

Preparation and analysis of silver nanoparticles (AgNPs) by plant extract techniques of green tea and study optical and structural properties

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Abstract: In order to study the green creation of silver nanoparticles (AgNPs) by decreasing the Ag⁺ ions in a silver nitrate solution, green tea plant extract was utilized. The generated AgNPs were examined by UV-Vis spectroscopy, field emission scanning electron microscopy, and Fourier transform infrared spectroscopy (FTIR). The produced AgNPs were 685 nm in size, spherical in shape, polydispersed in nature, and exhibited a maximum absorbance at 416 nm. *Escherichia coli* and *Staphylococcus* were successfully combated by the AgNPs.

Keywords: green synthesis; Ag; green tea; nanoparticles; antibacterial

1. Introduction

The development of experimental procedures for the creation of nanoparticles with varying sizes, shapes, and controlled disparities is the focus of nanotechnology. With a wide range of possible uses, including pharmaceuticals and medicine, this offers an effective control over numerous physical and chemical characteristics [1]. Currently, noble metals such as platinum (Pt), gold (Au), silver (Ag), and lead (Pb) are used most frequently to create metallic nanoparticles. Of these, silver (Ag) is the metal of choice in the field of biological systems [2]. Using ecologically friendly ingredients like plant leaf extract, bacteria, and fungus for the creation of nanoparticles is a new area of nanotechnology. As harmful chemicals are not used in the synthesis methods, it offers several advantages of environmental friendliness and compatibility for pharmaceutical and biomedical applications [3]. Resistance of disease-causing microorganisms to medication treatment is a growing public health issue. Therefore, the creation of novel bactericides is essential. Since antiquity, Ag salts have been shown to have antibacterial properties. Ag is now utilized in a number of applications, such as dental procedures, catheters, and bum sites, to inhibit bacterial development. In fact, it is well known that Ag ions and Ag-based compounds have severe biocidal effects on bacteria and are very poisonous to them [4]. Mosquitoes are significant human disease carriers, especially in the tropics where they cause the deaths of millions of people each year [5]. Although there are issues with chemical pesticides, such as their toxicity to nontarget organisms, concerns for the environment and human health, and the lack of vaccinations for many illnesses carried by mosquitoes, mosquitoes remain a significant vector insect from the perspective of public health [6,7]. In many tropical nations, notably India, controlling mosquito populations is the only way to lower the prevalence of vector-borne illnesses such malaria, filariasis, dengue, and chikungunya [5,8]. The necessity

to utilize environmentally friendly synthesized AgNPs for the control of disease vectors stems from the identification of the AgNPs as a viable alternative for synthetic chemical insecticides that are less likely to cause ecological harm. The *Delphinium denudatum* Wall, sometimes referred to as *Jadwar*, is a member of the Ranunculaceae family and is one of the most significant medications used in Indian traditional medicine, particularly in the Unani system. The bitter roots are said to be effective in treating a number of illnesses, including aconite poisoning, epilepsy, opium addiction, diuretic, analgesic, antipyretic, cardio, hepatocellular protective, stimulant, tonic, hemorrhoids, insanity, toothaches, rheumatism, snake bite, and more [9,10]. The anti-metal-reducing properties of Ag remain unknown, nevertheless. Therefore, the goal of the current work is to environmentally friendly synthesis silver nanoparticles using an aqueous extract of *D. denudatum*, characterize the created nanoparticles, and explore potential antibacterial and mosquito larvicidal properties.

2. Experimental

2.1. Material

2.1.1. Collection of plant sample

The dried green tea was purchased from reputable vendors in Hyderabad. Green tea's provenance and authenticity were verified.

2.1.2. Bacterial strains

The *Staphylococcus aureus* and *Escherichia coli* bacterial strains were a gift from the biology department at the University of Baghdad. The strains were kept alive at 4 °C on nutrient agar slants.

2.1.3. Hot water extraction

The green tea was cleaned with sterile, twice-distilled water to get rid of any surface impurities before being sun-dried for 10 days. Using a mechanical grinder, the dried plant material was ground into powder. A mixture of 4 g of fine powder and 40 mL of sterile, double-distilled water was prepared and held at 60 °C for 2 h in a water bath. Whatman No. 1 filter paper is then used to filter the slurry. For additional research, the filtered extract was kept in a refrigerator at 4 °C.

For the purpose of creating silver nanoparticles and comparing particle sizes under various conditions, the extract was prepared at both room temperature and boiling point and blended separately with a silver nitrate solution.

2.1.4. Biosynthesis of silver nanoparticles

1.5 mL of plant extract and 30 mL of a 1 mM aqueous silver nitrate solution were combined for the biogenesis of silver nanoparticles, and they were then incubated for 2 h at room temperature and in the dark. The UV-Vis spectroscopy (200–800 nm) of the solutions was measured on a regular basis to track the bio-reduction of the silver ions in the solution. The development of a solution with a yellowish brown hue suggested the emergence of silver nanoparticles. By repeatedly centrifuging the solution at 12,000 rpm for 20 min and dispersing the pellet in sterile deionized water three times, water-soluble biomolecules such proteins and secondary metabolites were removed from the silver nanoparticles that were produced.

Lyophilization was done on the water-suspended nanoparticles. Silver nanoparticles that had been frozen-dried were then employed to describe the composition and structure.

2.1.5. Characterization of green synthesized silver nanoparticles

Using a Cecil spectrophotometer (CE7001), UV-Vis absorption spectra were measured. Utilizing a Phillips 1830 X-ray diffractometer run at 40 kV, 30 mA, with Cu K α 1 radiation in h-2 θ configurations, crystalline metallic silver nanoparticles were studied. The Perkin-Elmer Spectrum One-FTIR 4200 spectrophotometer was used to produce FT-IR spectra for green tea powder and silver nanoparticles in the 4000–450 cm⁻¹ range using the KBr pellet technique. A (HITACHI SU6600) field emission scanning electron microscope (FESEM) was used to examine produced silver nanoparticles.

2.2. Antibacterial analysis

Agar well diffusion method

Using the Mueller Hinton agar (MHA) well diffusion technique, green tea AgNPs were first tested for their antibacterial effects at concentrations of 100, 250, and 500 μ g/well. The use of sterile 1 mM AgNO₃ solution as a blank revealed that it had no effect on any of the employed microorganisms.

3. Results

3.1. Results and discussion

Green synthesis of nano-scale silver particles

The creation of quick, dependable, and environmentally friendly processes promotes interest in the creation and use of nanoparticles that are advantageous to humanity [11,12]. A progressive change in color from clear to yellowish brown occurs after the reduction of silver nitrate into AgNPs after exposure to plant extracts as a result of the surface plasmon resonance (SPR) phenomenon. The color of the root extracts without AgNO₃ remained unchanged. One of the most used methods for characterizing the structure of silver nanoparticles is UV-Vis spectroscopy. Increased incubation period led to the development of the light brown tint. There was a noticeable peak in the absorption spectra of the light brown silver colloids at 416 nm, confirming the existence of a single, approximately spherical particle **Figure 1**.

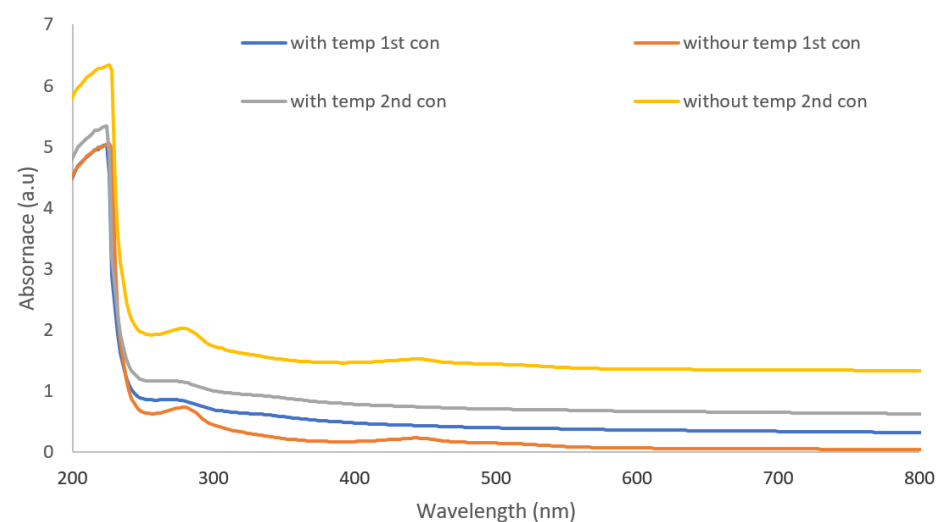


Figure 1. UV-Vis show the absorbance of green tea AgNPs.

SPR, which shifts to longer wavelengths with increasing particle size, dominates the optical absorption spectra of metal nanoparticles [13]. The particle size, the dielectric medium, and the surface-adsorbed species all have a significant impact on the location and form of plasmon absorption of silver nanoclusters [14,15]. Mie's theory [16] predicts that spherical nanoparticles will have a single SPR band in their absorption spectra, whereas anisotropic particles may have two or more SPR bands depending on their form. As the symmetry of the nanoparticle degrades, there are more SPR peaks [17]. The investigation's findings are quite intriguing in that they point to prospective plants in hill stations that may be used to make silver nanoparticles. Significant color shift before two hours suggested the root extract's rapid decrease of AgNO_3 . According to Shankar et al. [18], stable silver, gold, and bi-metallic Ag/Au core shell nanoparticles were quickly biosynthesized using 20 g of *Azadirachta indica* leaf biomass and 1 mM aqueous AgNO_3 , with a 90% reduction of the metal ions occurring in just 4 h. The different activity of the enzymes found in the aqueous leaf and root extracts of *A. indica* AgNps, respectively, may be the cause of the observed discrepancies in the rates of bioreduction.

3.2. FESEM and FTIR analysis

The green tea AgNPs are polydispersed and spherical in shape, according to the high-resolution analysis of the nanoparticles performed using FESEM. **Figure 1** shows the UV-Vis and visible spectra as a function of reaction time of the reaction between a green tea extract and a 1 mM AgNO_3 solution. Regarding **Figure 2**, it depicts the extract's structural makeup in conjunction with silver nanoparticles, clearly demonstrating the mixture's homogenous mixing and spherical shapes by using temperature and without temperature (room temperature soaking).

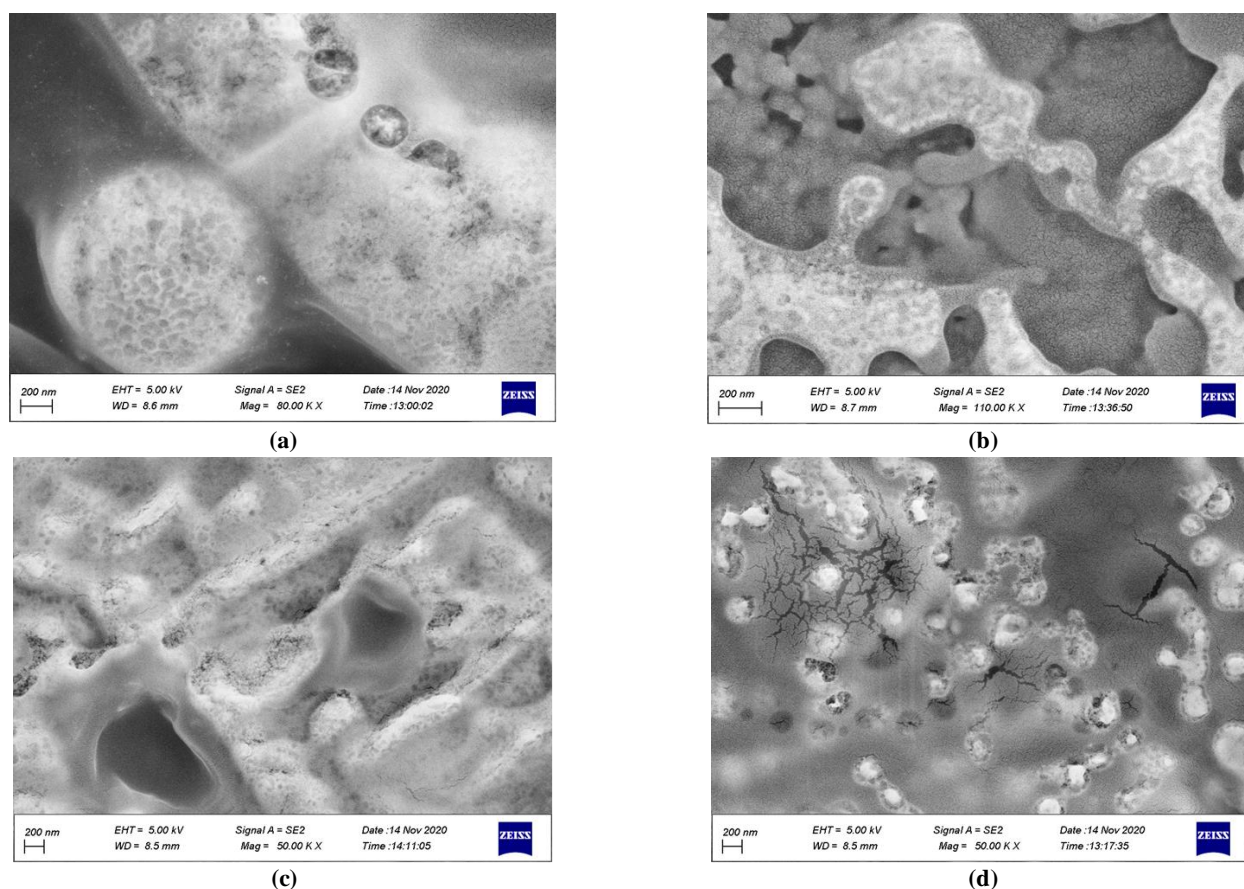


Figure 2. SEM images of particle production in the extract in the presence and absence of high temp, (a) and (c) without high temp; (b) and (d) with high temp.

The resulting particles were no larger than 85 nm in size (**Figure 2**).

To determine the potential biomolecules responsible for the reduction of the Ag^+ ions and capping of the bioreduced silver nanoparticles produced by the root extract, FTIR measurements were conducted on both the aqueous root extract and the manufactured dry silver nanoparticles. AgNPs' main FTIR peaks were found in the **Figure 3** at cm^{-1} 3354, 2952, 2063, 1651, 1419, 1383, 1354, 1171, 1093, 780, 672, and 605. The aqueous root extract's FTIR spectra also showed peaks at 3430, 2923, 2086, 1630, 1406, 1257, 1152, 1029, 860, 762, and 585 cm^{-1} . When comparing the peaks of the spectra of the *D. denudatum* aqueous extract and green tea AgNPs, it was found that after the bioreduction, the peaks at 1257 and 860 cm^{-1} completely vanished and the peaks at 1651, 1419, 1354, and 1171 cm^{-1} intensified. The decrease of Ag^+ ions caused by these modifications may have been primarily caused by the presence of polyols and phenols, which were oxidized to unsaturated carbonyl groups and produced a wide peak at 1660 cm^{-1} [19]. These groups show that various proteins and metabolites, such as terpenoids with amine, alcohol, ketone, aldehyde, and carboxylic acid functional groups, surround the silver nanoparticles produced from the extract [20].

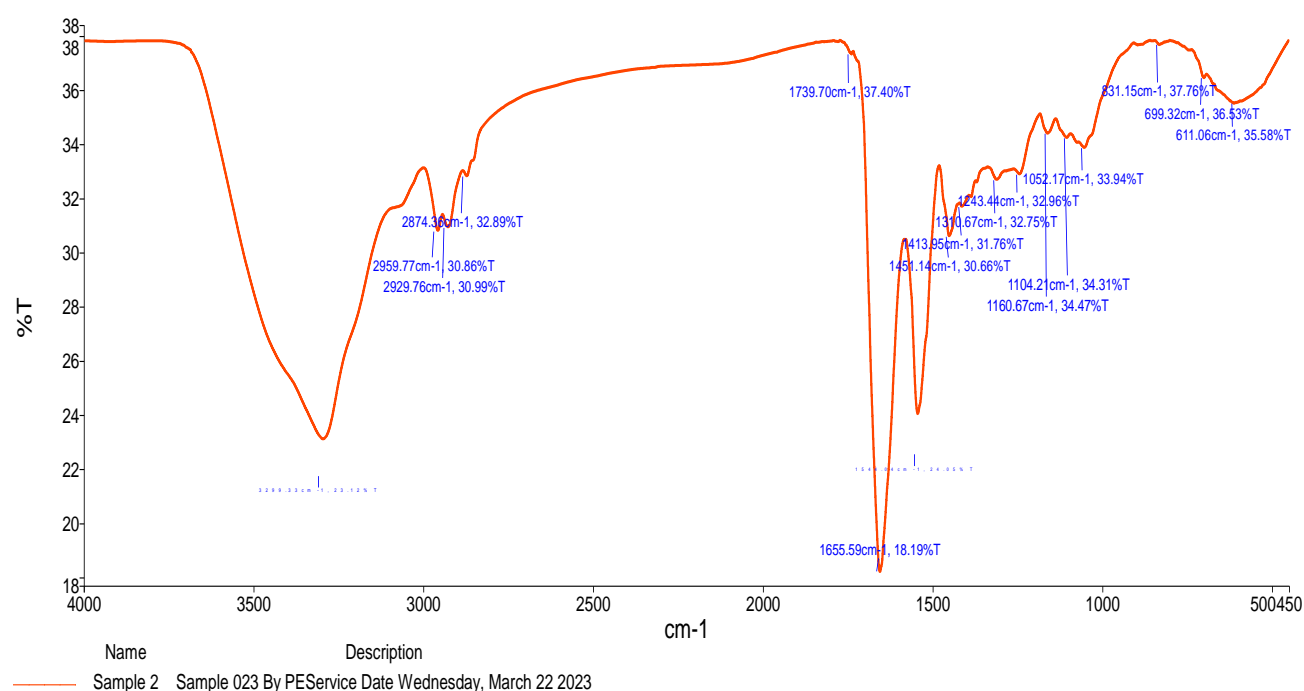


Figure 3. FTIR spectra of Ag nanoparticles synthesized using green tea leaf extracts.

3.3. Antibacterial screening

In **Figure 4**, the impact on bacterial isolates was depicted as ambient temperature for *E. coli* 28 mm (3–5 a), whereas for *S. aureus* 22 mm, the impact was minimal (3–5 b). The biggest inhibition zone, which measured 30 mm for *S. aureus* (3–5 b) and 26 mm for *E. coli* (3–31 a), was found at the high temperature.

For *E. coli* (3–5 a) and *S. aureus* (3–5 b), the largest inhibitory zone measured in ambient temperature high concentration was 25 mm and 32 mm, respectively. AgNPs' ability to stop bacterial cell development is obvious from the inhibition zone that surrounds their wells. AgNPs have gradually increased their activity in the inhibition zone images as a result of the wider inhibition region. We therefore came to the conclusion that the decreased biological activity is caused by C-AgNPs' lesser capacity to release Ag ions [20–23]. These findings unequivocally demonstrate that the various green materials utilized for metal ion stabilization and reduction have a crucial role in determining or adjusting the biological activity of the resulting nanoparticles. As a result, to determine the efficiencies of the acquired nanomaterials, more careful considerations in the selection of green chemicals for the nanomaterial manufacturing process and extensive screening regulations are necessary.

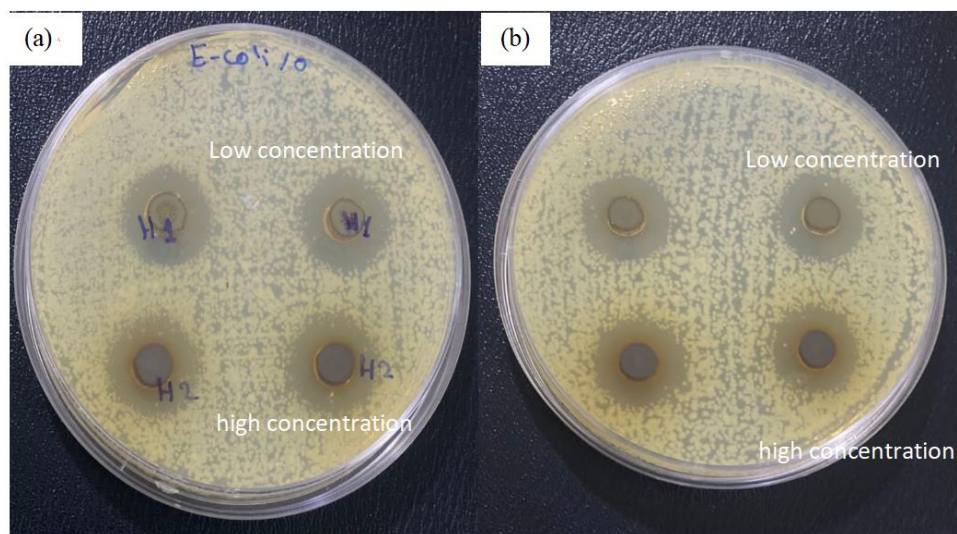


Figure 4. Inhibition zone (mm) of (a) *E. coli* for silver nanoparticles at ambient temperature and high temperature; (b) *S. aureus* for AgNPs.

Limited effectiveness as antibacterial agents due to a number of factors, including the interference caused by salts and the metal's intermittent release of insufficient quantities of silver ions. Silver nanoparticles, on the other hand, can be used to get around these restrictions since they are highly reactive species and have huge surface areas [23]. Against all of the test species, greenly produced silver nanoparticles demonstrated superior antibacterial activity. The antibacterial action that was seen was dose-dependent and grew linearly when the test sample's concentration was raised.

4. Conclusion

The current study attempted the first green manufacture of silver nanoparticles using the aqueous plant extract of green tea. The physical properties of the created nanoparticles were characterized using the proper techniques. We also demonstrated the potential use of green tea AgNPs in the medical field due to their antibacterial activity against human pathogenic pathogens. The knowledge gained from this study expands the corpus of research on the possibility of nanomaterials as an alternative to conventional medicine. But additional studies are needed to determine the crystal structure and toxicological mechanisms of AgNPs.

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