



<https://muthjas.mu.edu.iq/>

<http://doi.org/10.52113/mjas04/13.1/18>

Evaluation of antileishmanial activity and hemocompatibility of biosynthesized silver nanoparticles by *Olea europaea* L.

Athraa Ali Hussein¹, Yassir Dakheel Kremsh Alasadiy^{*2}

^{1,2} Department of Biology, College of Education for Pure Sciences, University of Al-Muthanna, Iraq.

*Corresponding author email: dr.yassiralasadiy@mu.edu.iq

Abstract

The focus of this study is the severe global health concern of Cutaneous Leishmaniasis, in particular the growing resistance of the parasites to treatments which are often highly toxic, and have numerous adverse effects. This study aimed to develop an environmentally benign nano-therapeutic approach through the green synthesis of silver nanoparticles. In this framework, the extract of *Olea europaea* L. acted as a natural reducing and stabilizing agent, yielding an environmentally friendly and physiologically safe approach for the synthesis of silver nanoparticles (AgNPs) without the use of toxic chemicals. A notable change in color was observed during the synthesis as an important first indication of the reduction of silver ions and the formation of nanoparticles. Several sophisticated methods were utilized to characterize the structure of the nanoparticles, such as FESEM, AFM, FTIR, and XRD. The analyses confirmed the formation of spherical silver nanoparticles of high structural stability. The hemolysis test revealed a 0% hemolysis rate and thus the complete absence of lysis of red blood cells. This demonstrated a high degree of compatibility of the silver nanoparticles for biomedical applications. The MTT assay was performed to assess the biological activity of the silver nanoparticles against clinical isolates of the *Leishmania tropica* parasite. The results showed a dose-dependent anti-parasitic effect of the silver nanoparticles against the parasite cultivated in 3N medium. The dose of 2000 µg/ml showed the greatest efficacy in suppression of parasite growth with strong statistical significance ($P < 0.0001$). The IC₅₀ value was around 853.7 µg/ml, showing the considerable efficacy of silver nanoparticles in the inhibition of Leishmania parasite

proliferation, and indicating that they could potentially be developed into an effective therapeutic option.

Key words: *Olea* , leaf extract, Hemocompatibility, 3N, MTT

1. Introduction

Leishmaniasis is a major public health concern, as recognized by the World Health Organization. It is classified as an endemic parasitic disease and constitutes a major problem for developing countries with limited healthcare resources. (AL-Hassani & AL-Bsherawee, 2022). It is caused by parasitic protozoa of the *Leishmania* genus, which is transmitted through the bites of infected sandflies. Of the various types of leishmaniasis, cutaneous leishmaniasis is the most common (Nawaz *et al.*, 2020). This disease occurs when the parasites invade and multiply inside the skin cells and is characterized by the development of skin lesions. The disease is usually characterized by the appearance of an itchy and reddish spot at the site of the insect bite, which turns into an ulcer. The lesions caused by cutaneous leishmaniasis can become chronic and often result in scars. The incubation period is 1-4 months and lesions tend to self-resolve within 3 months. Chronic lesions pose a continuing clinical problem (Balahbib *et al.*, 2023). Currently, there is no human vaccine available to prevent leishmaniasis. The traditional treatment options available are associated with a number of limitations such as reduced effectiveness, high costs of treatment, and the potential for toxic side effects. On top of that, the location of the parasites in the cells that macrophages work with creates difficulties in the successful distribution of drugs, which in turn creates the need for the administration of drugs in larger quantities, which could bring about the adverse effects of medications. In this respect, the employment of nanotechnology appears to

be a great strategy in which the modification of nanoparticles will allow the direct insertion of drugs to the target cells of the infected macrophages for the purposes of increasing the destruction of parasites (Sousa-Batista & Rossi-Bergmann, 2018). Silver nanoparticles, which via different cellular routes have potent antileishmanial activity, interact with the surface of the membrane of the parasite and cross the membrane, in turn increasing the permeability of the membrane, osmotic equilibrium disruption, and also produce an increased amount of reactive oxygen species (ROS) which produces an oxidative stress that leads to the death of the parasite cells (Fahim *et al.*, 2024). Anaerobic digestion of pasture with the help of the bacteria *Gluconobacter liquefaciens* produces a variety of organic acids which aid in the bioavailability of the copper, and also the uptake of the bacteria by reducing iron. The simple and rapid method that has been developed for the synthesis of olivaceous silver nanoparticles facilitates good contact between the bacteria and the surface of the copper; as a result, the bacteria colonize the surface quickly, and the organic acids in the solution will be utilized to produce a variety of secondary metabolites, which will be a potential food source for the bacteria. Olive leaves have been selected for the research due to the bioactive substances of the plant. The bioactive substances of this plant such as phenolic compounds, flavonoids, oleuropein, hydroxytyrosol, and others, are known to have various biological activities which along with their other properties, are responsible for the green synthesis of

nanoparticles by acting as reducing agents for the silver ions to form silver nanoparticles, and also as stabilizing or

capping agents to prevent the synthesized nanoparticles from aggregating and maintaining their stability.

The biological features and effectiveness of the nanoparticles for several biomedical usage are enhanced. Therefore, olive leaves are considered a feasible and eco-friendly renewable source for the manufacture of nanoparticles (Rahmanian *et al.*, 2015). There is a considerable therapeutic problem in the epidemiological context of *Leishmania tropica*, the primary cause of cutaneous leishmaniasis in many endemic regions, because the parasite strain has been proven to resist standard therapies, most notably the pentavalent antimonial drugs. The increased drug resistance, and the considerable systemic side effects of existing treatments, underscored the need for the development of alternative treatment options that are both effective and safe in

order to address the growing concern for this public health crisis (Balahbib *et al.*, 2023). Because of the proven antibacterial effectiveness and safety of silver nanoparticles, they can also be considered a responsive and practical approach to the problem of drug resistance (Lee *et al.*, 2018). Therefore, in line with the previously published studies, the present research sought to explore the biological viability of the *Leishmania tropica* parasites after exposure to extracts of *Stevia rebaudiana* and their green-synthesized nanoparticles. To evaluate cytotoxicity and determine the optimum formulation that inhibits the survival of the parasites, the MTT assay was employed.

2. Methods

2.1 Isolation and Identification of *Leishmania tropica*

This study isolated and identified *Leishmania tropica* from clinical samples procured from patients attending dermatology clinics in Al-Musayyib city. The study analyzed 50 clinical samples evenly split between males and females (25 males and 25 females). The study sample comprised individuals of multiple age groups including 13 children, 12 adolescents, and 25 adults.

The study sample's clinical presentations included skin lesions that began as small red papular lesions that later developed into ulcers characteristic of cutaneous leishmaniasis. Clinical samples were collected post treatment blood draws using capillary tubes. The tubes were then subsequently used to isolate and identify the parasites and assess the parasites' viability.

The subsequent use of the isolated parasites involved cytotoxicity studies to evaluate the biological viability of *Leishmania tropica* and to assess the antileishmanial activity of certain plant extracts and their green synthesized nanoparticle formulations against the parasite. Nanoparticle treatments in the MTT assay represented a more

reliable approach to quantifying the activity of parasites/metabolically active versus nonmetabolically active parasites and a more reliable approach to the determination of the half-maximal inhibitory concentrations (IC₅₀), as MTT assays are more reproducible than cell counts (Lee *et al.*, 2018; Mayaa & Alasadiy, 2015). This study is to determine, as suggested by previous studies, the biological viability of *Leishmania tropica* after *Olea europaea* L extracts and their green synthesized nanoparticles. The MTT assay to determine cytotoxicity and to determine the formulation that was most successful in reducing parasite lifespan.

2.2 Preparation of Aqueous *Olea europaea* L. Extract

Fresh *Olea europaea* L. leaves were collected, washed, then air-dried at room temperature for seven days. Once completely dry, the leaves were ground as thoroughly as possible. Five grams of the resulting powder were placed in 500 milliliters of distilled water and placed in an incubator set to 37 degrees Celsius for 24 hours to aid the extraction process, during which time the incubator was set to shake. Following this, the mixture was filtered through several layers of sterile gauze to remove large pieces, and then centrifuged for 15 minutes at 4,500 RPM. Finally, the aqueous extract was rendered clear and pure by additional filtration through Whatman No. 1 filter paper.

2.3 Preparation of Silver Nanoparticles Using *Olea europaea* L. Extract

Silver nanoparticles (AgNP_s) were prepared using an aqueous of *Olea europaea* L. .A 1 mM silver nitrate (AgNO₃) solution was mixed with the plant extract, and the mixture was incubated in a 154 rpm shaking incubator in the dark at room temperature for 48 hours to ensure complete bio reduction of the silver ions. After the incubation period, the sample was centrifuged at 4500 rpm for 30 minutes to separate the nanoparticles. The supernatant was discarded, and the sediment was washed with distilled water to remove impurities and unreacted material. This washing process was repeated three times, with centrifugation after each wash. The sediment was then collected in clean glass containers and dried in a hot air oven at 45 degrees Celsius for 24 hours. Finally, the dried nanoparticle powder was collected and stored in airtight containers for later use.

2.4 The Silver Nanoparticles Characterization

Multiple analytical methods were used to evaluate the physicochemical characteristics of silver nanoparticles derived from the aqueous extract of *Olea europaea* L. The Field Emission Scanning Electron Microscopy (FESEM) analysis was used to examine the morphology and particle size of the synthesized nanoparticles at acceleration voltages of 10 to 12 kV in low vacuum mode and varying magnification ranges. For silver nanoparticles, the surface topography, particle distribution, and nanoscale measurements were analyzed using Atomic Force Microscopy (AFM). Additionally, the reduction and stabilization processes of the nanoparticles were evaluated using Fourier

Transform Infrared (FTIR) spectroscopy to identify the associated functional groups.

2.5 Hemolytic Effect of Silver Nitrate Nanoparticles

To evaluate the hemolytic activity of the nanoplant extract, blood from a healthy volunteer was used. PBS was used as a negative control, while Triton X-100 was used as a positive control. Each of the samples was incubated on a shaking platform at room temperature for 2–4 hours, followed by centrifugation at 10,000 rpm. Supernatants were transferred to a 96-well microplate, and different concentrations of the nanoplant extract (125, 250, 500, and 1000 µg/ml) were added. Using a microplate reader, the absorbance at 550 nm was measured to determine the percentage of hemolysis.

2.6 Assessment of Cytotoxicity and Antileishmanial Activity (MTT Assay)

The antileishmanial activity of silver nanoparticles (AgNPs) generated from *Olea europaea* L. was evaluated via the MTT test. *Leishmania tropica* Promastigotes were grown on 96-well microtiter plates, 150 µL volumes of NNN media were used for each. Multiple concentrations of *Olea europaea* L. AgNPs (62.5, 125, 250, 500, 1000, and 2000 µg/mL) were added into the wells, and 250 µL was adjusted to each well. Culture of parasites without treatment served as negative control. The plates were kept at 26 °C for 24 hours. After this period, each well had 15 µL of MTT solution added, then the plates were assessed for 4 hours. DMSO in the volume of 40 µL was used to solubilize the formazan. An absorbance of

formazan at 620 nm was read with an ELISA plate reader. Cell viability was calculated with the following formula:

$$\text{Viability percentage of cells} = \frac{\text{Treatment} - \text{Blank}}{\text{Control} - \text{Blank}} \times 100\%$$

2.7 Statistical Analysis

Data were analyzed using two-way analysis of variance (ANOVA) with Dunnett's post hoc test. Statistical significance was set at $p < 0.05$. Data is represented as mean \pm standard deviation (SD). All analyses were performed using GraphPad Prism software version 8.4.3.

3. Result

Novel developments from herbal medicine have produced the concept of the 'green synthesis' of nanoparticles via the use of plant extracts. This method is a fast, cost-efficient, and more eco-friendly alternative to most other forms of biological synthesis, resulting in growing scientific curiosity in a multitude of fields. (Vanlalveni *et al*, 2022). Plant mediated nanoparticles also tend to have a wide range of size, and diverse metal use (Singh & Dhaliwal, 2019). The use of silver nanoparticles also has a positive safety profile, and helps combat microbial diseases, supporting their economic use (Singh & Dhaliwal, 2019).

Results from the study showcased the successful isolation and identification of *Leishmania tropica* from the clinical samples of a patient diagnosed with Cutaneous Leishmaniasis at a dermatology clinic in Musayyib. Blood samples were collected with capillary tubes from the lesion site after therapeutic injections, and

were used in the subsequent isolation of the parasite and related laboratory diagnosis. Microscopic examination of the smears stained with the Leishman stain demonstrated the presence of amastigotes in the cytoplasm of the phagocytic cells which

is a part of the histological spectrum of the parasite in the infected skin lesions, thus proving the efficiency of the isolation technique and the accuracy of the laboratory diagnosis.

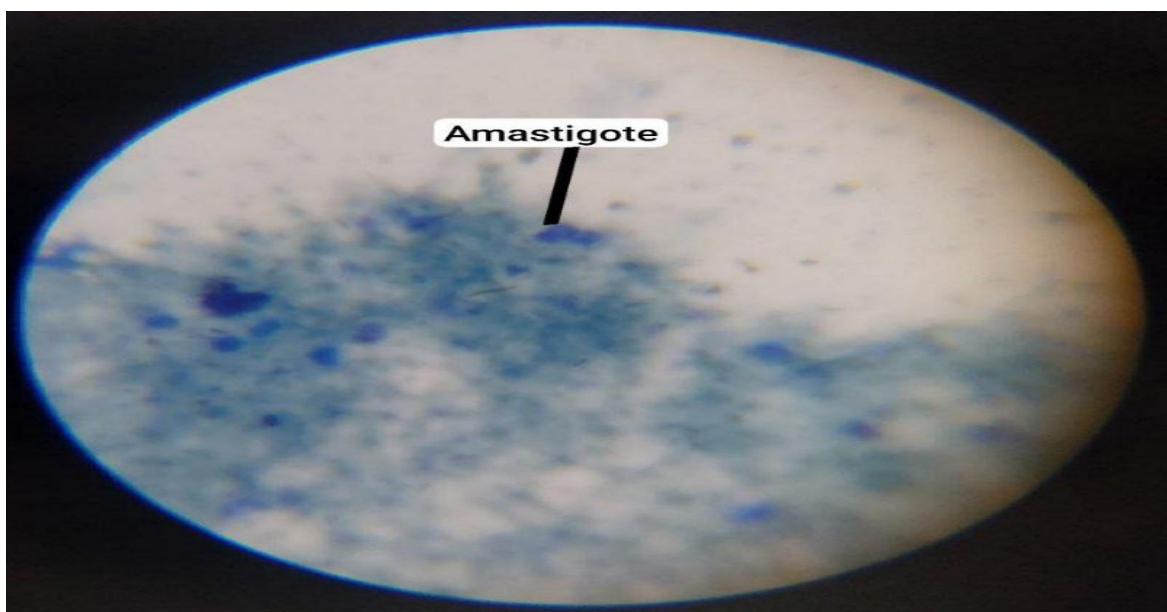


Figure (4-1) Appearance of the amastigote stage of *Leishmania tropica* in clinical specimens using Leishman stain.

Characterization results of silver nanoparticles (AgNPs) made using *Olea europaea* extract. In sections (A & B), Atomic Force Microscopy (AFM) displays some nanostructured surface topography consisting of a number of peaks and clusters, which shows that nanoparticles have been made. Furthermore, section (C) shows that the Field Emission Scanning Electron

Microscopy (FESEM) shows the presence of aggregated nanoparticles of random shapes. In part (D), the AFM line scan profile shows that the surface height has a lot of deviation, which shows that the particles are dispersed unevenly. In section (E), the Fourier Transform Infrared (FTIR) shows several functional groups of the phytochemicals of *Olea europaea* L. that served as the reducers

of the silver ions and as the stabilizers of the obtained nanoparticles. These results altogether prove the successful green

synthesis of silver nanoparticles using *Olea europaea* L. extract.

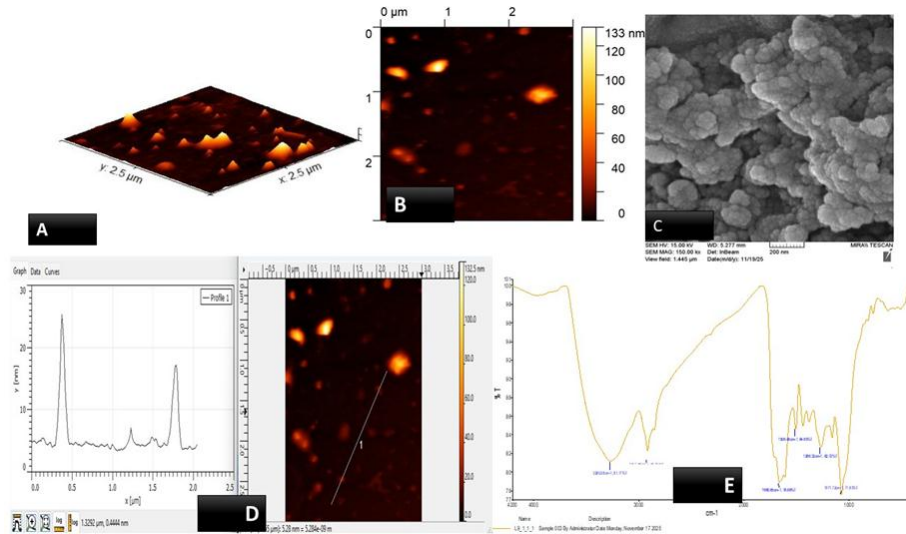


Figure (4-2) Structural and morphological characterization of silver nanoparticles (AgNPs) biosynthesized by *Olea europaea* L. extract

The effect of different concentrations of the tested compound on red blood cell (RBC) hemolysis. The results indicate that all tested concentrations (1, 0.5, 0.25, and 0.12 mg/mL) did not induce any noticeable

hemolysis, with a hemolysis percentage of 0% in all cases. This suggests that the compound is safe and non-toxic to RBCs at these concentrations

Table (4-1) Hemolytic Effect of Silver Nanoparticles at Different Concentrations

concentration (μg/ml)	test result	hemolysis %
1	0.5254	0
0.5	0.4781	0
0.25	0.4315	0
0.12	0.3895	0
POSITIVE	1.7018	100
NEGATIVE	0.3058	0

Table (2) shows the antileishmanial activity of the green synthesized silver nano extract of *Olea europaea* L. against the promastigote stage of *Leishmania tropica*. The findings continue to show a clear dose-dependent inhibitory effect; at concentrations of 2000 and 1000 $\mu\text{g/ml}$ parasites were significantly less compared to the untreated control ($P < 0.0001$). Biocidal

effect at 500 $\mu\text{g/ml}$ was also significant ($P = 0.0315$); in contrast, at lower concentrations (250, 125 and 62.5 $\mu\text{g/ml}$) there was no significant difference ($P > 0.05$) and parasitic promastigote vitality was high and similar to the control group. The data demonstrates the relationship between dose and nanoextract anti-parasitic effect.

Table (4-2) Effect of *Olea europaea* plant-derived nanoparticles in 3N medium

Olive on 3N compared to Untreated Group					
Concentration ($\mu\text{g/ml}$)	SEM	Mean \pm SD		Significant	p- value
2000	4.46	23.57 \pm 7.73		****	<0.0001
1000	4.32	48.31 \pm 7.49		****	<0.0001
500	4.71	84.28 \pm 8.17		*	0.0315
250	5.08	96.45 \pm 8.81		ns	0.9846
125	1.86	93.23 \pm 3.23		ns	0.7450
62.5	2.53	97.46 \pm 4.39		ns	0.9974

Regarding its concentration-dependent parasitic activity, *Olea europaea* L. has shown considerable results of changing *Leishmania tropica*'s structure. The extract

accomplished a semi-inhibitory concentration (IC_{50}) of about 853.7 $\mu\text{g/ml}$. This shows that the extract has the ability to change the survivability of the parasite

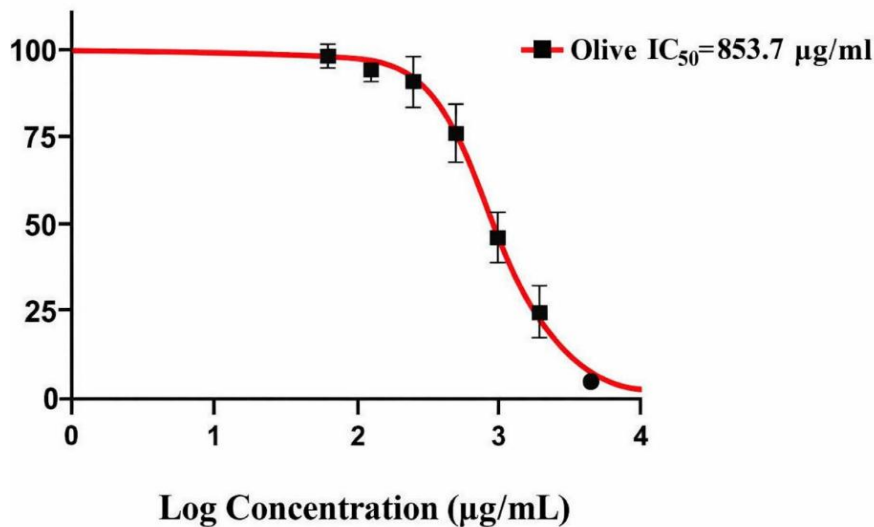


Figure (4-3) Antileishmanial Activity Against Promastigotes in 3N Medium

4. Discussion

This study identified and recorded *Leishmania tropica* from clinical samples of Cutaneous Leishmaniasis patients at Musayyib city dermatology clinics. The Leishman stain demonstrated the presence of the intracellular mastigote of the parasite within the infected skin lesions. The isolation and the diagnosis thus were confirmed (Ekşi *et al.*, 2017). The study further provided examples of the successful biogenesis of silver nanoparticles using the extract of *Olea europaea* L., and during the process, a distinctive color alteration was observed in the reaction solution. This is indicative of silver nanoparticles; the observed color change is attributed to the surface plasmon resonance that occurs as a result of the reduction of silver ions (Ag^+) to their metallic form (Ag^0), which is generally accepted as evidence of the occurrence of the nanoparticles even when prior studies were unsubstantiated (Sati *et al.*, 2025). The successful production of the particles is confirmed by all the tests performed.

In vitro study showed that the in situ synthesized Ag nanoparticles have anti leishmanial properties of the *Leishmania tropica* promastigote stage and appreciable cytotoxic effect on the in situ synthesized Ag nanoparticles on the human red blood cells (Sadeghinia *et al.*, 2024). Sadeghinia *et al.*, 2024 carried out an FTIR examination

which identified several functional groups present on the surface of the nanoparticles such as the hydroxyl and carbonyl groups and suggested that the phytochemicals act as both reducing and stabilizing agents for the nanoparticles. Sadeghinia *et al.*, 2024 carried out Morphological analysis using Field Emission Scanning Electron Microscopy (FESEM) which showed the presence of nanoparticles that were almost spherical and uniformly distributed across the entire nanoscale and that showed little agglomeration. In parallel, Sadeghinia *et al.*, 2024 carried out Atomic Force Microscopy (AFM) which showed the surface morphology of the particles along with the height and other dimensions of the particles that remained in the nanoscale which further confirmed the efficacy of the synthesis and surface distribution of the particles. Hemolysis testing showed that the effect of nanoparticles on the red blood cells, was Ag nanoparticles dependent and concentration dependent. The breakdown of the red blood cells fell at a low concentration and increased at a high concentration which showed that it was necessary to determine a safe concentration for the possible use of the nanoparticles in medicine (Elikaee *et al.*, 2025). In the MTT assay, the leader extract showed activity against the promastigote stage of the *Leishmania tropica* parasite.

The parasite's IC₅₀ value (approx. 853.7 µg/ml) shows that the parasite's survival rate decreases as extract concentration increases,

demonstrating the extract's anti-parasitic properties are dose dependent. The extract's anti-parasitic properties are likely a result of parasitic membrane integrity being compromised, or cell metabolism being disrupted due to antioxidants or phenolic compounds found in olives (Majeed *et al.*, 2023). The MTT assay, which is considered one of the most exact methods of determining cell viability, relies on the cell's ability to convert the yellow MTT compound to blue formazan, which can be measured for quantitative results (Ge *et al.*, 2023). The results of this research demonstrate the unique optical, chemical, and morphological properties of nanoparticles, which are biocompatible with red blood cells and have anti-parasitic properties as evidenced by the mtt assay. Additionally, the biosynthesis of nanoparticle uses olive extract which demonstrates its relative safeness and suggests its great potential in various fields including medicine and biology.

5. Conclusion

The study shows that the synthesis of silver nanoparticles using *Olea europaea* L. extract is a novel and ecofriendly method for the production of stable nanoparticles. A noticeable color change during the synthesis was a first sign of nanoparticles production caused by the reduction of silver ions. Characterization results from FTIR, FESEM, AFM, and XRD techniques suggested the formation of nanoparticles with a good crystalline structure and size uniformity, and the existence of plant functional groups which acted as natural stabilizers. Hemolysis test results showed

good biocompatibility at the tested doses; however, anti-parasitic efficacy was proven by the MTT Assay that showed decreasing viability of the *Leishmania tropica* parasite promastigote phase with increasing concentrations. The study showed that the generated nanoparticles have promising potential for biological and medical applications with concentration optimization needed to improve biocompatibility and therapeutic potential.

Conflict of Interest

The authors have no financial conflict of interest or personal relationships that could have influenced the content of this study.

6. Fund

The authors have self-financed this study. There was no financial support for the research from governmental, corporate, or non-profit entities.

7. data Availability Statement

The original images, raw data, and statistical analyses of this study, as well as the statistical analyses of the study, were deposited in the Figshare repository, and they can be accessed freely using this DOI: <https://doi.org/10.6084/m9.figshare/3136643>
5. Ander (CC BY 4.0)

8. Authors' contributions

The sample collection and laboratory work was performed by graduate student Athraa Ali, while Prof. Dr. Yassir Dkheel Alasadiy provided supervision for this work.

6. References

- Akhter, M. S., Rahman, M. A., Ripon, R. K., Mubarak, M., Akter, M., Mahbub, S., Al Mamun, F., & Sikder, M. T. (2024). A systematic review on green synthesis of silver nanoparticles using plants extract and their bio-medical applications. *Heliyon*, *10*(11).
- AL-Hassani, S. J. M., & AL-Bsherawee, A. I. M. (2022). Epidemiological and histological study of cutaneous leishmaniasis in humans in Al-Najaf Al-Ashraf governorate. *International Journal of Health Sciences*, *6*(S6), 8874–8895.
- Balahbib, A., Hmamouch, A., El Allam, A., Douhri, H., Dahaieh, N., El Omari, N., Chew, J., Ming, L. C., & Bouyahya, A. (2023). Cutaneous leishmaniasis: physiopathology, molecular diagnostic, and therapeutic approaches. *Progress In Microbes & Molecular Biology*, *6*(1).
- Ekşi, F., Özgöztaş, O., Karşlıgil, T., & Sağlam, M. (2017). Genotyping *Leishmania* promastigotes isolated from patients with cutaneous leishmaniasis in south-eastern Turkey. *Journal of International Medical Research*, *45*(1), 114–122.
- Elikaee, S., Taylor, J. K., Carter, N. S., Dwarka, K. M., & Roberts, S. C. (2025). Biogenically Synthesized Nanoparticles as Emerging Therapeutics for Leishmaniasis: A Review of Recent Advances and Potential Promises. *International Journal of Nanomedicine*, 12467–12484.
- Fahim, M., Shahzaib, A., Nishat, N., Jahan, A., Bhat, T. A., & Inam, A. (2024). Green synthesis of silver nanoparticles: A comprehensive review of methods, influencing factors, and applications. *JCIS Open*, *16*, 100125.
- Ge, R., Ye, C., Peng, Y., Dai, Q., Zhao, Y., Liu, S., Wang, P., Hu, L., & He, C. (2023). m6A-SAC-seq for quantitative whole transcriptome m6A profiling (vol 18, pg 626, 2023). *NATURE PROTOCOLS*, *18*(11), 3652.
- Lee, A. S., De Lencastre, H., Garau, J., Kluytmans, J., Malhotra-Kumar, S., Peschel, A., & Harbarth, S. (2018). Methicillin-resistant *Staphylococcus aureus*. *Nature Reviews Disease Primers*, *4*(1), 18033.
- Majeed, Q. A., Shater, A. F., & Alanazi, A. D. (2023). Green synthesis, characterization, and antileishmanial activity of the silver nanoparticles alone and along with meglumine antimoniate against *Leishmania major* Infection. *Iranian Journal of Parasitology*, *18*(4), 535.
- Mayaa, F. A. K., & Alasadiy, Y. D. K. (2015). Effect of cold aqueous plant extract (*Citrus aurantium* and *Solanum melongena*) against the *Giardia lamblia* parasite and *Entamoeba histolytica* in vitro and measuring the LD50 for extracts in Al-Muthanna province. *RESEARCH JOURNAL OF PHARMACEUTICAL BIOLOGICAL*

AND CHEMICAL SCIENCES, 6(6),
1241–1252.

Nawaz, M., Din, M., Khan, A., Khan, A., Ali, M., Din, S. U., & Aslam, K. (2020). Epidemiological features of cutaneous leishmaniasis endemic in hilly areas of district Karak, Khyber-Pakhtunkhwa province of Pakistan. *Journal of Parasitic Diseases*, 44(4), 725–729.

Rahmanian, N., Jafari, S. M., & Wani, T. A. (2015). Bioactive profile, dehydration, extraction and application of the bioactive components of olive leaves. *Trends in Food Science & Technology*, 42(2), 150–172.

Sati, A., Ranade, T. N., Mali, S. N., Ahmad Yasin, H. K., & Pratap, A. (2025). Silver nanoparticles (AgNPs): comprehensive insights into bio/synthesis, key influencing factors, multifaceted applications, and toxicity— a 2024 update. *ACS Omega*, 10(8), 7549–7582.

Sousa-Batista, A., & Rossi-Bergmann, B. (2018). Nanomedicines for cutaneous leishmaniasis. *Leishmaniases Re-Emerging Dis*, 181(10.5772).

Zanbili, F., & Poursattar Marjani, A. (2025). Innovative green and bio-based approaches for photosensitive nanoparticle synthesis: a review on methodologies, characterization, and applications. *Micro and Nano Systems Letters*, 13(1), 3.