



## Study on Green Zinc Nanoparticles from Plant Extract and Antimicrobial Effects on *Escherichia coli*

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### Abstract

**Background:** Urinary tract infection (UTI) is among the frequently occurring illnesses during pregnancy, and many of its symptomatic cases are caused by asymptomatic bacteria in the urinary tract. Zinc oxide nanoparticles (ZnO-NPs) are emerging antimicrobial agents that have gained considerable attention in the fields of medicine and food.

**Methods:** Synthesis of zinc (Zn) nanoparticles (NPs) was carried out using medicinal plant extract. Different tests were used to confirm the production of NPs, and finally, the antimicrobial property on pathogenic bacteria was determined using the microdilution method.

**Results:** *Escherichia coli* demonstrated the greatest resistance to ampicillin (19.4%), whereas the lowest resistance was recorded for vancomycin (0.9%). Regarding antibiotic susceptibility, gentamicin (22.2%) and imipenem (21.3%) showed the highest effectiveness. The study also determined that the maximum inhibitory concentration (3000 µg/mL) inhibited five strains, while the minimum inhibitory concentration (750 µg/mL) was effective against three strains.

**Conclusions:** The findings indicated that the synthesized ZnO-NPs possess notable antibacterial activity and hold potential for use in hygienic gels aimed at minimizing infections during pregnancy.

**Keywords:** Nanoparticles, Biosynthesis, *Escherichia coli*

### 1. Background

Urinary tract infections (UTIs) are prevalent infectious conditions, disproportionately impacting women, with a recurrence rate of 20 - 30%. Diagnosis involves clinical evaluations and microbiological testing (1, 2).

*Escherichia coli* is the primary bacterium identified in both UTIs and asymptomatic bacteriuria (ASB) during pregnancy, representing over half of reported cases globally (3). Nanotechnology was initially introduced by Richard Feynman in 1959. The extensive use of nano-based techniques over recent decades has created new

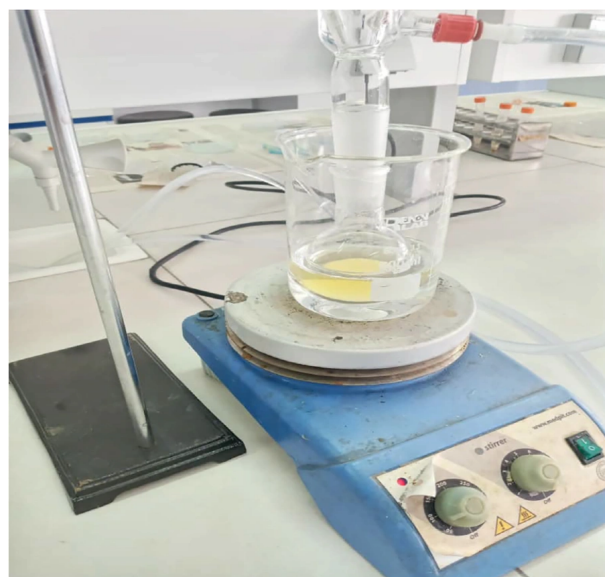
avenues in materials science, even as NPs have significantly transformed their chemical, biological, and physical properties. In addition, it has several pharmaceutical and scientific applications, including the synthesis of NPs for bioimaging, cancer treatment, carbon nanotubes, and drug delivery, as well as antimicrobial purposes (4-6). The advancement of metal oxide and metal NPs has recently captured the interest of the global scientific research community (7, 8).

Zinc oxide nanoparticles (ZnO-NPs) have been extensively developed and utilized across a variety of commercial products and additives (9, 10). Due to its distinctive features and a wide range of applications,

ZnO-NP has been the subject of extensive scientific research (11). Each year, 20,000 people are hospitalized in the UK due to foodborne illnesses (12, 13). The ZnO-NPs interact directly with the cell surface membrane, which subsequently infiltrates the cells of the microorganism and causes oxidative stress (14).

## 2. Methods

A questionnaire was completed on 50 urine samples from patients who had visited the outpatient clinic of Zabol City Hospital. In this study, we conducted the biosynthesis of oxide NPs using a shoot extract of *Ducrosia anethifolia*, a medicinal plant. Zinc (Zn) acetate dihydrate and sodium hydroxide were employed as precursors in the process (15, 16). The structure and morphology of the synthesized NPs were analyzed using several analytical techniques (Figure 1).



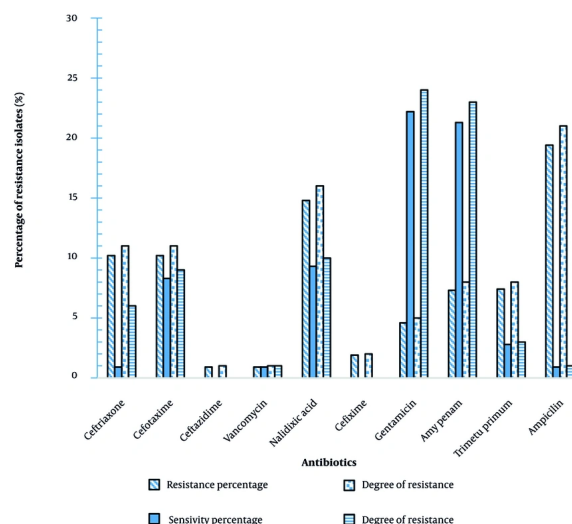
**Figure 1.** Green zinc (Zn) nanoparticles (NPs) synthesis process in *Ducrosia anethifolia*

Determination of MIC and MBC was carried out by assessing bacterial susceptibility to zinc (Zn) using the well-dilution method.

## 3. Results

### 3.1. Antibiotic Resistance Pattern

The highest resistance of *E. coli* was to ampicillin (19.4%) and the lowest resistance was to vancomycin (0.9%). The highest susceptibility to antibiotics was observed with gentamicin (22.2%) and imipenem (21.3%) (Figure 2).

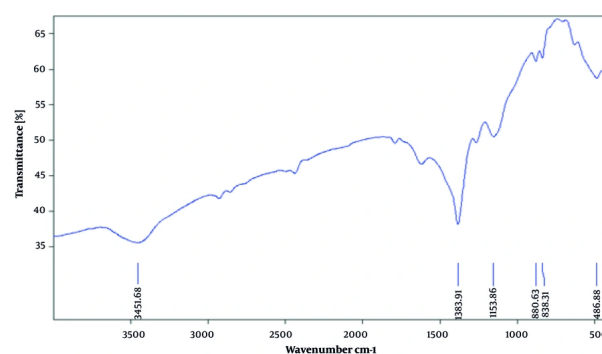


**Figure 2.** Percentage of bacterial resistance to different antibiotics

### 3.2. Fourier Transform Infrared Spectroscopy Results

#### 3.2.1. Analysis of Zinc Nanoparticles Synthesized in *Ducrosia anethifolia* Extract

The presence of a variety of functional groups in the sample is revealed (Figure 3).



**Figure 3.** Fourier transform infrared spectroscopy (FTIR) analysis of *Ducrosia anethifolia* samples

The Fourier transform infrared spectroscopy (FTIR) results obtained from seepage and extracts indicate an absence of clear and distinct structural information about the compounds present. This ambiguity can arise from several factors.

### 3.3. Determination of the MIC and MBC of Synthesized Zinc Nanoparticles

The highest inhibitory concentration was 3000 µg/mL, which inhibited 5 strains. The lowest inhibitory concentration was 750 µg/mL, with 3 strains being inhibited at this level. Additionally, 12 strains showed inhibition at a concentration of 1500 µg/mL.

## 4. Discussion

Bacterial infection during pregnancy has been very common, with the highest mortality rate observed between 2000 and 2008 (17-19). Bacteremia during pregnancy is also life-threatening for fetuses, with a retrospective study indicating a 10% fetal death rate (20). Pathogens can be a concern for expectant mothers. The anatomical and physiological changes that occur in the body during pregnancy can increase women's susceptibility to developing UTIs. *Escherichia coli* is the leading cause of bacteremia worldwide (21), and it is also prevalent during pregnancy, as indicated by Cape et al. (22). The data regarding antibiotic resistance patterns indicated that *E. coli* exhibited the greatest resistance to ampicillin, with a rate of 19.4%. Conversely, the lowest level of resistance in *E. coli* was observed for vancomycin, at 0.9%.

While the highest susceptibility to antibiotics was observed with gentamicin (22.2%) and imipenem (21.3%), it has been indicated (23). This research sought to contrast the antibiotic resistance profiles of *E. coli* found in the urine of generally healthy women who visited the Emergency Department (ED) due to either uncomplicated UTIs or pyelonephritis, with the resistance data documented in the ED's antibiogram (24). The results indicate that 45 patients tested positive for *E. coli*, with pyelonephritis suspected in nine of these subjects, representing 20% of the cases. Compared to the ED antibiogram, resistance rates were notably lower: Two percent for ciprofloxacin (versus 42%,  $P < 0.001$ ), 2% for levofloxacin (versus 26%,  $P < 0.001$ ), and 16% for trimethoprim-sulfamethoxazole (TMP-SMX) (versus 33%,  $P = 0.016$ ). Additionally, six patients were found to have non-*E. coli* uropathogens, and all of these cases were

susceptible to both levofloxacin and TMP-SMX (24). The findings of this research showed that *E. coli* was isolated more frequently from female individuals (70.7%) in comparison to male individuals (29.3%) (25). In urine samples, *E. coli* demonstrated a high degree of susceptibility to ertapenem (97.6%) and imipenem (96.4%). However, these isolates showed a considerable resistance rate of 87.8% to ampicillin (25).

In a study that examined the antibiotic resistance pattern in *E. coli* samples from pregnant women, the results showed that the highest susceptibility was to amikacin, nitrofurantoin, amoxicillin/clavulanic acid, and meropenem, respectively (26). The results of scanning electron microscopy photos showed that the highest inhibitory concentration was 3000 µg/mL, while the lowest inhibitory concentration was 750 µg/mL (27). In a study that biosynthesized NPs from plant extracts, NPs with a diameter of 20 to 30 nanometers were obtained that inhibited *E. coli* and *Klebsiella pneumoniae* bacteria (28). The photocatalytic and antibacterial properties of ZnO-NPs, synthesized via a green approach using extracts of *Lupinus albus* and *L. pilosus*, were examined. Antibacterial experiments demonstrated that the sample created a zone of inhibition against both gram-positive and gram-negative bacteria (29). Another study focused on the synthesis and characterization of ZnO nanomaterials from *Cassia sieberiana*. Synthetic NPs inhibited bacteria *S. typhi*, *S. aureus*, *E. coli*, and *C. albicans* at a concentration of 25 mg/mL (30). Naseer's study explored a green method for synthesizing ZnO-NPs using leaf extracts from *C. fistula* and *Melia zedaran*, assessing their antibacterial potential. The ZnO-NPs mediated by *C. fistula* and *M. zedaran* demonstrated strong antimicrobial activity (31).

### 4.1. Conclusions

The results of the study indicated that synthetic NPs possess a significantly high capability to inhibit bacterial growth and may be applicable in the treatment of female infections.

## Footnotes

**Authors' Contribution:** Study concept and design: T. E. and M. G.; Analysis and interpretation of data: A. M. and A. P.

**Conflict of Interests Statement:** The authors declare no conflict of interest.

**Data Availability:** All data analyzed are publicly available from the relevant study groups.

**Ethical Approval:** The study was approved by the Ethics Committee of Zabol University of Medical Sciences under the number: [IR.ZBMU.REC.1403.084](#).

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