ZULFAQAR Journal of Defence Science, Engineering & Technology
e-ISSN: 2773-5281
Vol. 9, Issue 1 (2026)
DOI: https://doi.org/10.58247/jdset-2026-0901-09
Journal homepage: https://zulfaqarjdset.upnm.edu.my

GREEN SYNTHESIS OF SILVER NANOPARTICLES USING *MORUS NIGRA* LEAF EXTRACT AND THEIR ANTIBACTERIAL POTENTIAL

Madihah Mohd Afzal[a], Siti Hajar Adam[b,*], Shirley Tang Gee Hoon[b], Nur Ezza Fazleen Mohd Fathil[c]

[a] Centre for Toxicology and Health Risk Studies (CORE), Faculty of Health Sciences, Universiti Kebangsaan Malaysia, 50300 Kuala Lumpur, Malaysia
[b] Department of Pre-Clinical Department, Faculty of Medicine & Defence Health, National Defence University of Malaysia, Sg. Besi Camp, 57000 Kuala Lumpur, Malaysia
[c] International Medical School, Management and Science University, University Drive, Off Persiaran Olahraga, Section 13, Shah Alam 40100, Malaysia

ARTICLE INFO

ARTICLE HISTORY
Received: 01-12-2025
Revised: 30-01-2026
Accepted: 01-03-2026
Published: 30-05-2026

KEYWORDS
Silver nanoparticles
Green synthesis
Morus nigra
Antibacterial activity

ABSTRACT

This study presents a simple optimisation approach for the green synthesis of silver nanoparticles (AgNPs) using Morus nigra (black mulberry) leaf extract and investigates their antibacterial properties. Three critical synthesis parameters which include aqueous temperature for plant extracts, incubation time and reaction temperature were optimised to enhance AgNP production. The biosynthesis of AgNPs was determined using UV-Visible spectroscopy, which confirmed the successful formation of nanoparticles through characteristic surface plasmon resonance peaks. The optimal conditions identified in this study facilitated efficient conversion of silver ions to stable AgNPs utilising the phytochemical constituents in *Morus nigra* extract as reducing and capping agents. The antibacterial activity of the optimized AgNPs was evaluated against Gram-positive (*Staphylococcus aureus*) and Gram-negative (*Klebsiella pneumoniae*) bacteria using disc diffusion assays. The biosynthesised AgNPs exhibited antibacterial activity against both bacterial strains, demonstrating their potential as effective antimicrobial agents. This research establishes a simple, eco-friendly protocol for AgNP synthesis contributing to the development of green nanomaterials with promising applications in addressing bacterial infections and antibiotic resistance.

1.0 INTRODUCTION

The rise of antibiotic resistance and the ongoing search for new therapeutic agents have driven interest in nanomaterials within biomedical research. Among various nanomaterials, silver nanoparticles (AgNPs) have garnered significant attention due to their remarkable antimicrobial properties and wide-ranging applications in healthcare, textiles, food packaging, and water purification systems. Their effectiveness against a wide range of pathogens, including multidrug-resistant bacteria, highlights their potential in combating the growing challenge of antimicrobial resistance [1]. The use of nanoparticles can improve bioavailability and receptor specificity, thereby reducing the frequency of administration, toxicity, and severe side effects of the drug. While various methods exist for nanoparticle synthesis, the green synthesis approach utilizing plant extracts for bio-reduction of metal compounds into nanomaterials represents an optimal solution. This is because the production of nanoparticles via green synthesis demonstrates high compatibility, lower toxicity, environmental friendliness, and cost efficiency compared to conventional physical and chemical approaches [2].

In recent years, plant-mediated synthesis of AgNPs has gained prominence as a sustainable alternative to traditional methods. *Morus nigra* (black mulberry), a member of the Moraceae family, presents an excellent candidate for green synthesis due to its rich phytochemical profile, including flavonoids, alkaloids, and phenolic compounds [3] that can act as effective reducing and stabilizing agents. These bioactive compounds facilitate the reduction of silver ions to silver nanoparticles while simultaneously providing a protective coating that enhances stability and biocompatibility. Despite the promising potential of *Morus nigra*-mediated synthesis of AgNPs, systematic optimization of synthesis parameters remains a critical

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

challenge. Key factors including incubation time, reaction temperature, and extraction temperature of the plant material significantly influence the formation and stability of the resulting nanoparticles, which directly affects their biological activities [4]. The optimisation of these specific parameters is essential to establish an efficient green synthesis protocol with enhanced antibacterial efficacy. This study aims to optimise three critical parameters (incubation time, reaction temperature, and plant extract preparation temperature) for the green synthesis of silver nanoparticles using *Morus nigra* leaf extract and evaluate their antibacterial properties against *Staphylococcus aureus* and *Klebsiella pneumoniae*. UV-Visible spectroscopy was employed to monitor nanoparticle formation under various synthesis conditions and assess the antibacterial efficacy of the optimized AgNPs. The findings of this research will contribute to the development of an eco-friendly protocol to produce AgNPs with antibacterial properties.

2.0 METHODS AND MATERIAL

Morus nigra leaves (2.0 kg) were obtained from a local supplier in Kuala Selangor, Malaysia. The leaves were thoroughly washed with distilled water, dried at room temperature for three days, followed by additional drying at 40°C for three days. The dried leaves were ground into a fine powder. For extract preparation, 1.25 g of *M. nigra* powder was mixed with 25 mL of autoclaved distilled water. To optimize the extraction process, the mixtures were heated in a water bath at three different temperatures: 40°C for 60 min, 60°C for 45 min, and 90°C for 30 min. After cooling to room temperature, the extracts were filtered through Whatman No. 1 filter paper to obtain clear aqueous extracts, which were stored at 4°C until further use. Silver nitrate solution (2 mM) was prepared by dissolving 0.0085 g of $AgNO_3$ in 25 mL of autoclaved distilled water [5]. The solution was stirred using a magnetic stirrer until completely dissolved, resulting in a clear solution without any visible precipitate. For optimization studies, 3 mL of 2 mM silver nitrate solution was added to separate test tubes labeled according to the extraction temperatures (40°C, 60°C, and 90°C). To each test tube, 100 µL of the corresponding *M. nigra* leaf extract was added and mixed thoroughly. These mixtures were incubated at different temperatures (40°C, 50°C, and 60°C) for varying time periods (2, 24, and 48 hours) to determine the optimal synthesis conditions. The formation of AgNPs was monitored using a JASCO UV-visible spectrophotometer in the wavelength range of 300-500 nm. Based on spectral analysis, the optimal conditions were determined to be 60°C for extract preparation, followed by incubation at 50°C for 24 hours. Following optimisation, a scaled-up synthesis was performed. *M. nigra* leaf powder (2.5 g) was mixed with 50 mL of autoclaved distilled water using a magnetic stirrer. The mixture was heated at 60°C for 45 min, cooled to room temperature, and filtered. For the silver nitrate solution, 0.204 g of $AgNO_3$ was dissolved in 700 mL of autoclaved distilled water with constant stirring.

The synthesis was carried out by adding 20 mL of the optimized *M. nigra* leaf extract to 600 mL of silver nitrate solution. This mixture was incubated at 50°C for 24 hours. The formation of AgNPs was confirmed by UV-visible spectroscopy. The synthesised AgNPs were purified by centrifugation at 40,000 rpm for 25 min, repeated for four cycles. The purified nanoparticles were rinsed to remove excess reagents and subsequently freeze-dried to obtain the final AgNP powder. The antibacterial activity of the synthesised AgNPs was evaluated against four clinical bacteria strains (*Staphylococcus aureus* and *Klebsiella pneumoniae*) in this study. These clinical strains were obtained from the Universiti Kebangsaan Malaysia Medical Centre (UKMMC), Wilayah Kuala Lumpur, Malaysia. Fresh bacterial cultures were prepared by incubating at 37°C for 24 hours. Single colonies were suspended in sterile saline solution, and the turbidity was adjusted to match 0.5 McFarland standard using a Wickerham card for visual comparison. The standardised bacterial suspensions were uniformly swabbed onto Mueller-Hinton agar plates using sterile cotton swabs. Sterile paper discs were impregnated with 20 µL of different test solutions: 2 mM silver nitrate solution, distilled water (negative control), gentamicin (10 µg/disc, positive control), *M. nigra* leaf extract (1.25 g/25 mL), and three different concentrations of synthesized AgNPs (5, 10, and 20 mg/mL prepared through sonication and serial dilution). The impregnated discs were placed onto the inoculated agar plates and incubated at 37°C for 24 hours. After incubation, the diameters of inhibition zones were measured in millimeters using a ruler to assess the antibacterial efficacy of the synthesized AgNPs compared to controls.

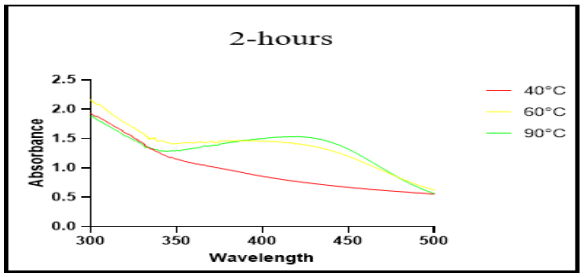
3.0 RESULTS AND DISCUSSION

3.1 Optimisation of Incubation Duration and Temperature on AgNPs Formation

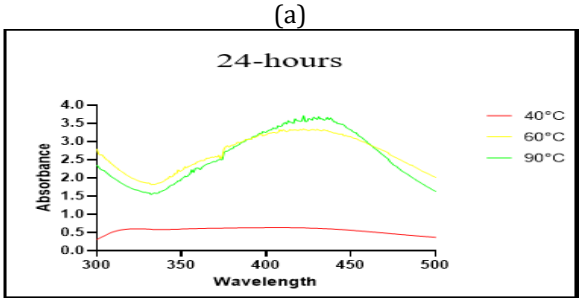
UV-Vis spectrophotometry was employed to confirm the successful synthesis of MN-AgNPs. The surface plasmon resonance (SPR) band, characteristic of AgNPs, was observed within the absorption range of 400–450 nm, aligning with previous studies on the green synthesis of AgNPs [6]. To optimize MN-AgNPs synthesis, key parameters such as temperature of aqueous extraction (which will determine the

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

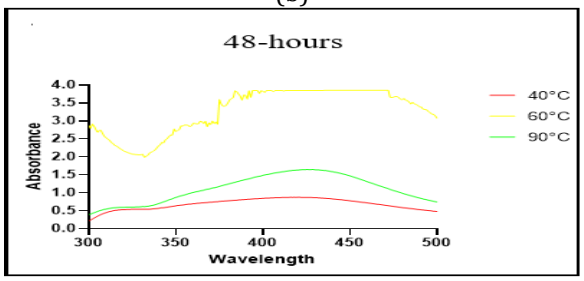
phytochemical compounds extracted), incubation period and temperature were evaluated. Figure 1 illustrate the UV-vis absorption spectra of AgNPs synthesised under different parameters. The UV-visible spectroscopy analysis confirmed the formation of AgNPs through the presence of characteristic surface plasmon resonance (SPR) peaks. The absorption spectra ranged from 400–451 nm, with the highest intensity observed at 423 nm for AgNPs synthesized with extract prepared at 60°C and incubated at 50°C for 24 hours. The sharp and distinct peak at this wavelength is consistent with the presence of well-dispersed AgNPs, as reported in previous literature [7]. Additionally, a single absorption peak suggests the formation of mostly spherical or near-spherical nanoparticles, as anisotropic shapes typically exhibit multiple resonance peaks.



(a)

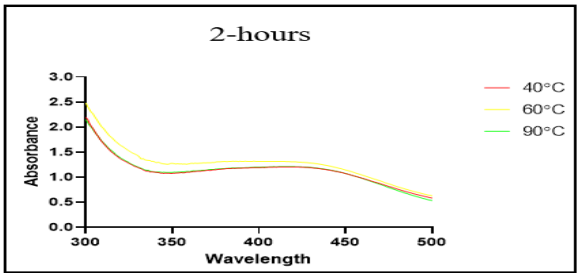


(b)

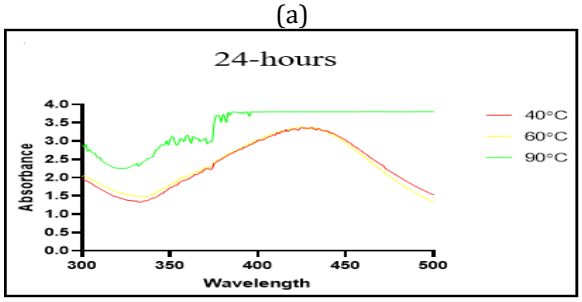


(c)

Figure 1. UV-vis absorption spectra of AgNPs synthesised using *Morus Nigra* extract at different temperatures 40°C for (a) 2 hours, (b) 24 hours, and (c) 48 hours

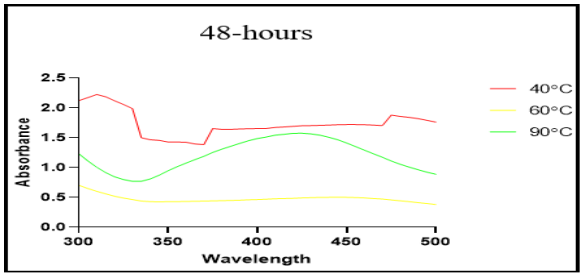


(a)



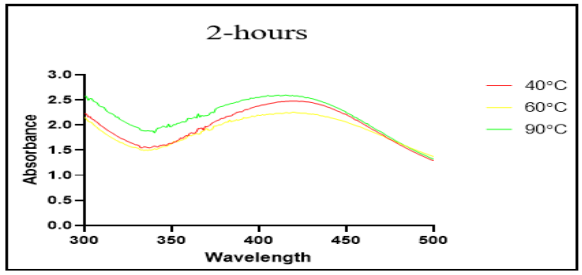
(b)

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

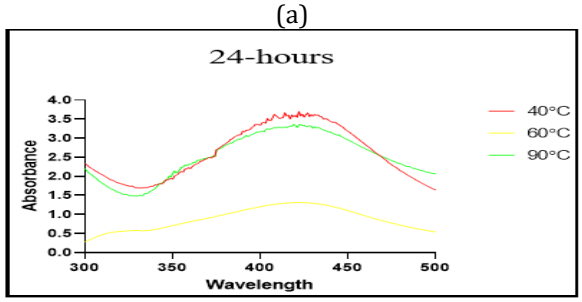


(c)

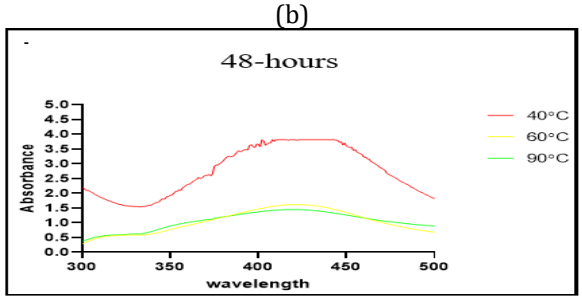
Figure 2. UV-vis absorption spectra of AgNPs synthesised using *Morus Nigra* extract at different temperatures 50°C for (a) 2 hours, (b) 24 hours, and (c) 48 hours



(a)



(b)



(c)

Figure 3. UV-vis absorption spectra of AgNPs synthesised using *Morus Nigra* extract at different temperatures 60°C for (a) 2 hours, (b) 24 hours, and (c) 48 hours

Temperature was another critical factor affecting nanoparticle yield and stability. Although AgNPs synthesised at 90°C exhibited a higher absorption peak (3.802 at 446 nm) compared to those synthesised at 60°C (3.386 at 423 nm), a lower temperature was preferred. This decision was based on the possibility that excessively high temperatures may cause solvent evaporation, degradation of thermolabile bioactive compounds, and potential contamination of the synthesised nanoparticles. Similar observations have been reported in previous studies, where high-temperature synthesis led to unwanted by-products and aggregation [4]. In contrast, samples synthesised at 90°C displayed broader absorption peaks at 446 nm with higher absorbance values (3.802), indicative of potential nanoparticle aggregation. The broader peak at higher temperatures suggests the presence of larger nanoparticles, which is an undesirable outcome as it may reduce antimicrobial efficiency [9]. Furthermore, the effect of the incubation period was assessed, as prolonged incubation can influence nanoparticle size and stability. Results showed that extended incubation (24 hours) led to a decline in absorbance intensity and a redshift in the absorption peak. This phenomenon suggests the aggregation of nanoparticles over time, resulting in broader peaks and a shift toward higher wavelengths, which aligns with previous findings on nanoparticle growth dynamics [10]. Consequently, an incubation period shorter than 24 hours was deemed optimal for MN-AgNPs synthesis.

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

This observation agrees with previous findings where prolonged incubation periods led to a decrease in nanoparticle stability due to secondary interactions [11].

The mass-produced AgNPs using the optimized conditions showed consistent characteristics with the small-scale optimisation batches. The UV-vis spectrum of the mass-produced AgNPs displayed a strong absorption peak at 425 nm with an absorbance value of 3.412 (Figure 4), confirming the successful scale-up synthesis. An extraction temperature of 60°C was chosen over 90°C, despite the latter exhibiting a higher absorption spectrum (3.802 at 446 nm) compared to 60°C (3.386 at 423 nm). The higher absorption spectrum at 90°C may indicate the presence of particles other than silver nanoparticles. This can be explained by the fact that excessive heat may lead to solvent evaporation, resulting in undesirable contamination and the degradation of thermolabile components, such as phenolic compounds [9]. Additionally, the results indicate that an increased incubation period leads to a decrease in both absorbance intensity and wavelength excitation. This phenomenon is likely due to the continued growth of nanoparticle size and potential aggregation, which causes the absorption peak to broaden and shift to a higher wavelength [7].

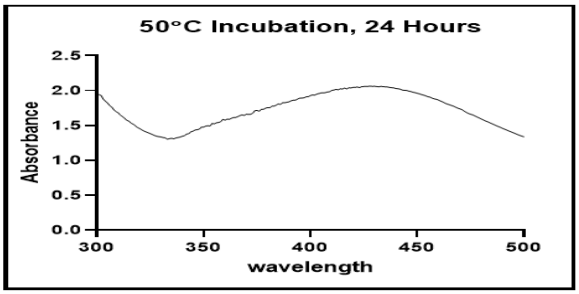
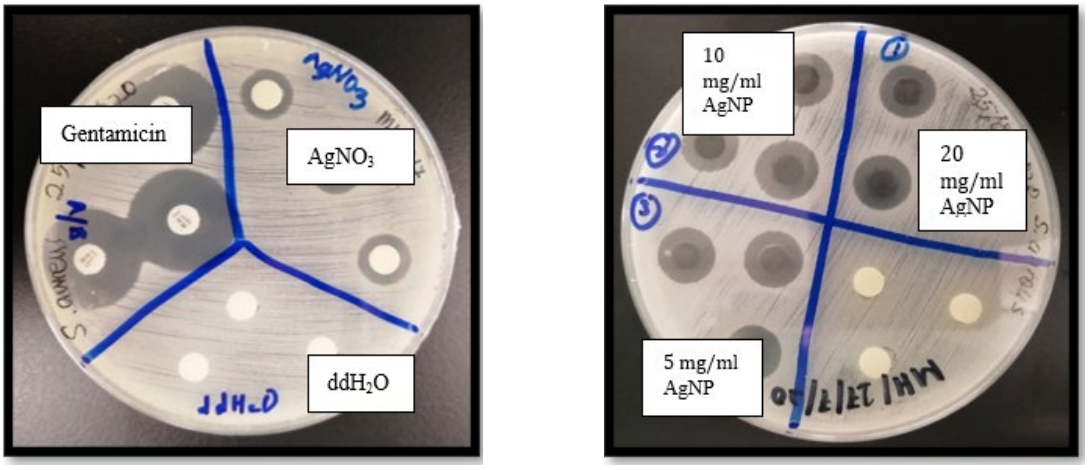


Figure 4. UV-vis absorption spectrum of mass-produced AgNPs synthesized using optimized conditions (60°C boiling temperature, 50°C incubation temperature, 24 hours incubation period)

3.2 Antibacterial activity of *M. nigra* synthesised AgNPs

The antimicrobial activity of biosynthesized AgNPs was evaluated using the disc diffusion method against *Staphylococcus aureus* (gram-positive) and *Klebsiella pneumoniae* (gram-negative). The antimicrobial activity of biosynthesized AgNPs was evaluated against *Staphylococcus aureus* using the disc diffusion method. As shown in Figure 5 and Table 1, all concentrations of AgNPs demonstrated significant antibacterial activity against *S. aureus* compared to both negative and positive controls. Inhibition zones showing antibacterial activity against *S. aureus* after 24-hour incubation at 37°C: (a) AgNPs at different concentrations labelled as 20 mg/ml, 10 mg/ml, and 5 mg/ml, (b) control groups with clear labels for silver nitrate solution, gentamicin (positive control), distilled water (negative control), and plant extract. Data represent mean ± SD of three replicates (n=3). Statistical comparisons performed using independent samples t-test against silver nitrate control. ([a] ; $p < 0.01$; [b] $p < 0.001$). AgNPs, silver nanoparticles. Positive control: gentamicin (30 µg/6 mm disc). Negative control: distilled water. All negative controls showed no inhibition zones.



(a) (b)

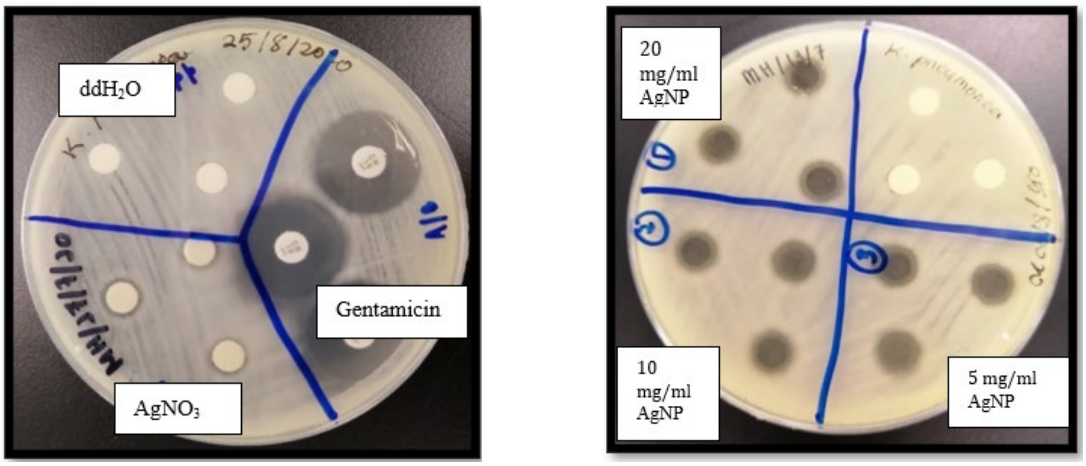
Figure 5. Inhibition zones showing antibacterial activity against *S. aureus* after 24-hour incubation at 37°C: (a) AgNPs at different concentrations labelled, (b) control groups

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

Table 1. Mean inhibition zone diameters (mm) against *S. aureus* for different treatment groups

Treatment	Average diameter of inhibition zone
AgNPs (20 mg/ml)	11.23 ± 0.06[a]
AgNPs (10mg/ml)	11.33 ± 0.06 [a]
AgNPs (5mg/ml)	11.23 ± 0.06 [a]
Silver nitrate	11.0 ± 0.0
M. nigra plant extract	6.0 ± 0.0[b]
Gentamicin (positive control)	20.2 ± 0.0[b]

The antibacterial activity against *Klebsiella pneumoniae* showed that the biosynthesized AgNPs were also effective against gram-negative bacteria, albeit with slightly smaller inhibition zones compared to those observed for *S. aureus* (Figure 6, Table 2). Inhibition zones showing antibacterial activity against *K. pneumoniae* after 24-hour incubation at 37°C: (a) AgNPs at different concentrations clearly labeled as 20 mg/ml, 10 mg/ml, and 5 mg/ml, (b) control groups with clear labels for silver nitrate solution, gentamicin (positive control), distilled water (negative control), and plant extract. Data represent mean ± SD of three replicates (n=3). Statistical comparisons performed using independent samples t-test against silver nitrate control. (ns = not significant; $p > 0.05$; [b] $p < 0.001$). AgNPs at 20 mg/ml showed the highest activity against *K. pneumoniae* (9.00 ± 0.00 mm vs 7.67 ± 1.15 mm for silver nitrate, representing a 17.3% improvement. The results demonstrated that AgNPs exhibited a greater inhibitory effect against *S. aureus* compared to *K. pneumoniae*, as evidenced by larger inhibition zones (Table 2). At all tested concentrations (5 mg/ml, 10 mg/ml, and 20 mg/ml), inhibition zones for *S. aureus* remained consistent at approximately 11.23 mm, whereas inhibition zones for *K. pneumoniae* ranged from 8.7 to 9.0 mm.



(a) (b)

Figure 6. Inhibition zones showing antibacterial activity against *K. pneumoniae* after 24-hour incubation at 37°C: (a) AgNPs at different concentrations, (b) control groups

Table 2. Mean inhibition zone diameters (mm) against *K. pneumoniae* for different treatment groups

Treatment	Average diameter of inhibition zone
AgNPs (20 mg/ml)	9.0 ± 0.0[ns]
AgNPs (10mg/ml)	8.7 ± 0.6 [ns]
AgNPs (5mg/ml)	8.7 ± 0.6 [ns]
Silver nitrate	7.7 ± 1.16
M. nigra plant extract	6.0 ± 0.0
Gentamicin (positive control)	20.13 ± 0.06 [b]

The greater susceptibility of *S. aureus* can be attributed to differences in bacterial cell wall composition. *S. aureus* possesses a thick peptidoglycan layer, which, although structurally rigid, has high affinity for metal nanoparticles, leading to increased nanoparticle adhesion and cellular disruption [12]. In contrast, *K. pneumoniae*, a gram-negative bacterium, has an outer membrane composed of lipopolysaccharides that act as a barrier, limiting AgNP penetration and reducing antimicrobial efficacy [13]. Interestingly, there was no difference in inhibition zones between different AgNP concentrations, suggesting that the antimicrobial effect is not dose-dependent within the tested range. However, it should be noted that formal statistical analysis for dose-response trends (such as regression analysis or trend testing) was not performed, which represents a limitation in interpreting concentration-dependent effects. This observation also highlights the potential limitations of using the disc diffusion method for dose-response evaluations. This implies that

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

once a threshold concentration is reached, increasing the dose does not proportionally enhance bacterial inhibition, though these findings should be interpreted with caution given the inherent limitations of the disc diffusion method, which provides only qualitative to semi-quantitative results and may not accurately reflect the dynamic interactions between nanoparticles and bacterial cells under physiological conditions. Similar findings have been reported in studies where AgNP efficacy plateaued beyond a certain concentration due to nanoparticle aggregation reducing bioavailability [14]. Comparison with control groups further supports the antimicrobial potential of AgNPs. Gentamicin (positive control) exhibited the highest inhibition zones against both bacteria (*S. aureus*: 20.2 mm, *K. pneumoniae*: 20.13 mm), confirming its broad-spectrum activity. Silver nitrate solution (2 mM) also demonstrated antimicrobial activity but with slightly smaller inhibition zones than AgNPs, indicating that the enhanced antibacterial effect is due to the nanoparticulate nature of silver rather than ionic silver alone. Notably, the *Morus nigra* plant extract alone exhibited negligible antibacterial activity (6.0 mm), reinforcing that the antimicrobial effect primarily stems from the AgNPs rather than plant-derived phytochemicals.

A key limitation of this study is the characterization of AgNPs solely through UV-visible spectroscopy. While SPR analysis provides an initial confirmation of nanoparticle synthesis, additional characterisation techniques such as transmission electron microscopy (TEM), X-ray diffraction (XRD), Fourier-transform infrared spectroscopy (FTIR), and dynamic light scattering (DLS) would provide more comprehensive insights into nanoparticle morphology, size distribution, and stability. Future studies should incorporate these techniques to validate nanoparticle formation and investigate their physicochemical properties in greater detail. Furthermore, while this study demonstrated promising antibacterial activity, additional testing against a wider range of bacterial strains, including multi-drug-resistant pathogens, is warranted. It is important to acknowledge the limitations of the disc diffusion method used in this study. The disc diffusion assay provides qualitative and semi-quantitative assessment of antimicrobial activity but does not determine minimum inhibitory concentration (MIC) or minimum bactericidal concentration (MBC) values. Additionally, the method may not fully reflect the complex interactions between nanoparticles and bacterial cells in dynamic physiological conditions. Future studies should incorporate complementary methods such as broth microdilution assays to determine precise MIC values and time-kill studies to evaluate bactericidal kinetics. The relatively small sample size (n=3) per group, while sufficient for initial screening, limits the statistical power to detect smaller differences between treatment groups. Future studies would benefit from larger sample sizes to enhance statistical robustness and detect smaller but potentially relevant differences. Furthermore, dose-response relationships observed in this study were not subjected to formal statistical analysis, which represents an additional limitation that should be addressed in future investigations through appropriate regression analysis or trend testing. In addition, growing evidence suggests that nanoparticles possess broad therapeutic potential, including antioxidant, anti-inflammatory, antimicrobial, and wound-healing properties, further supporting the need for continued investigation into nanoparticle-based biomedical applications [15].

4.0 CONCLUSIONS

This study successfully demonstrated the green synthesis of AgNPs using *Morus nigra* aqueous extract, with optimised conditions identified as 60°C extract preparation temperature and 50°C incubation for 24 hours. The biosynthesized AgNPs exhibited significant antimicrobial activity against both *S. aureus* and *K. pneumoniae*, with significantly higher activity against gram-positive bacteria. While the synthesized AgNPs showed promising antibacterial properties comparable to silver nitrate, further characterization and evaluation against clinically relevant resistant strains would be necessary before considering broader antimicrobial applications. This study provides a foundation for future research exploring the potential of plant-mediated AgNPs in biomedical applications, particularly with enhanced characterization and expanded antimicrobial testing protocols.

5.0 CONFLICT OF INTEREST

The authors declare no conflicts of interest

6.0 AUTHORS CONTRIBUTION

Mohd Afzal, M. (Writing - original draft; Conceptualization; Formal analysis)
Adam, S. H. (Supervision; Methodology; Writing - original draft ;Writing - review & editing)
Shirley Tang, G. H. (Methodology)
Mohd Fathil, N. E. (Methodology)

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my

7.0 ACKNOWLEDGEMENTS

The authors fully acknowledged Ministry of Higher Education (MOHE) and National Defence University of Malaysia (NDUM) which makes this important research viable and effective. We would like to express our sincere gratitude to the Medical Laboratory Technologist, and the staff at International Medical School (IMS), Management & Science University (MSU) for their guidance and support in using the laboratory apparatus and instruments. Their assistance was invaluable throughout this work.

List of Reference

[1] Abdelghany, T. M., Al-Rajhi, A. M. H., Abboud, M. A. A., Alawlaqi, M. M., Magdah, A. G., Helmy, E. A. M., & Mabrouk, A. S. (2017). Recent advances in green synthesis of silver nanoparticles and their applications: About future directions. A review. *BioNanoScience*, *8*(1), 5–16.

[2] Dada, A. O., Inyinbor, A. A., Idu, E. I., Bello, O. M., Oluyori, A. P., Adelani-Akande, T. A., Okunola, A. A., & Dada, O. (2018). Effect of operational parameters, characterization and antibacterial studies of green synthesis of silver nanoparticles using Tithonia diversifolia. *PeerJ*, *6*, e5865.

[3] De Leersnyder, I., De Gelder, L., Van Driessche, I., & Vermeir, P. (2019). Revealing the importance of aging, environment, size and stabilization mechanisms on the stability of metal nanoparticles: A case study for silver nanoparticles in a minimally defined and complex undefined bacterial growth medium. *Nanomaterials*, *9*(12), 1684.

[4] Escárcega-González, C. E., Garza-Cervantes, J. A., Vazquez-Rodríguez, A., Montelongo-Peralta, L. Z., Treviño-Gonzalez, M. T., Castro, E. D. B., Saucedo-Salazar, E. M., Morales, R. M. C., Regalado-Soto, D. I., Treviño-González, F. M., Rosales, J. L. C., Cruz, R. V., & Morones-Ramirez, J. R. (2018). In vivo antimicrobial activity of silver nanoparticles produced via a green chemistry synthesis using *Acacia rigidula* as a reducing and capping agent. *International Journal of Nanomedicine*, *13*, 2349–2363.

[5] 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.

[6] Jain, S., & Mehata, M. S. (2017). Medicinal plant leaf extract and pure flavonoid mediated green synthesis of silver nanoparticles and their enhanced antibacterial property. *Scientific Reports*, *7*(1), Article 15724.

[7] Lim, S. H., & Choi, C. (2019). Pharmacological properties of Morus nigra L. (black mulberry) as a promising nutraceutical resource. *Nutrients*, *11*(2), 437.

[8] Moodley, J. S., Krishna, S. B. N., Pillay, K., Sershen, N., & Govender, P. (2018). Green synthesis of silver nanoparticles from Moringa oleifera leaf extracts and its antimicrobial potential. *Advances in Natural Sciences: Nanoscience and Nanotechnology*, *9*(1), 015011.

[9] Rodrigues, A. S., Batista, J. G. S., Rodrigues, M. Á. V., Thipe, V. C., Minarini, L. A. R., Lopes, P. S., & Lugão, A. B. (2024). Advances in silver nanoparticles: A comprehensive review on their potential as antimicrobial agents and their mechanisms of action elucidated by proteomics. *Frontiers in Microbiology*, *15*, Article 1440065.

[10] Singh, H., Desimone, M. F., Pandya, S., Jasani, S., George, N., Adnan, M., Aldarhami, A., Bazaid, A. S., & Alderhami, S. A. (2023). Revisiting the green synthesis of nanoparticles: Uncovering influences of plant extracts as reducing agents for enhanced synthesis efficiency and its biomedical applications. *International Journal of Nanomedicine*, *18*, 4727–4750.

[11] Tesfaye, M., Gonfa, Y., Tadesse, G., Temesgen, T., & Periyasamy, S. (2023). Green synthesis of silver nanoparticles using Vernonia amygdalina plant extract and its antimicrobial activities. *Heliyon*, *9*(6), e17356.

[12] Wang, L., Wu, Y., Xie, J., Wu, S., & Wu, Z. (2018). Characterization, antioxidant and antimicrobial activities of green synthesized silver nanoparticles from Psidium guajava L. leaf aqueous extracts. *Materials Science and Engineering: C*, *86*, 1–8.

[13] Zhang, Q., Lin, L., & Ye, W. (2018). Techniques for extraction and isolation of natural products: A comprehensive review. *Chinese Medicine*, *13*(1), Article 20.

[14] Ferreira, A. M., Vikulina, A., Loughlin, M., & Volodkin, D. (2023). How similar is the antibacterial activity of silver nanoparticles coated with different capping agents? *RSC Advances, 13*(16), 10542–10555.

[15] Ahamad Tarmizi, A. A., Adam, S. H., Nik Ramli, N. N., Abd Hadi, N. A., Maisarah, A. M., Tang, S. G. H., & Mokhtar, M. H. (2023). The ameliorative effects of selenium nanoparticles (SeNPs) on diabetic rat model: A narrative review. *Sains Malaysiana, 52*(7), 2037–2053

*Corresponding Author | Adam, S. H. | siti.hajar@upnm.edu.my