag}^0$  (Table 1). Although FTIR spectra are commonly used to identify specific functional groups on nanoparticle surfaces, our study did not include FTIR analysis. Nonetheless, several reports have shown that hydroxyl, carboxyl, and phenolic groups from plant-derived compounds typically participate in the reduction and stabilization of AgNPs [38–40]. This is supported by the phytochemical constituents identified in *S. glauca* extract in this work (Table 1).

As *S. glauca* leaf extract was used for AgNP synthesis for the first time, optimization of reaction parameters such as time, temperature, and  $\text{AgNO}_3$  concentration was required. Fig. 2(A) shows UV spectra for reactions using different volumes of  $0.1\text{ M AgNO}_3$  (0.2, 0.4, 0.8, and 1 mL). Peak absorbance intensity increased with higher  $\text{AgNO}_3$  up to 1 mL, probable indicating more AgNP yield (Fig. 2A). Using 1.5 mL of  $0.1\text{ M AgNO}_3$  produced similar absorbance (data not shown), suggesting 1 mL was sufficient for complete silver reduction under the other conditions used ( $70^{\circ}\text{C}$ , 3 min).

Fig. 2(B) shows the effect of microwave irradiation time on AgNP formation, using 1 mL of  $0.1\text{ M AgNO}_3$  at  $70^{\circ}\text{C}$ . AgNP formation began after 1 min, with a light yellow-to-light-brown color change and a weak absorbance at  $\sim 454\text{ nm}$ ; absorbance intensity increased up to 3 min. After 3 min, absorbance plateaued, indicating 3 min as the optimal irradiation time for complete reduction of  $\text{Ag}^+$  to  $\text{Ag}^0$  by *S. glauca* extract. The UV peak intensity rose with time until silver ions were fully converted to nanoparticles [25].

Irradiation temperature was also optimized (Fig. 2C). Samples irradiated at  $60^{\circ}\text{C}$  were slightly paler than those at  $70^{\circ}\text{C}$ , and UV absorbance was lower at  $60^{\circ}\text{C}$  than at  $70^{\circ}\text{C}$ . At temperatures above  $70^{\circ}\text{C}$ , peak intensity decreased with a slight red shift, maybe reflecting formation of AgNPs with larger particle size at higher temperatures. Thus, optimal conditions were 1 mL of  $0.1\text{ M AgNO}_3$ ,  $70^{\circ}\text{C}$ , and 3 min irradiation using *S. glauca* leaf extract. Above mentioned optimisation parameter suggested that this microwave-assisted biosynthesis is rapid,

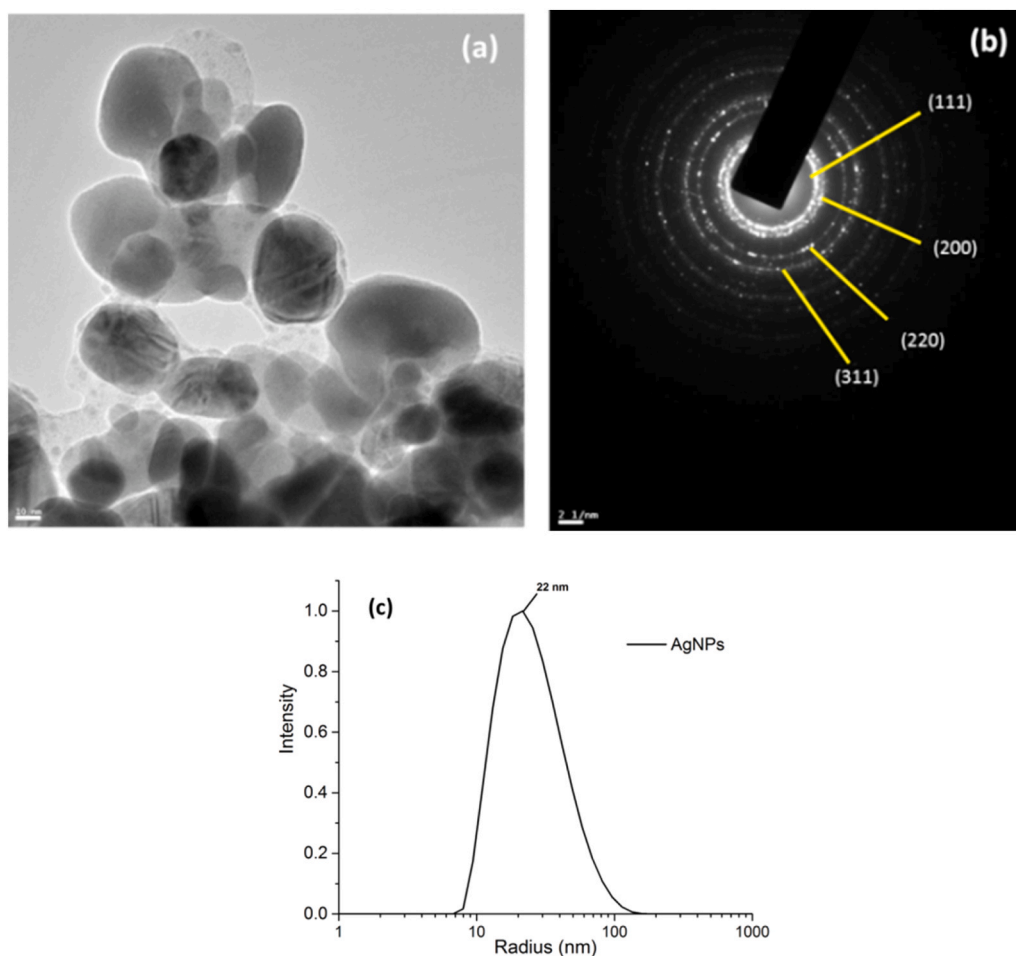


**Fig. 4.** (a) TEM images of AgNPs. Reaction conditions: *S. glauca* extract = 1 mL, 0.1 M  $\text{AgNO}_3$  solution = 1 mL, Irradiation temperature = 70 °C, and time = 3 min; (b) Selected area diffraction images of randomly selected Ag-nanocrystals and; (c) DLS of AgNPs.

eco-friendly, and consumes less energy than many previously reported methods in the presence of *S. glauca* extract.

The XRD pattern has been used for the determination of the crystalline structure of AgNPs. XRD pattern shows four distinct diffraction peaks at  $38.16^\circ$ ,  $44.21^\circ$ ,  $64.66^\circ$  and  $77.4^\circ$  of  $2\theta$  values which are assigned the plane of (111), (200), (220) and (311) correspondingly, indicating a structure of AgNPs is in crystalline form (Fig. 3).

The XRD pattern matches standard silver diffraction data (Joint

Committee on Powder Diffraction Standards, file no. 04-004-4773). Minor peaks at  $27.78^\circ$ ,  $32.34^\circ$ ,  $46.15^\circ$ , and  $57.42^\circ$  may arise from organic material from *S. glauca* bound to the AgNP surface (Fig. 3), consistent with previous biosynthesis reports [26].

The resulting AgNPs synthesised under optimised condition were further analysed to confirm the size and shape using high resolution TEM and DLS (Fig. 4).

TEM results show that this biosynthesis process produced the

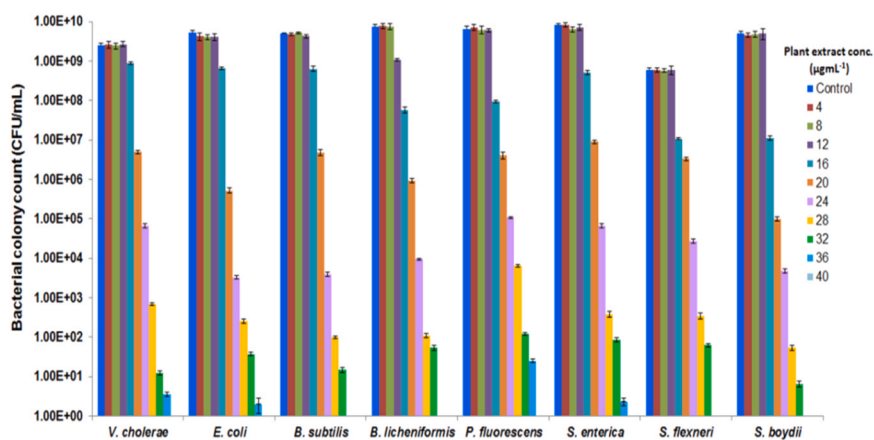


**Fig. 5.** Recovery of Gram-positive and Gram-negative bacterial cells after incubating with different concentrations of AgNPs and CAE as control given the figure legend. X and Y axis indicates the strains name and bacterial colony count respectively. Data are presented as the logarithm of mean  $\text{CFU} \pm \text{SD}$ .

**Table 2**MIC, MBC and IC<sub>50</sub> value of AgNPs against bacterial strains\*.

| Sl no | Strains used                   | MIC | MBC | IC <sub>50</sub> |
|-------|--------------------------------|-----|-----|------------------|
| 1     | <i>Vibrio cholerae</i>         | 28  | 40  | 24               |
| 2     | <i>Escherichia coli</i>        | 28  | 40  | 21.6             |
| 3     | <i>Bacillus subtilis</i>       | 28  | 36  | 23.2             |
| 4     | <i>Bacillus licheniformis</i>  | 28  | 36  | 21.6             |
| 5     | <i>Pseudomonas fluorescens</i> | 32  | 40  | 24               |
| 6     | <i>Salmonella enterica</i>     | 28  | 40  | 24               |
| 7     | <i>Shigella flexneri</i>       | 28  | 36  | 24               |
| 8     | <i>Shigella boydii</i>         | 28  | 36  | 20.8             |

\* All the experiments are performed in triplet and value is given as mean.

homogeneous (spherical) shape of silver nanoparticles in the presence of *S. glauca* leaves extract. Thus, this rapid and eco-friendly bioprocess could be utilised successfully to the synthesis of spherical AgNPs. In addition, TEM results indicated that the particle size of AgNPs were in the between  $\pm 10$ –15 nm (Fig. 4(a)). Fig. 4(b) shows the existence of rings patterns in the selected area electron diffraction, further discloses crystalline nature of the resulting spherical AgNPs with a favoured growth direction along the Ag (110), (200), (220), (311) and (331) planes. This is in the good agreement with XRD result (Fig. 4(b)). Furthermore, Dynamic Light Scattering (DLS) has been used for the size distribution of the AgNPs. DLS result revealed that the average hydrodynamic radius size was  $\pm 22$  nm (Fig. 4(c)). Furthermore, the colloidal stability of silver nanoparticles (AgNPs) synthesized through green methods depends heavily on their surface charge which researchers measure through zeta potential. The research indicates that systems achieve stability when zeta potential values exceed  $\pm 25$ –30 mV because electrostatic repulsion becomes more effective. The surface charge of AgNPs synthesized using plant extracts as capping agents becomes negative when the pH value approaches the isoelectric point from below. Thus, phytochemical capping agents contribute both to charge and steric stabilization of AgNPs [20,41–43].

### 3.2. Antibacterial activity

Antibacterial activity of the resulting AgNPs was tested using different strains (Fig. 5).

The antibacterial activity was evaluated through the incubation of

exponentially grown bacterial strains with various concentrations of silver nanoparticles (AgNPs), while the crude aqueous extract (CAE) was used as the control. Colony-forming unit (CFU) counts were performed for each strain after overnight incubation on LB agar plates. Comparative analyses were conducted between AgNP-treated cultures at different concentrations and untreated control groups. As presented in Fig. 4, inhibitory effects were exhibited by AgNPs against all tested bacterial strains. Bacterial growth was not affected at concentrations between 4–12  $\mu\text{g/mL}$ , whereas concentrations above 16  $\mu\text{g/mL}$  caused substantial logarithmic reductions in all strains except *Bacillus licheniformis*, in which similar inhibition was observed at 12  $\mu\text{g/mL}$ . Increasing concentrations of AgNPs resulted in progressively greater inhibition of bacterial growth (Fig. 4). At 24  $\mu\text{g/mL}$  AgNPs, bacterial growth inhibition of 50 % or higher was observed in most strains. After 4 h of incubation, different levels of bacterial growth suppression were observed depending on the bacterial strain, while no significant effect was observed with CAE.

The bactericidal properties of AgNPs were evaluated using the broth dilution method. The minimum bactericidal concentration (MBC) was defined as the lowest concentration of AgNPs required to achieve a 99.9 % reduction in bacterial cells, indicated by the absence of bacterial colonies on agar plates. No bacterial growth was detected on plates corresponding to the MBC values when compared with the control group. As shown in Table 2, the MBC values of AgNPs against the eight tested bacterial strains were found to range from 36 to 40  $\mu\text{g/mL}$ .

### 3.3. Bacterial viability assessment

LIVE/DEAD BacLight bacterial viability assay revealed that AgNPs obtained under optimised condition was effective against both Gram-negative and Gram-positive bacteria. We observed that in the presence (21  $\mu\text{g/mL}$ ) and absence AgNPs in bacterial culture, the survival rate more than 50 % has declined in *E. coli* and *B. licheniformis* (Fig. 6(a), b, c, d).

After 4 h incubation with AgNPs, viable cells fluoresced green (SYTO 9), while dead cells fluoresced red (PI) (Fig. 6a–d). AgNP-treated cells (Figs. 6b, 6d) predominantly fluoresced red, indicating high PI permeability and cell death; *E. coli* exhibited a larger proportion of dead cells. No morphology changes were noted for cells exposed to plant extract

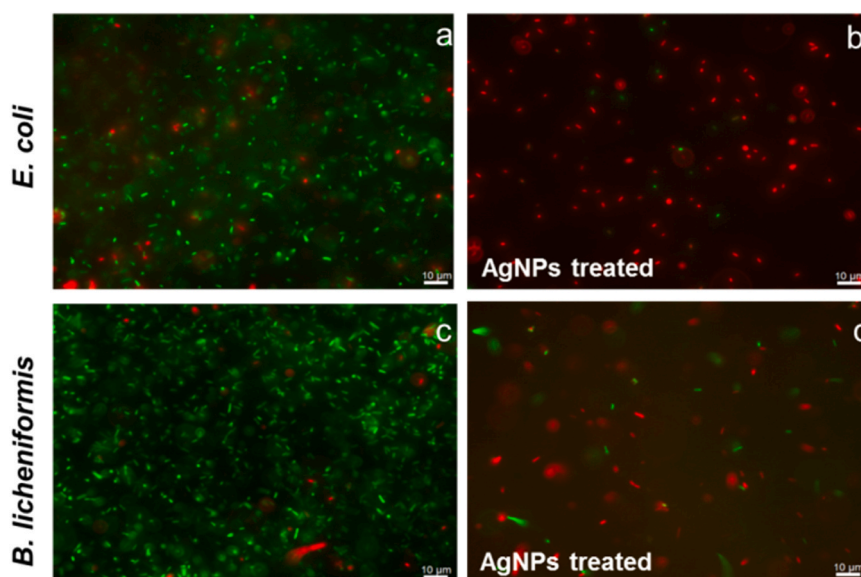


**Fig. 6.** (a) Fluorescence image of *E. coli* in the control (with plant extract); (b) presence of 21  $\mu\text{g/mL}$  of AgNPs. (c) Fluorescence image of *B. licheniformis* in the control and; (d) presence of 21  $\mu\text{g/mL}$  of AgNPs. *E. coli* and *B. licheniformis* all stained by the live/dead BacLight staining kit. The green (fluorescence intensities was measured at 525 nm) and red (fluorescence intensities was measured at 605 nm) fluorescence images are merged together for better assessment of living and dead cells.

alone.

The antibacterial effects of silver nanoparticles (AgNPs) stem from multiple mechanisms which work together to produce their effects. The nanoparticles interact with bacterial cell membranes through two mechanisms which cause membrane damage and permeability increase and silver ion ( $\text{Ag}^+$ ) release from the particle surface. Silver ions have the ability to bind with cellular proteins that contain thiol groups which leads to the disruption of vital enzymes and cellular functions. Research shows that AgNPs produce their broad spectrum of antimicrobial activity through two distinct mechanisms which include direct nanoparticle contact and silver ion release. The antimicrobial properties of AgNPs depend on their surface area and size distribution and their crystalline structure which controls the relative impact of each mechanism on the total effect. Thus, the antibacterial effects reported in this work likely arise from the combined action of silver nanoparticles and silver ion release [16,44,45].

#### 4. Conclusion

A rapid, green, plant-mediated method for AgNP production was developed using microwave irradiation of *S. glauca* aqueous leaf extract. Optimal synthesis occurred within 3 min at 70 °C. The AgNPs were characterized by UV–vis spectroscopy, XRD, TEM, and DLS to determine size and morphology. Antibacterial testing against eight Gram-positive and Gram-negative strains showed significant activity for the bio-synthesized AgNPs, achieving strong antibacterial effects within 4 h across all tested pathogens. MIC and MBC values fell within a narrow range, and IC50 values for AgNPs were notably lower than for the crude aqueous leaf extract. Both Gram-positive and Gram-negative bacteria were susceptible to low concentrations of the AgNPs.

#### CRediT authorship contribution statement

**Saule Z. Nauryzova:** Methodology. **Gopal Bhojani:** Formal analysis. **Dana K. Bolatkan:** Methodology. **Sana K. Kabdrakhmanova:** Writing – review & editing, Validation. **Faisal Kholiya:** Writing – original draft. **Aigul Zh. Kerimkulova:** Validation, Funding acquisition. **Shruti Chatterjee:** Validation. **Anastassiya D. Kukhareva:** Software, Methodology. **Ramavatar Meena:** Writing – review & editing, Supervision. **Ainur K. Kabdrakhmanova:** Software, Data curation.

#### Declaration of Competing Interest

The authors declare that they 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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#### Data availability

Data will be made available on request.

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