

Republic of Iraq
Ministry of Higher Education
& Scientific Research
AL-Muthanna University
College of Education for pure sciences
Department of Biology



Antibacterial Activity of Silver Nanoparticles Synthesized from *Azadirachta indica* Leaves Against Some Urinary Tract Pathogens

A Thesis Submitted to the Council of college of Education for pure sciences/Al Muthanna University as Partial Fulfillment of the Requirements for the Degree of Master in Education Biology

By:

Hussein Khader Ali

B.Sc. Biology / College of Education /AL-Iraqi University / 2023

Supervisors:

Assist. Prof. Dr. Dhay Ali Azeez

Assist. Prof. Dr. Israa A.A. Hamdan

2026 A.D

1448 A.H

Urinary tract infections (UTIs) are among the most common bacterial infections worldwide, and the increasing antimicrobial resistance of their causative pathogens has highlighted the need for alternative antibacterial approaches. *Azadirachta indica* is a medicinal plant rich in biologically active phytochemicals with recognized antimicrobial properties, making it a promising candidate for green nanotechnology applications. The present study aimed to synthesize silver nanoparticles (AgNPs) using aqueous and ethanolic leaf extracts of *Azadirachta indica* and to evaluate their antibacterial activity against bacterial pathogens associated with UTIs in comparison with the corresponding conventional extracts.

The study was conducted from October 1 to December 30 and included 100 urine samples collected from patients with UTIs. The participants comprised 31 males (31.0%) and 69 females (69.0%), with ages ranging from 5 to 70 years. The highest frequency of infection was recorded among the 20–29-year age group. Bacterial isolation and identification yielded 181 isolates, including *Enterococcus faecalis* 65(35.9%), *Staphylococcus aureus* 53 (29.3%), *Escherichia coli* 38 (21.0%), *Klebsiella pneumoniae* 13(7.2%), and *Pseudomonas aeruginosa* 12 (6.6%). Bacterial identification was confirmed using Hichrom agar UTI, while antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method according to Clinical and Laboratory Standards Institute (CLSI) guidelines.

The antibacterial activity of aqueous and ethanolic neem leaf extracts and their corresponding AgNPs was evaluated at concentrations of 10 mg/mL, 15 mg/mL, 20 mg/mL, 25 mg/mL, and 30 mg/mL using the agar well diffusion method. The synthesized AgNPs were characterized using UV–Visible spectroscopy, FT-IR, SEM, TEM, EDX, and XRD analyses. A characteristic UV–Visible absorption peak at 440–460 nm, together with the observed color change during synthesis, confirmed the formation of AgNPs. Microscopic characterization revealed

Abstract

predominantly spherical and relatively uniform nanoparticles, while EDX confirmed the presence of silver.

The antibacterial activity of the nano-extracts was significantly higher than that of the corresponding conventional extracts ($P = 0.001$). The aqueous conventional extract exhibited limited antibacterial activity, whereas the aqueous nano-extract showed substantially enhanced activity, with maximum inhibition zones of 25mm, 40mm, 30mm, and 30 mm against *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, and *Pseudomonas aeruginosa*, respectively. The conventional ethanolic extract demonstrated limited to moderate activity, while the ethanolic nano-extract exhibited greater antibacterial activity, producing maximum inhibition zones of 40 mm against *Staphylococcus aureus*, 30 mm against *Enterococcus faecalis* and *Escherichia coli*, and 20 mm against *Pseudomonas aeruginosa*.

Overall, the findings demonstrate that green synthesis of AgNPs using *Azadirachta indica* leaf extracts substantially enhances the antibacterial activity of the plant extracts against both Gram-positive and Gram-negative UTI-associated bacteria. The nano-extracts, particularly the ethanolic nano-extract, showed promising antibacterial.