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GREEN SYNTHESIS OF SILVER NANOPARTICLES (AGNPS) FROM FIVE SPECIES OF *Cyperus* GENUS AND CHARACTERIZATION OF THEIR BIO-FUNCTIONAL AND CATALYTIC PROPERTIES

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Nanotechnology offers a sustainable approach for synthesising materials at the nanoscale, enhancing their physicochemical and biological properties. This study investigated the green synthesis of silver nanoparticles (AgNPs) using aqueous root extracts (AEs) of five *Cyperus* species, namely *C. mindorensis*, *C. esculentus*, *C. alternifolius, C. rotundus*, and *C. eragrostis*. Phytochemical screening revealed the presence of carbohydrates, saponins, proteins, and quinones in all AEs, while tannins and carboxylic acids were absent. SEM analysis confirmed spherical AgNPs of 20–50 nm. Compared to AEs, AgNPs showed significantly higher total flavonoid content (TFC), total phenolic content (TPC), and total antioxidant capacity (TAC). *C. esculentus* AEs showed 746.09 g/QE/100g for TFC, 543.34 g/GAE/100g for TPC, 52.988 g/AAE/100g for TAC, and AgNPs exhibited the highest TFC level (4.2 g/QE/100g), the highest TAC levels (319 g/AAE/100g), and the highest TPC levels (2158 g/GAE/100g). The antibacterial potential of the synthesised AgNPs was evaluated using the well diffusion method with gentamycin serving as positive control, revealing significant inhibitory activity against *E. coli* and *S. aureus*. Notable zones of inhibition were observed: *C. esculentus* – 11 mm, *C. alternifolius* – 11 mm, *C. rotundus* – 10 mm, and *C. eragrostis* – 11 mm. But *C. mindorensis*-AgNPs did not exhibit antibacterial properties. Cytotoxicity tests using *Artemia salina* indicated 100% viability across all AgNP concentrations, confirming biocompatibility. In catalysis studies, AgNPs facilitated the reduction of para-nitrophenol (PNP) to para-aminophenol (PAP), with *C. esculentus*-AgNPs showing the highest rate constant (0.1351 min^{-1}). Degradation of PNP was observed within 20 min at 420 nm, and formation of PAP was observed at 320 nm. Photocatalytic degradation experiments were performed with both methylene blue (MB) and methyl orange (MO). Photocatalytic degradation of MB and MO was faster with 4000ppm AgNPs, while 267ppm AgNPs showed enhanced degradation when combined with $NaBH_4$. 4000ppm showed higher degradation with AgNPs and AgNPs with $NaBH_4$. Bioinformatic docking analysis revealed silver ion binding with key residues (Ser127, Asn124) of the TRAF1 (1KZN) protein (pbd_00001kzn), suggesting potential anticancer applications. These results demonstrate that *Cyperus*-derived AgNPs possess multifunctional properties, highlighting their potential use in biomedical and environmental fields.

Keywords: AgNPs, *Cyperus*, green synthesis, antioxidant, photocatalysis, molecular docking

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INTRODUCTION

Nanotechnology utilises nanoscale materials (1–100 nm) with superior physical, chemical, and biological properties. Green synthesis of nanoparticles, especially using plant extracts, offers a sustainable alternative to conventional chemical methods. Silver is favoured due to its antimicrobial strength, optical properties, and ease of reduction. Roots of *Cyperus spp*. provide a rich source of phytochemicals useful for reducing and stabilising AgNPs (Mirsasaani *et al.,* 2019). The synthesised nanoparticles were characterised and tested for antioxidants, catalytic, cytotoxic, and antimicrobial activity. The study aims to synthesise silver nanoparticles (AgNPs) using aqueous root extracts (AE) of five Cyperus species under varying temperature and time conditions. The extracts will be screened qualitatively for phytochemicals and quantitatively for total flavonoid, phenolic, and antioxidant content. The synthesised AgNPs will be evaluated for catalytic, antimicrobial, cytotoxic (via *Artemia salina* assay), and photocatalytic activities, as well as bioinformatically analysed for potential anticancer effects. This integrated approach will provide insights into the synthesis, properties, and diverse applications of AgNPs.

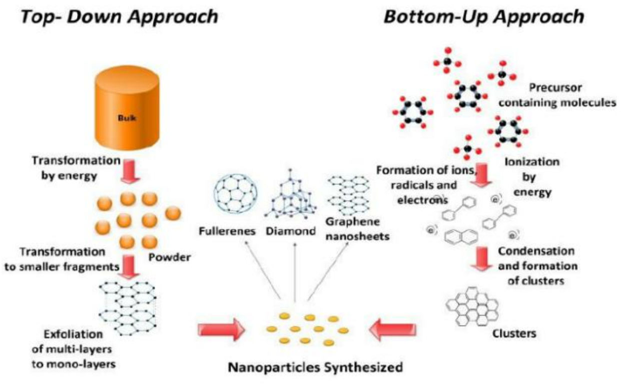


Figure 27: The Top-Down Approach involves breaking down bulk materials into nanoparticles, while the Bottom-Up Approach constructs nanoparticles by combining molecular precursors like atoms or ions (Khaled Habiba *et al*., 2014).

MATERIALS AND METHODS

Roots from five Cyperus species, *C. mindorensis*, *C. esculentus*, *C. alternifolius*, *C. rotundus*, and *C. eragrostis* were collected, shade-dried, and ground before being extracted in distilled water to obtain aqueous root extracts. For the synthesis of

AgNPs, 1 mM $AgNO_3$ solution was mixed with each root extract and incubated at two different temperatures, 60°C and 90°C, for time intervals ranging from 15 to 60 minutes. Successful nanoparticle formation was indicated by a colour change and further confirmed by UV-Visible spectroscopy, which revealed characteristic absorbance peaks.

Phytochemical screening was performed qualitatively to detect the presence of key bioactive compounds such as phenols, flavonoids, proteins, saponins, and others following the method suggested by (Cilesizoglu et al., 2022), (Godlewska et al., 2022), (Kushwaha, 2019), (Rondon et al., 2018). Characterisation of the synthesised AgNPs was carried out using SEM to observe particle morphology, while antioxidant properties were quantified using TFC (Zahidin *et al.,* 2018), TPC (Anokwuru *et al.,* 2011), and TAC assays (Kandiah, Bhaskaran and Perera, 2024).

A series of bioassays was conducted to assess biological activity. Cytotoxicity was evaluated using the *Artemia salina* lethality assay to determine the safety of AgNPs (Kandiah, Bhaskaran and Perera, 2024). Catalytic efficiency was assessed by measuring the degradation of PNP in the presence of $NaBH_4$ (Shimoga *et al.,* 2020). Photocatalytic activity was tested through the degradation of two common textile dyes, MB and MO, under sunlight, both with and without $NaBH_4$ (Elbadawy *et al.,* 2023). Antibacterial properties were assessed using the well diffusion method on Mueller-Hinton agar plates against two bacterial strains, *Escherichia coli* and *Staphylococcus aureus*. Finally, a bioinformatics analysis was performed using molecular docking simulations to investigate the interaction of silver ions with the TRAF1 protein (PDB ID: 1KZN), a cancer-related protein, in order to explore the nanoparticles' potential anticancer activity.

Figure 28: Selected plant species from the Cyperus genus used for the green synthesis of AgNPs. A: *Cyperus mindorensis* (Cy.min), B: *Cyperus esculentus* (Cy.esc), C: *Cyperus alternifolius* (Cy.alt), D: *Cyperus rotundus* (Cy.rot), E: *Cyperus eragrostis* (Cy.era).

RESULTS AND DISCUSSION

Phytochemical analysis of the aqueous root extracts from all five *Cyperus* species revealed the presence of carbohydrates, proteins, saponins, and quinones, while flavonoids and steroids were present selectively among the species. These bioactive compounds played a key role in reducing and stabilising silver ions during nanoparticle synthesis. The formation of AgNPs was visually indicated by a colour change and confirmed by a characteristic surface plasmon resonance peak at 420 nm using UV-Visible spectroscopy. Scanning SEM further validated the synthesis, revealing well-dispersed spherical nanoparticles ranging from 20 to 50 nm. The optimal synthesis conditions were identified as 90°C for 60 minutes.

Antioxidant assays demonstrated that AgNPs exhibited significantly higher TPC, TFC, and TAC compared to their respective plant extracts. This enhancement indicates the successful capping and stabilisation of nanoparticles by phytochemicals during synthesis, contributing to their bioactive potential. In catalytic reduction studies, the AgNPs facilitated the rapid conversion of PNP to PAP in the presence of $NaBH_4$. Among the tested samples, *C. esculentus*-derived AgNPs exhibited the highest rate constant, signifying the most efficient catalytic performance.

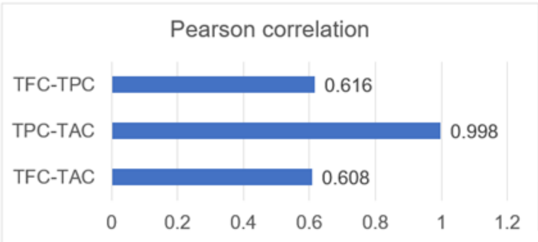
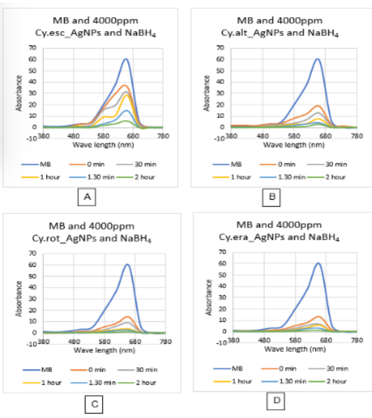


Figure 3. Pearson correlation (TPC, TFC, TAC). TPC has a strongly positive correlation with TAC. All are strongly positive because these are above 0.5.

Photocatalytic degradation experiments further demonstrated that both MB (Figure 4) and MO (Figure 5) dyes were effectively broken down under sunlight. The presence of $NaBH_4$ enhanced the degradation rate, with 4000 ppm AgNP concentrations showing the most significant performance.



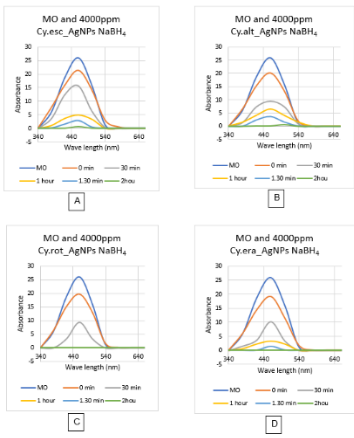


Figure 4. Photocatalytic degradation of methylene blue at 4000 ppm of different AgNPs extracts with $NaBH_4$. A). *C. esculentus* B). *C. alternifolius* C). *C. rotundus* D). *C. eragrostis*

Figure 5. Photocatalytic degradation of methyl orange at 4000 ppm of different AgNPs extracts with $NaBH_4$. A). *C. esculentus* B). *C. alternifolius* C). *C. rotundus* D). *C. eragrostis*.

Antibacterial testing revealed that AgNPs produced larger zones of inhibition compared to crude extracts, with *E. coli* being more susceptible than *S. aureus*, likely due to differences in cell wall structure that affect permeability and oxidative stress response.

Cytotoxicity was assessed using the *Artemia salina* lethality assay, and results showed 100% viability at both 200 ppm and 800 ppm AgNP concentrations. This indicates the non-toxic and environmentally safe nature of the nanoparticles, supporting their potential for biomedical applications. Finally, bioinformatics analysis using molecular docking revealed that silver ions formed stable interactions with the TRAF1 (1KZN) protein, specifically binding to residues Ser127 and Asn124. The observed binding energy of -0.22 kcal/mol suggests a favorable interaction that may trigger reactive oxygen species (ROS) production and apoptosis, highlighting a possible anticancer mechanism of action for Cyperus-derived AgNPs.

This study successfully demonstrated the green synthesis of silver nanoparticles (AgNPs) using aqueous root extracts from five *Cyperus* species, *C. mindorensis*, *C. esculentus*, *C. alternifolius*, *C. rotundus*, and *C. eragrostis*. Among the conditions tested, optimal synthesis was achieved at 90°C for 60 minutes, producing well-dispersed, spherical nanoparticles ranging from 20–50 nm in size. Phytochemical analysis revealed that all extracts were rich in bioactive compounds such as carbohydrates, proteins, saponins, and quinones, which played crucial roles as reducing and stabilising agents in nanoparticle formation.

The synthesised AgNPs exhibited significantly enhanced antioxidant properties, as evidenced by higher TPC, TFC, and TAC compared to the crude plant extracts. This indicates that the bioactive compounds not only mediated nanoparticle synthesis but also contributed to their stability and biological efficacy. The catalytic potential of the AgNPs was evident from their efficient reduction of PNP to PAP, with *C. esculentus*-derived nanoparticles exhibiting the highest reaction rate, thereby highlighting their suitability for detoxifying harmful environmental pollutants.

In photocatalytic assays, AgNPs facilitated the effective degradation of MB and MO, especially at higher concentrations and when combined with sodium borohydride ($NaBH_4$). This demonstrates the potential application of these nanoparticles in wastewater treatment and environmental remediation (Shimoga *et al.,* 2020). Furthermore, the AgNPs displayed strong antibacterial activity against *E. coli* and *S. aureus*, with greater effectiveness against the Gram-negative *E. coli*, indicating their potential use in antimicrobial formulations for healthcare and sanitation (Ismail *et al.,* 2025)

Cytotoxicity assessments using the *Artemia salina* model confirmed that the synthesised AgNPs were non-toxic at both high and low concentrations, reinforcing their biocompatibility and environmental safety (Arshad *et al.,* 2023). In addition, bioinformatic molecular docking studies showed that silver ions could bind favorably to active residues in the TRAF1 protein, suggesting a potential mechanism for anticancer activity via induction of oxidative stress and apoptosis (Lafitte *et al.,* 2002).

CONCLUSION

Overall, this research highlights the feasibility and effectiveness of using *Cyperus* plant root extracts for the green synthesis of multifunctional AgNPs. These nanoparticles exhibit a wide range of valuable properties, including antioxidants, catalytic, photocatalytic, antibacterial, and possibly anticancer activities making them strong candidates for applications in biomedicine, environmental science, and nanotechnology. Future work can further explore their therapeutic potential, scale-up production, and assess long-term impacts in real-world applications.

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