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Green Synthesis of Zinc Oxide Nanoparticles from Fruits and Leaf Extract of *Cordia dichotama*

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Abstract

The study was conducted from February, 2023 to May, 2025 in the Department of Food Safety and Quality Assurance, College of Food Science and Technology, Acharya N. G. Ranga Agricultural University, Pulivendula, with an aim to prepare a uniform, ultrafine ZnO NPs using a process that saves time and energy. The solution combustion synthesis (SCS) approach was adopted for the preparation of zinc oxide (ZnO) nanoparticles using *Cordia dichotoma* L. leaf extract as a fuel and reducing agent, aiming to obtain uniform, ultrafine ZnO nanoparticles through a time- and energy-efficient process. Green synthesis using plant materials, vegetables, enzymes, and microorganisms had gained considerable attention due to its eco-friendly, economical, and simple nature. Among biological methods, plant extract-based synthesis had emerged as a promising alternative. In this work, ZnO nanoparticles were synthesized using both leaf and fruit extracts of *C. dichotoma* through a self-sustaining reaction between zinc acetate and the extracts. The synthesized nanoparticles were characterized using UV-Visible spectroscopy, scanning electron microscopy (SEM), energy-dispersive X-ray (EDX) analysis, and X-ray diffraction (XRD). UV-Vis spectra exhibited absorption peaks between 350–390 nm, confirming nanoparticle formation. XRD revealed a strong peak at 37°, corresponding to the wurtzite structure. SEM analysis showed particle sizes ranging from 3–10 nm, while EDX confirmed high purity. The antioxidant activity of leaf-extract-derived ZnO nanoparticles was higher than that of fruit-extract-derived ones and increased with nanoparticle concentration. Antibacterial activity was more effective against *Escherichia coli* (Gram-negative) than *Staphylococcus aureus* (Gram-positive). Overall, *C. dichotoma*-mediated ZnO nanoparticles exhibited excellent antioxidant and antibacterial potential for biomedical and food safety applications.

Keywords: EDX, SEM, XRD, zinc oxide nanoparticles, *Cordia dichotoma*

1. Introduction

Metal oxides likeZinc Oxide (ZnO) nano/micro-structures have attracted considerable interest in the field of engineering as well as in optical, chemical, mechanical, and biological sciences due to their unique functions and stimulation characteristics (Zhang et al., 2009). Fascinating properties of ZnO due to their high surface area make it a remarkable agent for various applications like electronics (biosensing and bio detection) gas sensors fuel production (Wang et al., 2012), cosmetics (Cross et al., 2007) and environmental protection (Zhou et al., 2006). It's environmental friendly property and biocompatibility makes it desirable especially for biomedical applications and therapeutic intervention, such as drug delivery and nanomedicine (Rasmussen et al., 2010), biological sensing/bioimaging (Xiong, 2013), biological labelling, gene delivery, DNA delivery cell culturing, anti-diabetic activities (Alkaladi et al., 2014), antibacterial defence, antifungal protection and pediculocidal/larvicidal control (Kirthi et al., 2011). Several approaches such as chemical precipitation (Dakhlaoui et al., 2009), sol-gel spin coating (Vafaee and Ghamsari, 2007), spray pyrolysis, thermal decomposition and electrochemical techniques (Pourmortazavi et al., 2015) have been developed to synthesize the nano/micro-structure of ZnO. Recently, owing to the biological importance of the ZnO nano/micro-structure, synthesis of these materials using eco-friendly routes has gained considerable attention. The green synthesis of bioactive ZnO nano/micro-structures from various plant resources and their antibacterial, antifungal and photocatalytic activities have been documented by many researchers (Lakshmeesha et al., 2014, Tripathi et al., 2014, Yuvak Kumar et al., 2014, Kavyashree et al., 2015, Odoom-Wubah et al., 2016). Generally, metal nitrates are preferred over other salts as the oxidizing agent due to their solubility, thus resulting in a highly homogeneous solution. Many studies have been revealed

that carbohydrates, amines, sugar, urea and glycine can be used as a fuel to prepare nanocrystalline ZnO (Dinesha et al., 2009). Recently, the trend of preparing metal/metal oxides NPs using the extracts of various plants has gained popularity. So far, several plants such as, *Pulicaria glutinosa*, *Salvadora persica* L. and so on have been employed for this purpose (Al-Marri et al., 2015). Apart from these, different plants from the genus Cordia such as *Cordia dichotoma* and *Cordia myxa*, which are widely distributed in regions of tropical and subtropical climates such as Central and South America, Asia and Africa, have also been applied for the synthesis of nanoparticles. However, to the best of our knowledge, *Cordia dichotama* L. which is another plant of the genus Cordia, has not been used so far for the preparation of nanoparticles. This plant, belongs to the family Boraginaceae and exhibit excellent biological, and pharmacological properties, The phytochemical studies have revealed the presence of flavonoids, alkaloids, saponins, sterols and terpenes in leaves and fruits of *Cordia dichotama* L. plant. Particularly, the Cordia leaves extract has shown significant antioxidant activity due to the presence of many phenolic derivatives such as chlorogenic and caffeic acid (Bharathi et al., 2018). In this study, SCS approach was adopted for the synthesis of ZnO utilizing *Cordia dichotama* L. leaves extract as fuel (reducing agent) with an aim to prepare a uniform, ultrafine ZnO NPs using a process that saves time and energy. It is assumed that phenolic contents in *C. dichotama* extract may act as a reducing agent during solution combustion synthesis for the production of ZnO. This study placed special emphasized on detailed characterization of *C. dichotama* mediated ZnO structures. The effect of extract concentration on ZnO morphology and sizes was evaluated. Furthermore, potential antimicrobial activity of resultant ZnO nanomaterials were investigated against selected microbial pathogens and antioxidant activity of ZnO nanoparticles were determined.

2. Materials And Methods

The study was conducted from February 2023 to May 2025 in the Department of Food Safety and Quality Assurance, College of Food Science and Technology, Acharya N. G. Ranga Agricultural University, Pulivendula.

2.1. Collection of raw material

Fruits and leaves of *Cordia dichotama* were collected from local fields.

2.2. Preparation of cordia dichotama leaf extract

Cordia dichotama (leaves)

⇩

Oven drying (90°C for 24 hrs)

⇩

Grinding into fine powder

⇩

10 g of dried leaf powder macerated with 100 ml distilled water with occasional stirring

⇩

Filtering through Whatmann no-1 filter paper

⇩

Leaf extract is obtained (Saif et al., 2019)

Figure 1: Preparation of leaf extract of *Cordia dichotama*

2.3 Preparation of Cordia dichotama Fruit Extract

Cordia dichotama(fruits)

⇩

Oven drying (90°C for 24 hrs)

⇩

Grinding into fine powder

⇩

10 g of dried leaf powder macerated with 100ml distilled water with occasional stirring

⇩

Filtering through Whatmann no-1 filter paper

⇩

Fruit extract is obtained (Saif et al., (2019)

Figure 2: *Preparation of* fruit extract of *Cordia dichotama*

2.4. Synthesis of zinc oxide nanoparticles from leaf extract

Leaf extract and zinc acetate (4:1)

⇩

Magnetic stirrer for 6 hrs at 70°C

⇩

Centrifugation at 5000 rpm for 15 min

⇩

Collect the retentate

⇩

Drying for 24 h at 60°C in oven

⇩

Keep it in muffle furnace for 2 h at 400°C (Saif et al., 2019)

Figure 3: ZnO NPs synthesized from leaf extract of *Cordia dichotama*

2.5. Synthesis of zinc oxide nanoparticles from fruit extract

Fruit extract and zinc acetate (4:1)

⇩

Magnetic stirrer for 6 hrs at 70°C

⇩

Centrifugation at 5000 rpm for 15 min

⇩

Collect the retentate

⇩

Drying for 24 h at 60°C in oven

⇩

Keep it in muffle furnace for 2 h at 400°C (Saif et al., (2019)

Figure 4: ZnO NPs synthesized from fruit extract of *Cordia dichotama*

2.6. UV-vis spectroscopy

The optical properties of nanoparticles were investigated by recording UV absorption and transmission spectra. The spectrum of the ZnO NPs was in the range between 300–400 nm (Nilavukkarasi et al., 2020).

Dilute zinc oxide nanoparticles in distill water (0.1 mg ml^{-1})

⇩

Measure absorbance range from 200–780 nm

⇩

Note the readings of absorbance (KavyaSree et al., 2015)

2.7. X-ray diffraction

To examine the structure of zinc oxide nanoparticles, X-ray diffraction spectroscopy was used. In this technique, X-ray radiations were used. Through the cathode ray tube that was present in spectroscope, X-ray radiations were formed. The structure of zinc oxide nanoparticles was evaluated when these radiations were bombarded on the sample. Diffractometer was used to produce the spectrum (Janakiet al., 2015).

2.8. Scanning electron microscope

The scanning electron microscope (SEM) revealed information about the sample including externalmorphology, crystalline structure. Morphology of zinc oxide nanoparticles synthesized from *C. dichotama* was determined by using Scanning Electron Microscope. SEM slides were prepared and thin layer of carbon, potassium and silicon was coated on the smears to make them conductive. Already formed sample of ZnO NPs was observed to determine magnification power and resolution. Then the samples were monitored in SEM (Hasnidawani et al., 2016).

2.9. Energy dispersive X-ray

For chemical characterization (composition and proportion) of zinc oxide nanoparticles, energy dispersive X-ray spectroscopy was used. On the surface of zinc oxide nanoparticles beam of electrons collide (10–20 KeV). After collision of beam of electrons on the surface of biosynthesized zinc oxide nanoparticles, image was formed (Yedurkar et al., 2016).

2.10. Anti-oxidant activity

Preparation of reagent

⇩

(0.588 ml sulphuric acid+0.049 g ammonium molybdate+0.036 g sodium phosphate)

⇩

Make up the volume upto 10 ml with distilled water

⇩

Dissolve 0.01 mg of zinc nanoparticles in 10 ml of Di-methyl sulfoxide (DMSO)

⇩

Water bath at 95°C for 90 min

⇩

Read the absorbance at 695 nm

⇩

The phosphomolybdenumreduction potential (PPR) of the studied extracts were reported (Prieto et al.,1999)

2.11. Anti-bacterial activity

Nutrient agar plates were prepared

⇩

Spread the bacterial culture (gram positive, gram negative) using swab into different Petri plates

⇩

Dissolve (0.001 mg $(10\ ml)^{-1}$, 0.005 mg $(10\ ml)^{-1}$, 0.01 mg $(10\ ml)^{-1}$) concentrations of ZnO NPS in distilled water

⇩

Perform disc diffusion

⇩

Incubate for 24 h at 30°C (Prieto et al.,1999)

3. Results and Discussion

3.1. UV-vis spectroscopy

The formation of zinc oxide nanoparticles (ZnO NPs) synthesized using *Cordia dichotoma* fruit and leaf extracts (Figure 1 to 4) was confirmed by UV-Visible spectroscopy. The absorption spectra of the synthesized ZnO NPs were recorded in the range of 200–800 nm. A characteristic and sharp absorption peak was observed at 370 nm for the fruit extract-mediated ZnO NPs and 368 nm for the leaf extract-mediated ZnO NPs.

These absorption maxima were consistent with the intrinsic band-gap absorption of ZnO nanoparticles due to electron transitions from the valence band to the conduction band. The peak at approximately 370 nm (Figure 5) confirmed the successful formation of ZnO nanoparticles, as also reported in previous studies (Ezealisiji et al., 2018).

The slight blue shift observed in the leaf extract-derived nanoparticles (368 nm) indicated a smaller particle size, which was in accordance with the quantum confinement effect seen in nanoscale semiconductors (Ogunyemi et al., 2019). These results suggested that phytochemicals present in *Cordia dichotoma* not only acted as reducing agents but also

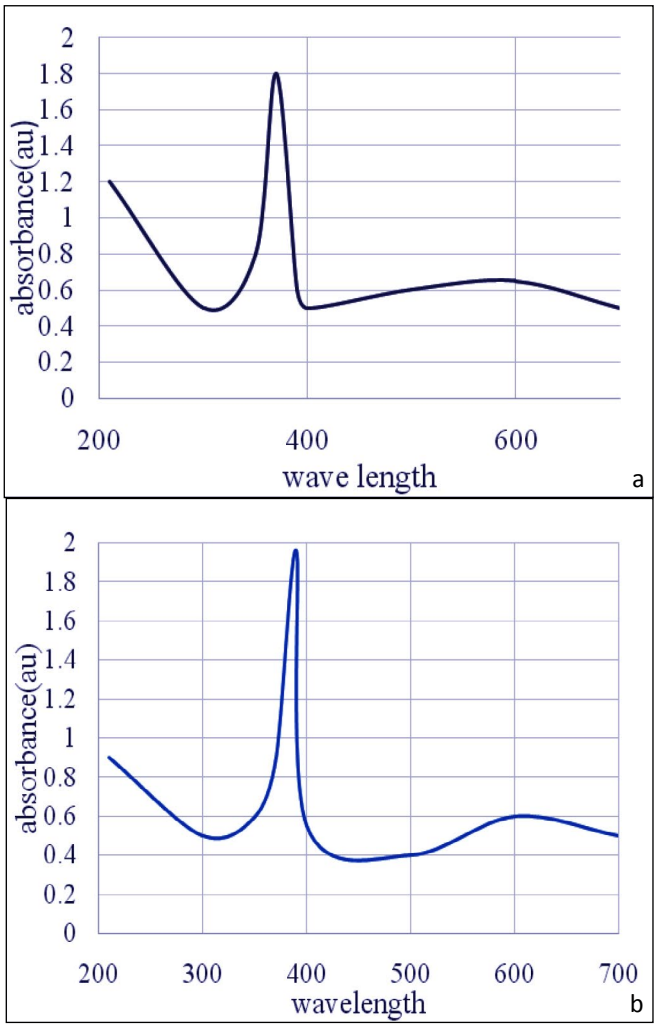

Figure 5: UV-Vis absorbance of ZnO NPs synthesised from *C.dichotama* (a) leaf extract (b) fruit extract

effectively cap and stabilized the nanoparticles, influencing their size and morphology.

3.2. X-ray diffraction

X-Ray Diffraction (XRD) analysis was conducted to confirm the crystalline nature and phase purity of the synthesized zinc oxide nanoparticles (ZnO NPs) obtained from *Cordia dichotoma* fruit and leaf extracts. The diffraction patterns were recorded over a 2θ range of 20° to 80° using Cu-Kα radiation (λ=1.5406 Å).

The XRD patterns of both fruit- and leaf-mediated ZnO nanoparticles exhibited prominent and well-defined peaks at 2θ values of approximately 31.7°, 34.4°, 36.2°, 47.5°, 56.6°, 62.8°, 66.4°, 68.0°, and 69.2°, corresponding to the crystallographic planes (100), (002), (101), (102), (110), (103), (200), (112), and (201) respectively (Table 1). These peaks match the standard data of the hexagonal wurtzite structure of ZnO (JCPDS card no. 36–1451), confirming the formation of crystalline ZnO nanoparticles (Figure 6). The absence of any additional peaks indicates that the synthesized nanoparticles

Table 1: XRD Peak Positions and Assigned Planes for ZnO Nanoparticles

2θ (degrees)	Miller Indices (hkl)	Crystal Phase	Remarks
31.7°	(100)	Hexagonal	Strong intensity peak
34.4°	(002)	Hexagonal	Sharp, indicates high purity
36.2°	(101)	Hexagonal	Dominant peak
47.5°	(102)	Hexagonal	Minor peak

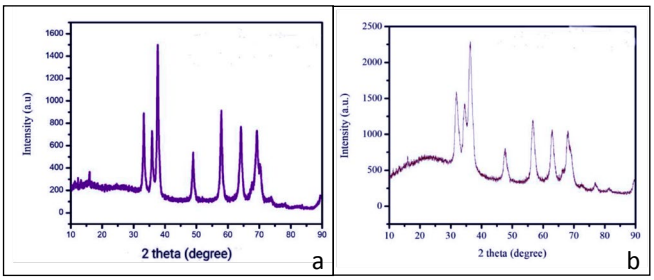

Figure 6: X-ray diffraction spectra of synthesized ZnO NPs from fruits and leaves of *C. dichotama* (a) leaf extract (b) fruit extract

are highly pure and free from impurities or secondary phases. The average crystallite sizes were found to be in the range of 20–35 nm, indicating nanoscale dimensions. The slightly smaller crystallite size of leaf-mediated ZnO NPs compared to fruit-mediated ones might be attributed to stronger phytochemical capping or stabilization effects. These findings were consistent with previous studies reporting crystalline ZnO NPs synthesized via green routes using plant extracts (Ezealisiji et al., 2018; Ogunyemi et al., 2019).

3.3. Scanning electron microscope

Scanning Electron Microscopy (SEM) was used to investigate the surface morphology and size distribution of zinc oxide nanoparticles synthesized using *Cordia dichotoma* fruit and leaf extracts. The SEM micrographs revealed that the ZnO nanoparticles were predominantly spherical to hexagonal in shape with slight agglomeration, which was commonly observed in nanoparticles synthesized through green methods due to the presence of plant biomolecules acting as capping agents. The particle sizes were found to range approximately between 20 nm to 60 nm (Figure 7), with some variation depending on the plant part used. The fruit extract-mediated ZnO nanoparticles exhibited relatively larger agglomerates, possibly due to the presence of higher sugar and polysaccharide content, which promoted particle clustering. In contrast, the leaf extract-mediated nanoparticles were more uniformly distributed, possibly due to better capping and stabilizing efficiency of polyphenols and flavonoids presented in the leaves. No sharp edges or irregular surfaces were observed, suggesting that the synthesis method provided smooth and homogeneously coated surfaces. These findings aligned with

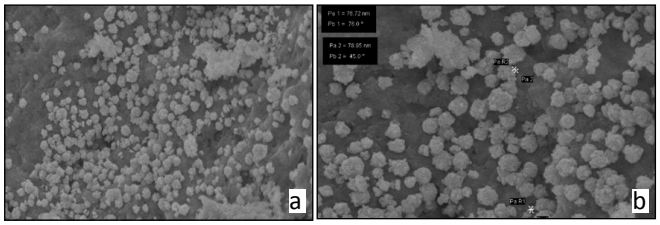

Figure 7: SEM images of ZnO NPs synthesized from fruits and leaves of C.dichotoma (a) leaf extract (b) fruit extract

previous SEM-based studies on green-synthesized ZnO NPs (Ramesh et al., 2016; Abomuti et al., 2021). Overall, SEM analysis confirmed that the green synthesis method using *Cordiadichotoma* was effective in producing nanoparticles of nanoscale dimension and controlled morphology, making them suitable for applications in food packaging, antimicrobial coatings, and biosensors.

3.4. Energy dispersive x-ray

Energy-Dispersive X-ray Spectroscopy (EDX) was employed to confirm the elemental composition of the zinc oxide nanoparticles (ZnO NPs) synthesized using *Cordia dichotoma* fruit and leaf extracts. The EDX spectrum showed strong signals for zinc (Zn) and oxygen (O), confirming the successful formation of ZnO nanoparticles (Figure 8). The major peaks observed were at energies around 1.0 keV corresponding to the Kα transitions of oxygen and Lα transitions of zinc, respectively. The absence of any significant peaks from other elements suggested a high degree of purity and no major contamination in the synthesized nanoparticles. Minor signals of carbon (C) were also detected, which might be attributed to phytochemical residues from the plant extract used during the synthesis process. These organic residues were known to act as capping and stabilizing agents, helping to reduce particle aggregation and improved colloidal stability (Ezealisiji et al., 2018; Ramesh et al., 2016). Quantitative EDX analysis indicated the atomic percentages of Zn and O to be approximately 48–55% for Zn and 20–25% for O (Table 2), with slight variations between fruit- and leaf-mediated samples. These results confirmed that the synthesized materials were indeed stoichiometrically consistent with ZnO.

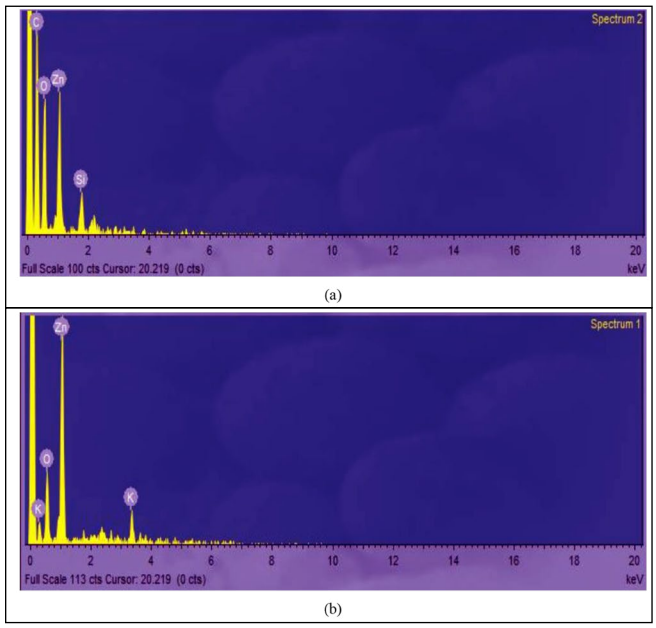

Figure 8: EDX spectrograph of synthesized ZnO NPs from fruit and leaf extract of *C. dichotama* (a) leaf extract (b) fruit extract

Table 2: Elemental composition of ZnO nanoparticles

Element	Weight %	Atomic %
O	23.09	39.42
Zn	53.18	69.93

3.5. Anti-oxidant activity

Total antioxidant capacity assay was a spectroscopic method for the quantitative determination of antioxidant capacity, through the formation of phosphomolybdenum complex. The assay was based on the reduction of Mo (VI) to Mo (V) by the sample analyte and subsequent formation of a green phosphate Mo (V) complex at acidic pH. Total antioxidant capacity could be calculated by this method described by Prieto et al. (1999). Leaf and fruit extract samples antioxidant activity was analyzed using this method at different concentrations. ZnONPs synthesized from leaf extracts have more antioxidant activity than synthesized from fruit extracts. By increasing the concentrations of samples both the samples antioxidant activity also increased (Figure 9). The higher antioxidant activity in leaf-mediated ZnO NPs might be attributed to the presence of abundant phytochemicals such as flavonoids, tannins, and phenolic compounds, which were known for their radical-scavenging capabilities and might remain partially bound to the nanoparticle surfaces, enhancing their bioactivity (Ogunyemi et al., 2019). These findings were consistent with earlier studies, which reported that ZnO nanoparticles synthesized using plant extracts exhibited notable antioxidant activity due to synergistic effects between the metal oxide core and phytochemical capping agents.

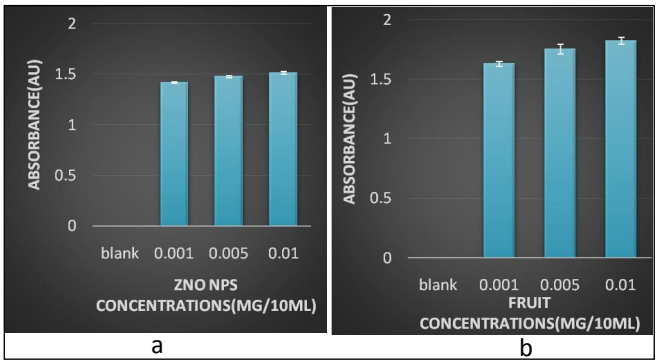

Figure 9: Antioxidant activity of ZnO NPs from *C.dichotama* (a) leaf extract (b) fruit extract

3.6. Anti-bacterial activity

In this study, the bactericidal activity of crystalline ZnO microstructures synthesized using *Cordia dichotoma* was

evaluated against both Gram-positive (*Staphylococcus aureus*) and Gram-negative (*Escherichia coli*) bacteria using the disc diffusion method. The antibacterial activity of the prepared ZnO samples was tested in a dose-dependent manner, and the inhibition zone values obtained from the assay were presented in Tables 2 and 3. All synthesized ZnO samples demonstrated significant growth inhibition against both Gram-positive and Gram-negative bacterial strains.

Table 3: Antibacterial activity of ZnO NPs synthesized from *C. dichotama* leaf extract

Zone of inhibition in mm	ZnO NPs leaf extract (mg 10 ml^{-1})	
	E. coli	*S. aureus*
Control	1.7	0.7
0.001	2.1	1.1
0.005	3.8	2.5
0.01	4.5	2.9

The antibacterial activity of ZnO nanoparticles synthesized from leaf extract was found to be greater than that of nanoparticles obtained from fruit extract. For both leaf and fruit samples, antibacterial activity increased with increasing concentrations of ZnO nanoparticles. Additionally, both samples exhibited stronger antibacterial effects against *Escherichia coli* compared to *Staphylococcus aureus*, indicating that Gram-negative bacteria were more susceptible to ZnO nanoparticles than Gram-positive bacteria. These observations were consistent with previous studies, which reported similar trends in bacterial susceptibility. The variation in antibacterial activity between Gram-positive and Gram-negative bacteria was attributed to differences in cell wall composition and membrane structure (Yuvak Kumar et al., 2014).

It was further observed that higher concentrations of ZnO nanoparticles led to increased bacterial growth inhibition in both Gram-positive and Gram-negative strains. Dutta et al. (2013) reported effective inhibition of *E. coli* using ZnO at concentrations as high as 175 mg l^{-1}. This enhanced antibacterial effect was believed to be associated with the increased generation of reactive oxygen species (ROS) at higher ZnO concentrations, which induced lipid peroxidation in bacterial membranes. The damage to bacterial cell membranes likely resulted in leakage of intracellular contents, ultimately leading to cell death. Moreover, the release of free zinc ions from ZnO nanoparticles has been implicated in contributing to bacterial toxicity (Li et al., 2009).

4. Conclusion

ZnO nanoparticles were effectively synthesized using fruit and leaf extracts of *Cordia dichotoma* through a green synthesis approach. Characterization confirmed wurtzite-structured nanoparticles (3–10 nm) with high purity. Leaf-extract-derived ZnO NPs exhibited superior antioxidant and antibacterial activities compared to fruit-extract-derived ones, with effectiveness increasing at higher concentrations. The nanoparticles were more active against *E. coli* than *S. aureus*. Overall, *Cordia dichotoma*-mediated ZnO NPs demonstrate strong bioactive potential for biomedical and environmental applications.

5. Acknowledgement

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