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ORIGINAL ARTICLE

Green Synthesis of Silver Nanoparticles Using *Euphorbia Microsphaera*: Tuning Size and Stability via Precursor Concentration

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Abstract

Background/Aim: The green synthesis of silver nanoparticles (AgNPs) by plant extracts provides an environmentally friendly alternative to traditional chemical methods. This study aimed to report the biosynthesis of AgNPs using *Euphorbia microsphaera*, a native plant of the Middle East and to investigate how different precursor concentrations affect the physicochemical properties of the synthesised nanoparticles.

Methods: The methanolic extracts prepared from *E microsphaera* leaves were employed for the fabrication of AgNPs at varying precursor concentrations such as 1, 3 and 5 mM in silver nitrate ($AgNO_3$). The formation of NPs was observed visually and by measurement with UV-VIS spectroscopy. Dynamic light scattering (DLS) was used to determine the hydrodynamic size, polydispersity index (PDI) and zeta potential. Morphological and elemental analyses were conducted by utilising scanning electron microscopy (SEM), transmission electron microscopy (TEM) and energy dispersive X-ray spectroscopy (EDS).

Results: A clear colour change from pale yellow to dark crimson pink suggested a successful biosynthesis of AgNPs. UV-Vis spectroscopic analysis showed a distinct surface plasmon resonance (SPR) peak at 452.5 nm for the 1 mM sample, indicating the formation of small, monodispersed nanoparticles. In contrast, at 5 mM, a pronounced red shift to 530.5 nm was observed, accompanied by a substantial increase in hydrodynamic size from 238.8 nm to more than 2000 nm. TEM and SEM images supported the results obtained from UV–vis, depicts a well-dispersed spherical shaped nanoparticles in size range 20–50 nm at concentration of 1 mM, whereas 5 mM showed massive agglomeration associated to the saturation of the capping agent.

Conclusion: This study successfully showed the green synthesis of AgNPs by using *E microsphaera*. The results revealed the significant effect of precursor concentration on nanoparticle stability and 1 mM was found to be the optimal value for high stability of monodisperse nanoparticles. *E microsphaera* can be considered as a promising bio-resource for nanotechnology in the biomedicine area.

Key words: Green chemistry technology; *Euphorbia, microsphaera*; Silver; Nanoparticles; Precursor; Concentration.

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Introduction

Nanotechnology has transformed science by enabling manipulation and modification at the atomic and molecular levels, leading to new materials with novel physicochemical properties. Particularly, silver nanoparticles (AgNPs) have aroused much interest due to their outstanding electric conductivity, catalytic performance as well as robust antimicrobial and antiinflammatory activity.[1, 2]

These characteristics make AgNPs especially advantageous for various medical applications, such as wound healing, drug delivery and dermatological treatment.[1] AgNPs are conventionally synthesised by physical and chemical approaches for instance laser ablation and chemical reduction. Nevertheless, these processes generally require high energy consumption, expensive equipment and the use of hazardous chemicals, leading to environmental pollution and preventing their medical applications.[3] In this regard, green synthesis using biological systems, particularly plant extracts, has been recognised as a better alternative because it is environmentally benign and cost-effective. In this approach, plant extracts act as dual-function agents, serving as both reducing agents for metal ions and stabilising agents for the resulting nanoparticles.[4]

Euphorbia (Family: *Euphorbiaceae)* is one of the largest and most diverse genera in flowering plant, distributed widely in tropical and subtropical areas. This genus is reported for its highly bioactive phytochemical constitution such as diterpenoids, triterpenoids, flavonoids and phenolic compounds.[5, 6] Some important species including *Euphorbia hirta, Euphorbia milii* and *Euphorbia helioscopia* have been successfully utilised for the green synthesis of metallic nanoparticles, acting as efficient reducing agents.[7-9] Compared with the widely studied *Euphorbia* species, *Euphorbia microsphaera* remains poorly explored despite being a native plant of the Middle East and possessing a distinctive phytochemical profile.[10] Previous studies have suggested that its unique chemical fingerprint may be useful for nanoparticle synthesis. The novelty of this study lies in reporting, for the first time, the green synthesis of AgNPs using *E microsphaera* extract. Therefore, this work investigates how variations in $AgNO_3$ concentration affect the size, dispersity and stability of the synthesised nanoparticles, with mechanistic insight into the role of phytochemicals in the reduction process. Hence, this study intends to report for the first time the green synthesis of AgNPs by employing methanolic extract of *E microsphaera*. Effort was aimed at studying how alterations in the concentration of precursor ($AgNO_3$) affect the size, dispersity and stability of the achieved nanoparticles with a mechanistic insight on phytochemicals influence in reduction using UV-VIS spectroscopy and dynamic light scattering (DLS).

The aim of this work was to synthesise AgNPs using *E microsphaera* extract and to evaluate how different precursor concentrations influence nanoparticle size, morphology and stability and to clarify that tuning these parameters was expected to optimise nanoparticle characteristics for potential biological applications.

Methods

Plant material collection and authentication

Fresh aerial parts including leaves, stems and flowers of *Euphorbia microsphaera* Boiss were collected during the full blooming period (July–August 2025) at the Baherz area, Baqubah City, Diyala Province, Iraq. This time period was specifically selected to maximise the bioactive secondary metabolite content, as it falls within the pharmacognostic standard for *Euphorbiaceae*.[11, 12] The botanical identification and authentication of the plant materials were carried out by Ali Halub, a specialist at the Directorate of Seed Testing and Certification (DSTC), Ministry of Agriculture, Iraq. A voucher specimen was deposited in the herbarium for future reference.

Post-harvest processing and preparation

Approximately 10 kg of fresh biomass was collected, from which leaves were hand-picked to avoid impurities. The leaves were washed extensively with tap water, followed by rinsing with distilled water to remove soil and contaminants. To prevent thermal degradation of labile compounds (terpenoids and flavonoids), the cleaned

leaves were air-dried in a shaded, ventilated room at 25 °C for two weeks. This process successfully prevented exposure to direct sunlight, which could result in the photodecomposition of chlorophyll and other photosensitive metabolites.[13, 14] The dried leaves were ground into a fine powder using an electric analytical grinder and passed through a 40–60 mesh screen to ensure uniform particle size.[12] The obtained fine powder (400 g) was stored in dark, air-tight amber glass vials at 4 °C until extraction to minimise enzymatic oxidation and maintain metabolite stability.[10, 14]

Sequential Soxhlet extraction protocol

A total of 400 g of powdered plant material was exhaustively extracted using a 2-L Soxhlet apparatus. The extraction was performed sequentially using solvents of increasing polarity: n-hexane, chloroform, ethyl acetate and methanol, respectively.[13, 15] This fractionation strategy was employed to separate non-polar compounds from polar phenolics.[12] First, defatting was performed with 4 L of n-hexane at 75 °C for 25 hours to eliminate lipids, waxes and chlorophyll, yielding 18 g (4.5 % w/w). The dried residue was then extracted with 4 L of chloroform at 65 °C for 20 hours to remove moderately polar ingredients, yielding 13 g (3.25 % w/w).[15, 16] Subsequently, the residue was re-extracted with 4 L of ethyl acetate at 80 °C for 30 hours, yielding 9 g (2.25 % w/w).[17, 18] Finally, the marc was extracted with 4 L of methanol at 70 °C for 18 hours to obtain the polar phenolics and glycosides fraction, which yielded 23 g (5.75 % w/w).[19, 20] All extracts were filtered through Whatman No 1 filter paper, concentrated using a rotary evaporator and dried to a constant weight.

Quality control measures

Strict quality control measures were implemented to ensure the reproducibility and safety of the extracts. The moisture content of the plant powder was maintained below 10 % (measured by oven drying at 105 °C for 3 h) to ensure effective extraction efficiency.[21] Furthermore, the content of residual solvents in the obtained extracts was monitored and reduced to less than 0.1 % to prevent cytotoxicity. The percentage yield was gravimetrically determined using the following equation: Yield (%) = (Weight of extract / Weight of starting powder) × 100.[13]

Preparation of extract solution for nano-synthesis

The dried methanol extract, selected because of its richness in phenolic compounds, was used for the preparation of silver nanoparticles. The methanolic extract can act as natural reducing and stabilising agents and methanol is a commonly used solvent for recovering such bioactive constituents. For the stock solution, 0.1 g of dry extract was dissolved in 10 mL of deionised water (to a concentration of 10 mg/mL, ie 1 % w/v). The samples were sonicated for 10 min to dissolve and homogenise completely. It was then filtered through a 0.45 µm syringe filter to remove any dissolvable particles.[22] The clear, amber filtrate obtained was used directly in the synthesis.

Green synthesis of silver nanoparticles (AgNPs)

Silver nitrate ($AgNO_3$) was used as metal precursor in the present report. Three different concentrations (1 mM, 3 mM and 5 mM) of aqueous $AgNO_3$ solution were prepared using deionised water. The synthesis reaction was initiated by mixing 1 mL of the plant extract solution (10 mg/mL) with 9.0 mL of each $AgNO_3$ solution at a 1:9 v/v ratio in separate flasks. The reaction mixtures were magnetically stirred at 60 °C under continuous stirring and kept in the dark for 90 min, then excess was shaken with another 60 min at room temperature to ensure total reduction.[23] The methanolic extract was mixed with aqueous $AgNO_3$ solution under controlled conditions. The reaction mixture was maintained at room temperature and the pH was adjusted to a neutral to slightly alkaline range to favour nanoparticle formation. The synthesis was allowed to proceed for a fixed incubation time until a visible colour change indicated the formation of AgNPs. All reactions were performed under the same conditions to ensure reproducibility. The visual confirmation of the initial conversion of Ag+ into AgNPs appeared as a colour change from pale yellow to a bright crimson pink.

Purification and characterisation of AgNPs

Synthesised nanoparticles were separated by centrifugation (10,000 rpm, 15 min) from the reaction mixtures. The supernatant was removed and the resulting pellets were resuspended in deionised water and centrifuged again. The washing was carried out two times to completely remove any unreacted silver ions and remaining plant extract.[24]

The optical properties of the nanoparticles were

analysed by Shimadzu UV-1900I spectrophotometer (*Shimadzu Corp*, Kyoto, Japan) in the range 350–700 nm at BPC Analysis Centre in Iraq. Further, the hydrodynamic size (Z-average) and polydispersity index (PDI) were determined using DLS, with a Zetasizer system (*Malvern Panalytical*, UK) at the College of Pharmacy Laboratories, Iraq University–Baghdad.

Results

Visual observation of biosynthesis

Upon addition of *E microsphaera* leaf extract to the aqueous silver nitrate solution, a gradual colour change was observed in the reaction mixture. After 90 min of incubation at 60 °C, the colour changed from light yellow to deep red-brown (carmine pink), which confirmed the reduction of silver ions (Ag+) to zero-valent elemental AgNPs (Ag0). This colour change was observed at all concentrations tested, with significant differences in intensity associated to the precursor concentration. The 1 mM solution was a bright crimson pink, suggesting that small and stable nanoparticles were present. On the other hand, reddish-brown colour intensity was slightly darker than 1 mM solution indicating the higher concentration of produced particles. The 5 mM sample appeared dark brown and turbid, implying the formation of larger aggregates. Significance no colour change was observed in the control solutions, silver nitrate or plant extract alone (under same experimental condition).

UV-Vis spectroscopy analysis

UV-Vis spectroscopy was used to monitor the formation of silver nanoparticles by detecting the surface plasmon resonance band. The appearance and shift of the absorption peak provide preliminary evidence of nanoparticle formation and changes in particle size or aggregation. The synthesis of AgNPs was confirmed by UV-Vis spectrophotometry through the presence of surface plasmon resonance (SPR) bands. Figure 1(A-C) shows the absorption spectra of AgNPs produced at different precursor concentrations. At the concentration of 1 mM, A sharp, well isolated absorption peak was found at 452.5 nm which corresponds with a molar extinction coefficient of around 1.2 (au). This activity indicates that monodispersed nanoparticles are formed. The 3 mM concentration exhibited also a peak (though similar to the previous one) at 454.0 nm, but with slightly broadened appearance and lower intensity compared with the 1 mM sample. By comparison, the SPR peak at 5 mM red-shifted significantly to 530.5 nm and was broadened and flattened as well. This shift suggests a change in particle size or aggregation state.

Particle size and zeta potential analysis (DLS)

DLS was used to determine the hydrodynamic diameter and size distribution of the nanoparticles in suspension. This technique also provides information about particle dispersion and possible aggregation behaviour in the liquid medium.

The hydrodynamic size (Z-average) and polydis-

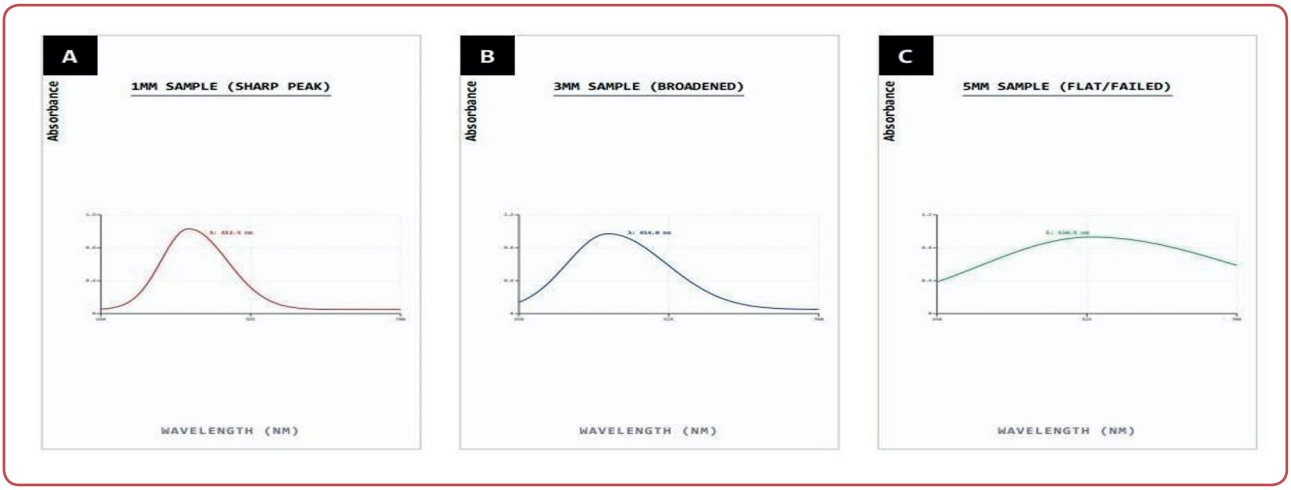

Figure 1: UV-Vis spectra of silver nanoparticles (AgNPs) synthesised using different $AgNO_3$ concentrations (1, 3 and 5 mM) with E microsphaera extract. The surface plasmon resonance (SPR) peak shifted from 452.5 to 454.0 nm and broadened to 530.5 nm at higher precursor concentration, indicating increased particle size and aggregation

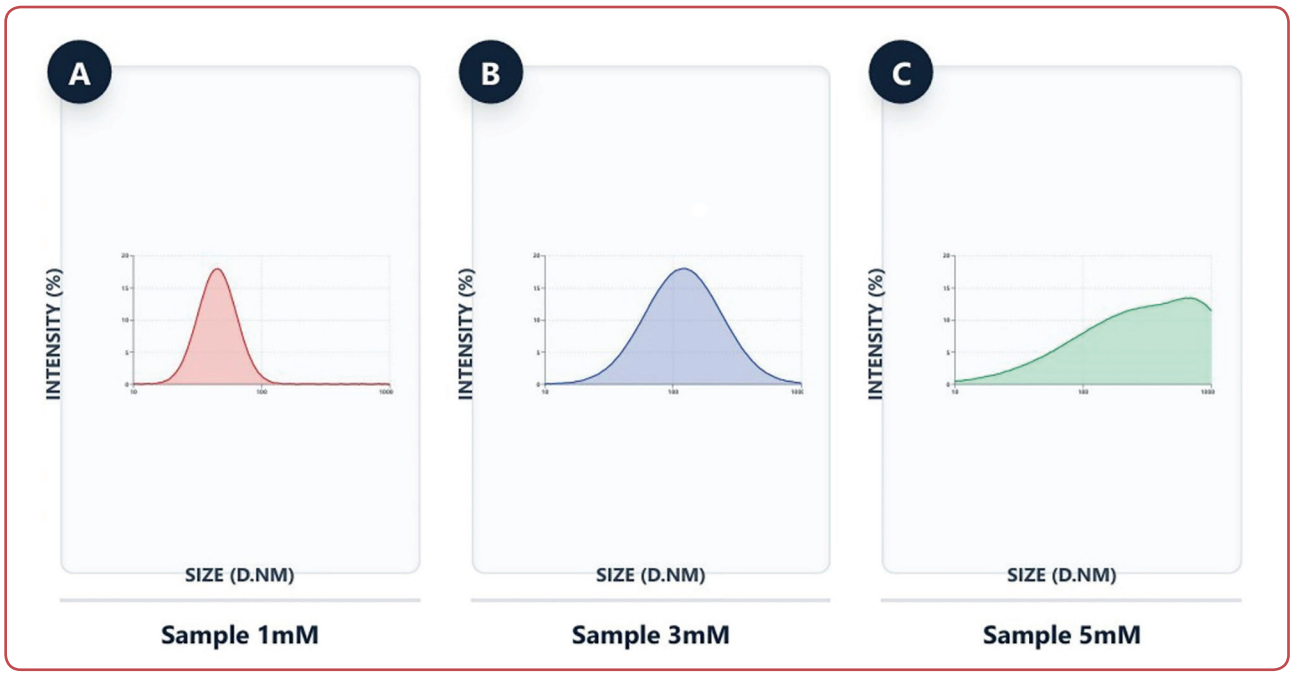

Figure 2: Dynamic light scattering (DLS) size distributions of silver nanoparticles (AgNPs) synthesised at 1, 3 and 5 mM $AgNO_3$. The 1 and 3 mM samples showed relatively narrow distributions with similar Z-average sizes, while the 5 mM sample exhibited a broader distribution and larger hydrodynamic diameter, indicating increased aggregation

persity index (PDI) of the prepared nanoparticles were measured by DLS. The results are summarised in Table 1. The AgNPs prepared at 1 mM showed a Z-average particle size of 238.8 nm and a PDI of 0.380, indicating a fairly monodisperse sample); Thus, indicating a fairly monodispersed in size. By comparison, the sample prepared at 3 mM exhibited a slightly higher Z-average of 256.1 nm, but maintained a similar PDI (0.371). The 5 mM sample exhibited a drastic increase in particle size with Z-average value of 2021 nm and PDI of 0.271. This dramatic change in behaviour is indicative of formation of large, rather than stable nanoscale dispersions (Figure 2).

Morphological and elemental analysis (SEM, TEM and EDS)

The surface morphology, structural features and elemental composition of the fabricated AgNPs were thoroughly studied using scanning electron microscopy (SEM), transmission electron microscopy (TEM) and energy-dispersive X-ray spectroscopy (EDS).

Scanning electron microscopy was employed to examine the surface morphology and overall shape of the synthesised nanoparticles. This technique provides detailed information about particle distribution, surface texture and the degree of aggregation.

It was observed that particle dispersion is highly dependent on precursor concentration (Figure 3) by SEM analysis. At the lowest nanoparticle concentration of 1 mM, smooth surface morphology and well-dispersed spherical shaped particles were observed which suggests that the phytochemicals present in *E microsphaera* extract acted as efficient capping agent and prevented

Table 1: Hydrodynamic size (Z-average) and polydispersity index (PDI) of silver nanoparticles (AgNPs) synthesised using E microsphaera extract at different silver nitrate concentrations

Sample concentration ($AgNO_3$)	Z-average (d. nm)	PDI	Observation
1 mM	238.8	0.380	Monodispersed, stable nanoparticles
3 mM	256.1	0.371	Slight increase in size, stable
5 mM	2021.0	0.271	Extensive agglomeration (micro – scale)

their agglomeration. However, as the precursor concentration was further increased to 3 mM and 5 mM, a remarkable change in morphology was observed. Particles were densely packed and appeared to show signs of surface agglomeration into larger irregular conglomerates. This can be attributed to saturation of the bioactive capping agents by the increased number of metal ions, resulting in insufficient coverage and subsequent aggregation.[25]

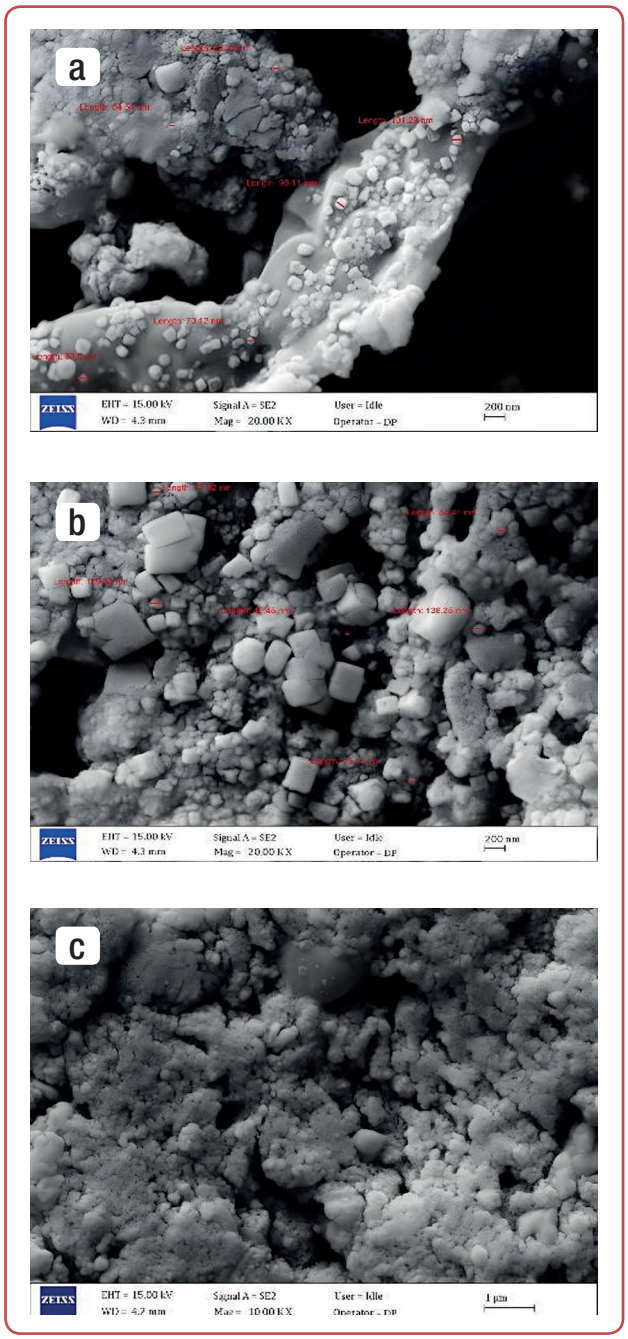

Figure 3: Scanning electron microscopy (SEM) micrographs of silver nanoparticles (AgNPs) synthesised at 1, 3 and 5 mM $AgNO_3$. The 1 mM sample showed well-dispersed nanoparticles, whereas 3 mM showed increased particle density and 5 mM showed marked agglomeration

TEM was used to observe the internal structure and precise morphology of the nanoparticles at high resolution. It allows direct visualisation of particle shape, size and the presence of agglomeration or capping layers. TEM provided insight into the internal core diameter and aggregating property of the nanoparticles (Figure 4). The micrographs confirmed that the primary particles were predominantly spherical or near spherical.

At 1 mM (Figure 4A) were uniform, monodispersed and isolated with a mean core size of 20–50 nm for the AgNPs. This dark metallic core was surrounded by a weak, shapeless layer, which proved that organic capping provides steric stabilisation. At 3 mM (Figure 4B), the images represented an intermediate phase of the aggregation initiation. Although single particles could still be distinguished, they became more densely packed loose clusters due to reduced particle-to-particle spacing. At 5 mM (Figure 4C), dramatic morphological changes occurred. The nanoparticles were no longer distinguishable and formed big bundles with high density. TEM at high magnification revealed direct evidence of particle sintering and coalescence, leading to the formation of silver networks. This is in accordance with the increased hydrodynamic size (> 2000 nm) as observed from DLS data. This indicated that at 5 mM, the electrostatic and steric repulsion forces of the extract were not strong enough to offset the high surface energy of a large number of nanoparticles, resulting in irreversible deposition.

The difference between DLS and TEM sizes can be explained by the fact that DLS measures the hydrodynamic diameter of particles in suspension, including the solvent layer and any surface-bound molecules or small aggregates, whereas TEM measures the actual physical core size of dried particles. Therefore, DLS values are usually larger than TEM values, especially for nanoparticles with surface coatings or some degree of aggregation.

Energy-dispersive X-ray spectroscopy was used to confirm the elemental composition of the biosynthesised nanoparticles. It verifies the presence of silver and may also detect elements associated with plant-derived capping agents or sample preparation.

The elemental composition of prepared NPs was proved EDS analysis that is a critical method to identify and quantify the element and elements in

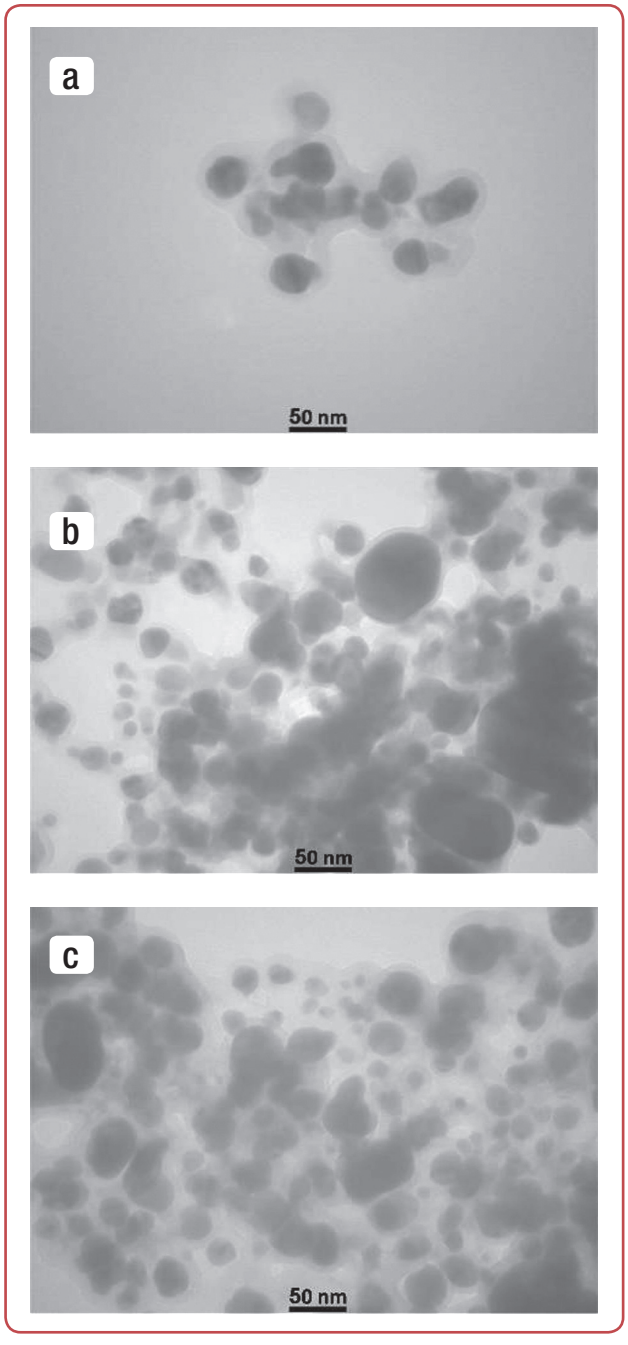

Figure 4: Transmission electron microscopy (TEM) images of silver nanoparticles (AgNPs) synthesised at 1, 3 and 5 mM $AgNO_3$. The 1 mM sample showed isolated spherical nanoparticles, the 3 mM sample showed early aggregation and the 5 mM sample exhibited strong agglomeration

a sample (Figure 5). In the EDS spectrum, a sharp optical absorption peak was found at about 3 keV characteristic of metallic Ag, thus demonstrating the reduction of silver ions. An Oxygen (O) signal, was also present associated with the organic phytochemicals adsorbed on the surface of the nanoparticle. The Gold (Au) also got peaks which could be related to the thin gold coating applied during sample preparation for making it conductive. This elemental composition has not only verified the purity of AgNPs but also revealed the capping by organic agents.[26]

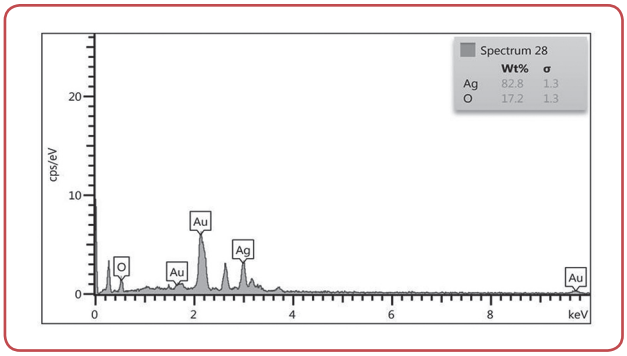

Figure 5: Energy-dispersive X-ray spectroscopy (EDS) spectrum of the biosynthesised silver nanoparticles (AgNPs) showed a strong Ag peak at round 3 keV, confirming the presence of silver. The O peak at 0.52 keV indicates phytochemical capping, while Au peaks are attributed to the gold coating used before analysis

The effect of precursor concentration on AgNP formation was evident across all characterisation techniques. At 1 mM $AgNO_3$, the nanoparticles showed a sharp UV-Vis SPR peak at 452.5 nm, a relatively narrow DLS distribution with a Z-average of 238.8 nm and well dispersed particles in SEM and TEM images. At 3 mM, only slight changes were observed, including a small red shift in the UV-Vis peak to 454.0 nm and a modest increase in DLS size to 256.1 nm, suggesting limited growth and early aggregation. In contrast, at 5 mM, the UV-Vis peak became broader and strongly red-shifted to 530.5 nm, while SEM and TEM revealed extensive agglomeration and larger particle networks. Overall, increasing precursor concentration led to progressively less stable nanoparticles with greater aggregation and larger effective particle size.

Discussion

The present work demonstrates a potential biosynthesis of AgNPs with the methanolic extract of *Euphorbia microsphaera*. Among the sequentially obtained fractions, the methanolic extract was prioritised for green synthesis due to its expected higher content of redox-active phytochemicals, particularly phenolics and flavonoids, which are effective in nanoparticle formation and stabilisation. A significant colour change from light

yellow to deep pink was observed as a main indication of the formation of AgNPs, corresponding to the excitation of SPR.[25]

This reduction is attributed to the presence of phytochemicals in the extract, mainly phenols and flavonoids. These molecules act as electron donors and convert Ag+ to Ag0, which reduces the ions to elemental silver while capping the nanoparticles for stabilising them.[26, 27]

The interconnection between particle size, stability and precursor concentration is a key observation made in this study. Characterisation UV-VIS spectroscopy as seen in Figure 3b, the UV-VIS spectra of Ag NPs with a concentration of 1 mM showed a sharp SPR peak at 452.5 nm, suggesting the formation of small size and narrow distributed Ag NPs.[28] In contrast to the previous sample, the red-shifting of 530.5 nm and broadening in larger size of at least 5 mM indicates that there is still a substantial increment in particle size as well as anisotropy.[24] This may be caused by competition of nucleation and growth kinetics.

At the lower concentration (1 mM), the high ratio of capping agents to metal ions enabled faster nucleation and efficient surface coating, thereby preventing further growth. On the other hand, at higher concentrations (5 mM), available capping molecules are no longer present in sufficient quantities to fully cover excess silver ions; a phenomenon known as 'capping agent starvation'. This transition gives rise to a growth dominated scenario in which particles coalesce via mechanisms such as Ostwald ripening and coalescence process, reducing their surface energy.[29, 30]

The results obtained from spectroscopic study were confirmed by DLS and TEM measurements. The 1 mM sample has a > DLS Z-average of 238.8 nm, which is higher than what was found for the metallic core size in TEM (20–50 nm) and reflect the hydrodynamic characteristic of DTS. This hydrodynamic size includes the metallic core, the solvation shell and the capping layer of phytochemicals.[31] This natural coating serves a key function of steric hindrance needed to preserve stability. Noticeably, the much larger hydrodynamic size (> 2000 nm) for the 5 mM sample correlates well with the TEM images which showed formation of interconnected networks and fused agglomerates. This observation indicates that in higher precursor concentration the balance between electrostatic and steric repulsion forces are disturbed, leading towards irreversible aggregation and formation of clusters at micro-scale.

The present findings are in line with previously reported plant-mediated green synthesis studies, which have shown that phytochemicals in plant extracts can serve as both reducing and capping agents during AgNP formation. Similar to earlier reports, the synthesis outcome in our study appears to depend on key reaction parameters such as extract composition, $AgNO_3$ concentration, mixing ratio, pH, temperature and reaction time, all of which are known to influence nanoparticle size, morphology and colloidal stability. Previous studies have also demonstrated that different plant extracts may produce AgNPs with variable physicochemical characteristics, even under broadly similar experimental conditions, reflecting differences in the abundance of phenolics, flavonoids and other redox-active constituents. Therefore, the properties observed in the current work are consistent with the broader literature on green synthesis, while also highlighting that optimisation of the synthesis protocol is essential for obtaining nanoparticles with desirable characteristics.

The effect of precursor concentration on AgNP formation was evident across all characterisation techniques. At 1 mM $AgNO_3$, the nanoparticles showed a sharp UV-Vis SPR peak at 452.5 nm, a relatively narrow DLS distribution with a Z-average of 238.8 nm and well-dispersed particles in SEM and TEM images. At 3 mM, only slight changes were observed, including a small red shift in the UV-Vis peak to 454.0 nm and a modest increase in DLS size to 256.1 nm, suggesting limited growth and early aggregation. In contrast, at 5 mM, the UV-Vis peak became broader and strongly red-shifted to 530.5 nm, while SEM and TEM revealed extensive agglomeration and larger particle networks. Overall, increasing precursor concentration led to progressively less stable nanoparticles with greater aggregation and larger effective particle size.

Although the present study demonstrates the successful green synthesis of AgNPs using *E microsphaera* extract, some limitations should be acknowledged. First, the scalability of the method may be constrained by the natural variability in phytochemical composition among plant batches, which can affect nanoparticle formation and stability. Second, the reproducibility of the synthesis may vary depending on extraction con-

ditions, plant origin and seasonal factors. Therefore, further optimisation and standardisation will be necessary before considering large-scale or industrial applications.

Conclusion

This manuscript describes for the first time in literature an ecofriendly synthesis of silver nanoparticles using the methanolic extract of *Euphorbia microsphaera*. The synthesis method is simple, environmentally friendly and highly dependent on precursor concentration. Complete physicochemical characterisation, by means of UV-VIS, DLS, SEMTEM and EDS spectroscopy indicates that a lower precursor concentration (1 mM $AgNO_3$) is the best one to produce with spherical shape, monodispersed and stable AgNPs. These NPs showed a unique SPR peak at 452.5 nm, with a controlled hydrodynamic size. However, using 5 mM gave severe instability with fusion of particles (coalescence) into large interconnected agglomerates. The findings of this study demonstrate that *E microsphaera* potentially can be used as an excellent bio-source in the facile, clean and tuneable fabrication of nanomaterials. Further studies focusing on the biological efficacy of these optimised nanoparticles, particularly against microbial and anti-psoriatic activity, are warranted to verify their therapeutic potential.

Ethics

The study protocol was approved by the Research Ethics Committee of the College of Pharmacy, University of Baghdad (Approval No: RECAUB-CP420250512R), dated 12 May 2025. This study involved plant extracts and did not include experiments on human or animal subjects.

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Conflicts of interest

The authors declare that there is no conflict of interest.

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Data access

The data that support the findings of this study are available from the corresponding author upon reasonable individual request.

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Validation: DAA
Formal analysis: AQH
Investigation: AQH
Resources:
Data curation: AQH
Writing - original draft: AQH
Writing - review and editing: DAA
Supervision: DAA.

References

1. Pradeep M, Kruszka D, Kachlicki P, Mondal D, Franklin G. Uncovering the phytochemical basis and the mechanism of plant extract mediated synthesis of silver nanoparticles. ACS Sustain Chem Eng. 2021;9. doi: 10.1021/acssuschemeng.1c06960.

2. Sati A, Ranade TN, Mali SN, Ahmad Yasin HK, Pratap A. Silver nanoparticles (AgNPs): comprehensive insights into bioapplications and green synthesis approaches. ACS Omega. 2025. doi: 10.1021/acsomega.4c11045.
3. Iravani S, Korbekandi H, Mirmohammadi SV, Zolfaghari B. Synthesis of silver nanoparticles: chemical, physical and biological methods. Res Pharm Sci. 2014;9(6):385-406. PMID: 26339255.
4. Mittal AK, Chisti Y, Banerjee UC. Synthesis of metallic nanoparticles using plant extracts. Biotechnol Adv. 2013;31(2):346-56. doi: 10.1016/j.biotechadv.2012.09.002.
5. Ernst M, Grace OM, Saslis Lagoudakis CH, Nilsson N, Simonsen HT, Rønsted N. Global medicinal uses of Euphorbia L. (Euphorbiaceae). J Ethnopharmacol. 2015;176:90-101. doi: 10.1016/j.jep.2015.10.025.
6. Vasas A, Hohmann J. Euphorbia diterpenes: isolation, structure, biological activity, and synthesis (2008–2012). Chem Rev. 2014;114(17):8579-612. doi: 10.1021/cr400541j.
7. Annamalai A, Nallamuthu T. Green synthesis of silver nanoparticles: characterization and determination of antibacterial potency. Appl Nanosci. 2016;6(2):259-65. doi: 10.1007/s13204-015-0433-z.
8. Caroling G, Priyadharshini MN, Vinodhini E, Ranjitham AM, Shanthi P. Biosynthesis of silver nanoparticles using aqueous leaf extract of Euphorbia milii and study of its antibacterial activity. Int J Curr Microbiol Appl Sci. 2015;4(6):528-35.
9. Bong SM, Kim SH, Kim I, Yang JA, Kim KS. Antioxidant and anticancer activities of silver nanoparticles synthesized from Euphorbia helioscopia leaf extract. J Ind Eng Chem. 2020;84:37-46. doi: 10.1016/j.jiec.2019.12.040.
10. El-Saadony MT, Saad AM, Mohammed DM, Korma SA, Alshahrani MY, Ahmed AE, et al. Medicinal plants: bioactive compounds, biological activities, combating multidrug-resistant microorganisms, and human health benefits - a comprehensive review. Front Immunol. 2025 Apr 28;16:1491777. doi: 10.3389/fimmu.2025.1491777.
11. Evans WC. Trease and Evans Pharmacognosy. 16th ed. London: Saunders Elsevier; 2009. ISBN:9780702029332.
12. World Health Organization. Quality control methods for herbal materials. Geneva: World Health Organization; 2011. ISBN: 9789241500739.
13. Harborne JB. Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis. 3rd ed. London: Chapman and Hall; 1998. ISBN: 0412572605.
14. Azwanida NN. A review on the extraction methods uses in medicinal plants, principle, strength and limitation. Med Aromat Plants. 2015;4(3):196. doi: 10.4172/2167-0412.1000196.
15. Tiwari P, Kumar B, Kaur M, Kaur G, Kaur H. Phytochemical screening and extraction: a review. Int Pharm Sci. 2011;1(1):98-106.
16. Silva GO, Abeysundara AT, Aponso MM. Extraction methods and bioactivity of natural products. Ann Biol Res. 2017;8(1):19-24.
17. Bioactive compounds of plant based food: extraction, isolation, identification, and functional properties. Food Sci Nutr. 2025;13:e70351. doi: 10.1002/fsn3.70351.
18. Abubakar AR, Haque M. Preparation of medicinal plants: basic extraction and fractionation procedures for experimental purposes. J Pharm Bioallied Sci. 2020;12(1):1-10. doi: 10.4103/jpbs.JPBS_175_19.
19. Dai J, Mumper RJ. Plant phenolics: extraction, analysis and their antioxidant and anticancer properties. Molecules. 2010;15(10):7313-52. doi: 10.3390/molecules15107313.
20. Do QD, Angkawijaya AE, Tran Nguyen PL, Huynh LH, Soetaredjo FE, Ismadji S, et al. Effect of extraction solvent on total phenol content, total flavonoid content, and antioxidant activity of Limnophila aromatica. J Food Drug Anal. 2014;22(3):296-302. doi: 10.1016/j.jfda.2013.11.001.
21. Zhang QW, Lin LG, Ye WC. Techniques for extraction and isolation of natural products: a comprehensive review. Chin Med. 2018;13:20. doi: 10.1186/s13020-018-0177-x.
22. Ahmed S, Ahmad M, Swami BL, Ikram S. A review on plant extracts mediated synthesis of silver nanoparticles for antimicrobial applications: a green expertise. J Adv Res. 2016;7(1):17-28. doi: 10.1016/j.jare.2015.02.007.
23. Zaky AA, Akram MU, Rybak K, Witrowa Rajchert D, Nowacka M. Bioactive compounds from plants and by products: novel extraction methods, applications, and limitations. Molecules. 2024;29(2). doi: 10.3390/molecules2902010.
24. Bhadange YA, Carpenter J, Saharan VK. A comprehensive review on advanced extraction techniques for retrieving bioactive components from natural sources. ACS Omega. 2024 Jul 8;9(29):31274-31297. doi: 10.1021/acsomega.4c02718.
25. Mulvaney P. Surface plasmon spectroscopy of nanosized metal particles. Langmuir. 1996;12(3):788-800. doi: 10.1021/la9502711.
26. Marslin G, Siram K, Maqbool Q, Selvakesavan RK, Kruszka D, Kachlicki P, et al. Secondary metabolites in the green synthesis of metallic nanoparticles. Materials (Basel). 2018;11(6):940. doi: 10.3390/ma11060940.
27. Ahmad N, Sharma S, Alam MK, Singh VN, Shamsi SF, Mehta BR, et al. Rapid synthesis of silver nanoparticles using dried medicinal plant of Basil. Colloids Surf B Biointerfaces. 2010;81(1):81-6. doi: 10.1016/j.colsurfb.2010.06.029.
28. Valodkar M, Jadeja RN, Thounaojam MC, Devkar RV, Thakore S. Biocompatible synthesis of peptide capped copper nanoparticles using Euphorbia milii latex. Mater Lett. 2011;65(4):544-7. doi: 10.1016/j.matlet.2010.11.040.
29. Thanh NT, Maclean N, Mahiddine S. Mechanisms of nucleation and growth of nanoparticles in solution. Chem Rev. 2014;114(15):7610-30. doi: 10.1021/cr400544s.
30. Voorhees PW. The theory of Ostwald ripening. J Stat Phys. 1985;38(1 2):231-52. doi: 10.1007/BF01017860.
31. Stetefeld J, McKenna SA, Patel TR. Dynamic light scattering: a practical guide and applications in biomedical sciences. Biophys Rev. 2016;8(4):409-27. doi: 10.1007/s12551-016-0218-6.