Uttar Pradesh Journal of Zoology

Volume 45, Issue 12, Page 179-188, 2024; Article no.UPJOZ.3567
ISSN: 0256-971X (P)

Momordica charantia Leaf Extract Mediated Zinc Oxide Nanoparticles for Antibacterial Performance

P. Subramanian [a] and S. Alwin David [b*]

[a] Department of Educational Planning and Administration, Tamil Nadu Teachers Education University, Chennai – 97, Tamil Nadu, India.
[b] PG & Research Department of Chemistry, V. O. Chidambaram College, Tuticorin, India.

Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

DOI: https://doi.org/10.56557/upjoz/2024/v45i124116

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://prh.mbimph.com/review-history/3567

Original Research Article

Received: 17/03/2024
Accepted: 19/05/2024
Published: 23/05/2024

ABSTRACT

We uncovered a simple and conservational process for the production of zinc oxide nanoparticles (ZnO NPs). This was accomplished by a biosynthetic method that included the leaf extract of *Momordica charantia* as a reducing agent. The UV-Vis absorption spectrum of them exhibits maxima at 352 nm. The presence of bands at 630cm^{-1} in the Fourier transform infrared spectrum verifies the production of zinc oxide nanoparticles. XRD determined that the particle size ranged between 20 and 58nm. Field emission scanning electron microscopy exposed that the ZnO NPs were irregular in nature and ranged in size from 20 to 60nm. The existence of oxygen and zinc in ZnO NPs is verified by energy-dispersive X-ray analysis. ZnO NPs exhibited strong antibacterial action against *Klebsiella pneumoniae* and *Staphylococcus aureus*. This biosynthetic technique could pave the way for simple, cost-effective, environmentally friendly products that avoid toxic chemicals and are helpful for applications in medication and the large-scale manufacture of nanoparticles.

**Corresponding author: Email: alwindavid1986@gmail.com;*

Cite as: *Subramanian, P., & David, S. A. (2024). Momordica charantia Leaf Extract Mediated Zinc Oxide Nanoparticles for Antibacterial Performance. UTTAR PRADESH JOURNAL OF ZOOLOGY, 45(12), 179–188. https://doi.org/10.56557/upjoz/2024/v45i124116*

Keywords: *Biosynthesis; ecofriendly; bitter gourd; Klebsiella pneumoniae; Staphylococcus aureus; plant extract; FESEM.*

1. INTRODUCTION

Momordica charantia is usually known as a bitter gourd because of its bitter taste. *Momordica charantia* is generally grown in Asian countries and African countries for its exceedingly bitter and wholesome fruit. Momordica means "to bite," referring to the jagged margins of the leaves that look to have been bitten. It has been scientifically proven to be anticancer, antileukemic, antibacterial, antitumorous, antiviral, antiprotozoal, antiparasitic, antifungal, anti-obesity, hypoglycemic, anti-ulcer, and immunologically stimulating.

Momordica charantia has been used by natural medical professionals for the treatment of high blood sugar, cancer, cholesterol problems, bacterial infections, and viral septicemia. Alkaloids, proteins, steroids, and triterpenes are the primary ingredients of *Momordica charantia,* which are accountable for its beneficial activities [1-9]. These qualities persuaded researchers to use it in the production of ZnO NPs.

ZnO is one of the most extensively utilized n-type semiconductors owing to its uses in light-emitting diodes (LED), photodiodes, solar cells, memory devices, piezoelectric transducers, light emitting diodes, photodetectors, sensors, anticancer therapeutic agents, photocatalysis, and industrial wastewater treatment [10-16].

Several ways have established that ZnO NPs may be produced using chemical and physical methods; however, because of the use of a large amount of expensive reagents, hazardous chemicals, prolonged time, and high temperature conditions, an alternate method is required. The green chemistry approach highlights that the use of natural plants has provided a simple, dependable, nontoxic, and environmentally beneficial solution.

Plant extracts *Myristica fragran* (fruit), Aloe vera (leaf), Cassava starch (fruit), *Cassia fistula*(leaf), *Cocos nucifera* (leaf), *Elaeagnus angustifolia* L (leaf), *Hibiscus subdariffa* (leaf), *Coriandrum sativum* (leaf), *Allium sativum* (bulb), *Zingiber officinale* (root), *Raphanus sativus var. Longipinnatus* (leaf), *Passiflora caerulea* (leaf), *Phoenix dactylifera L* (leaf), *Azadirachta indica* (leaf), *Calotropis gigantea* (leaf), have been utilized to reduce Zn^{2+} during the synthesis of ZnO NPs [17-35].

The research on the plant-mediated production of ZnO NPs utilizing leaf extract of *Momordica charantia* as a reductant is presented here, and they were characterized using UV-Vis diffuse reflectance spectroscopy, X-ray diffraction analysis, Fourier transform infrared spectroscopy, Field emission scanning electron microscopy, and energy-dispersive X-ray analysis. This investigation also includes evaluating the antibacterial efficacy of ZnO NPs against *Klebsiella pneumoniae* and *Staphylococcus aureus* bacteria.

2. MATERIALS AND METHODS

2.1 Chemicals Used

For the synthesis of ZnONPs, zinc sulphate heptahydrate ($ZnSO_4.7H_2O$) was acquired from Nice Chemicals. *Momordica charantia* leaves were obtained locally in Thoothukudi, Tamil Nadu, India, and utilized in this research.

2.2 Preparation of *Momordica charantia* Leaf Extract

In order to eliminate dust particles, about 20g of fresh *Momordica charantia* leaves were picked and suitably rinsed with water. The freshly rinsed leaves were finely chopped and steeped in 200 mL distilled water in a RB flask with a condenser for a whole 30-minute boil. By filtering the leaf extract through Whatman filter paper, purified leaf extract was obtained.

2.3 Green Synthesis of Zinc Oxide Nanoparticles

A solution of 0.1M $ZnSO_4.7H_2O$ was mixed with 125 mL of freshly produced *Momordica charantia* leaf extract. In a RB flask with a condenser, the above mixture was heated to 110°C for one hour. Subsequently, the ZnO NPs that were synthesized underwent filtration and oven drying at 80°C.

2.4 Antimicrobial Study

The microbicidal activity of the synthesized nanoparticles against bacteria was examined by the agar - well diffusion method. Nutrient broth medium was used to subculture the microorganisms, and the resulting homogenous microbial growth plate was created by spreading the medium out on Muller Hinton Agar plates and incubating them for 1 day at 37°C. The strains of bacteria were *Klebsiella pneumoniae* and *Staphylococcus aureus*.

For the purpose of comparing the antibacterial activity of the antibiotic and nanoparticles, chloramphenicol was used as the positive control and dimethyl sulfoxide (DMSO) as the negative control. Finally, a full day of incubation at 37°C was spent on the petri plates. The zone of inhibition (ZOI) diameter in millimeters was measured in order to assess the antibacterial performance of the synthesized nanoparticles. The formula below was used to obtain the inhibition percentage.

$$\text{Percent inhibition} = \left(\frac{\text{Zone of inhibition of test sample (mm)}}{\text{Zone of inhibition of standard drug (mm)}}\right) \times 100$$

2.5 Characterization

The UV-visible spectrum of the ZnO NPs was noted by the JascoV-600 spectrophotometer. FTIR was measured using a FTIR spectrometer with the model name Thermo Scientific Nicolet iS5. The average particle size of the nanoparticles was determined using Cu K radiation and an XPERT-PRO X-ray diffractometer (Philips - CM200). Energy dispersive X-ray analysis and field emission scanning electron microscopy were taken using a TESCAN MIRA3 XMU device.

3. RESULTS AND DISCUSSION

3.1 UV-Vis Diffuse Reflectance Spectroscopy

One of the best techniques for examining metal oxide nanoparticles is UV-visible spectroscopy, which may be used to determine their optical properties. The green - produced ZnO NPs' UV-visible spectrum is displayed in Fig. 1. A distinctive ZnO absorption peak can be seen in the spectra at a wavelength of 352 nm.

3.2 FTIR Analysis

The FTIR spectrum of ZnO NPs is shown in Fig. 2. Major peaks in the FTIR spectra of ZnO NPs may be found at 630, 751, 1097, 1400, 1635, and 3240cm^{-1}. The spectrum clearly displays bands at roughly 1635 and 3240 cm^{-1} for asymmetric stretching vibration of C-O and the stretching vibration of the OH group, respectively. The C-H stretching vibration may be accountable for the band at 1400 cm^{-1}. The band around 1097 cm^{-1} is typical of aromatic amine C-C stretching vibrations. The 751cm^{-1} band corresponds to C-N stretching amine. The band around 630 cm^{-1} corresponds to the Zn-O bond stretching vibration and indicates ZnO NPs formation.

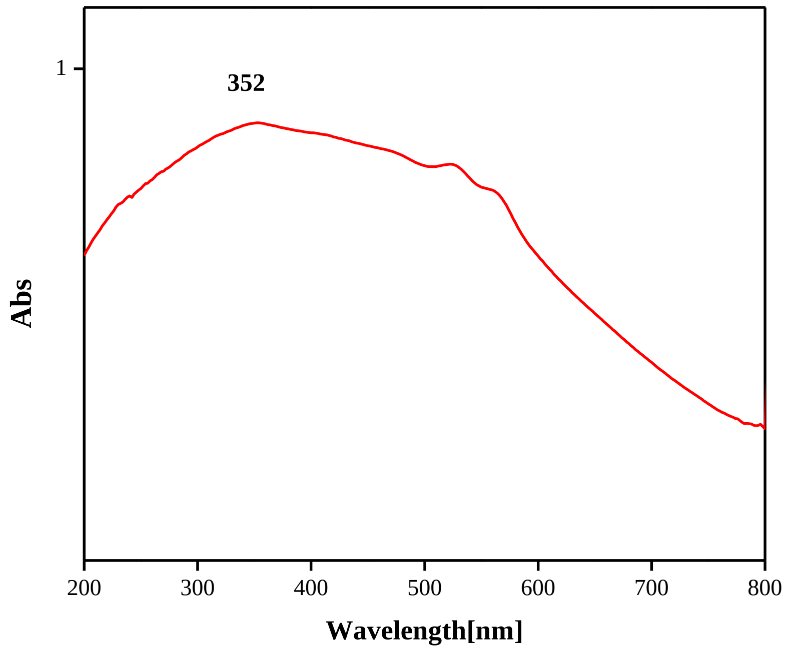


Fig. 1. UV-Visible diffuse reflectance spectrum of ZnO NPs

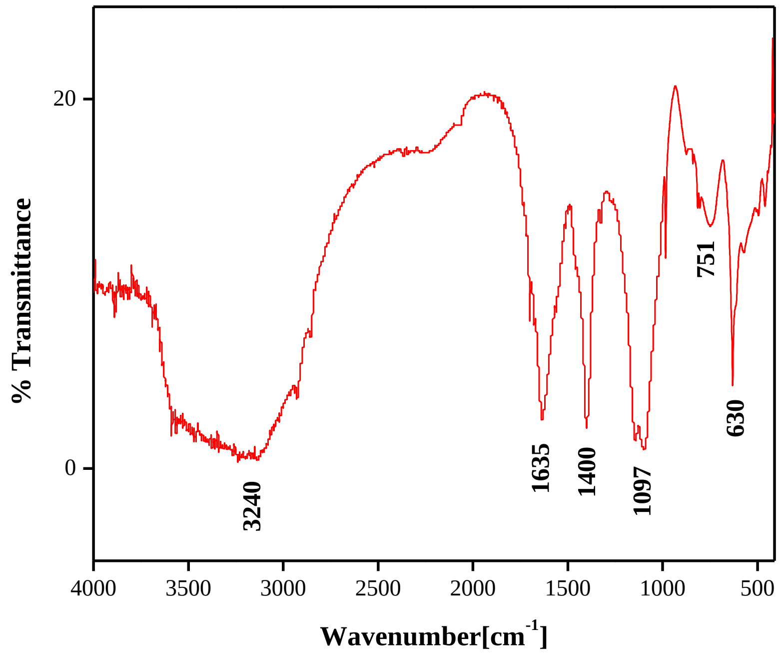


Fig. 2. FTIR spectrum of ZnO NPs

3.3 X-ray Diffraction Analysis

The Debye-Scherer formula, used to analyze XRD data, yields an average crystallite size of 33.81 nm. The Scherrer equation estimates the crystallite size of the ZnO NPs to be between 20 and 58 nm.

The XRD pattern of ZnO NPs is displayed in Fig. 3. The peaks of XRD at 2θ values of 31..84°, 34.52°, 36.28°, 47.36°, 56.48°, 62.88° , 66.44°, 67.76°, 69.08° , 72.28° and 77.08° can be ascribed to the (100), (002), (101), (102), (110), (103), (200), (112), (201), (004) and (202) planes of ZnO nanoparticles, respectively, which are

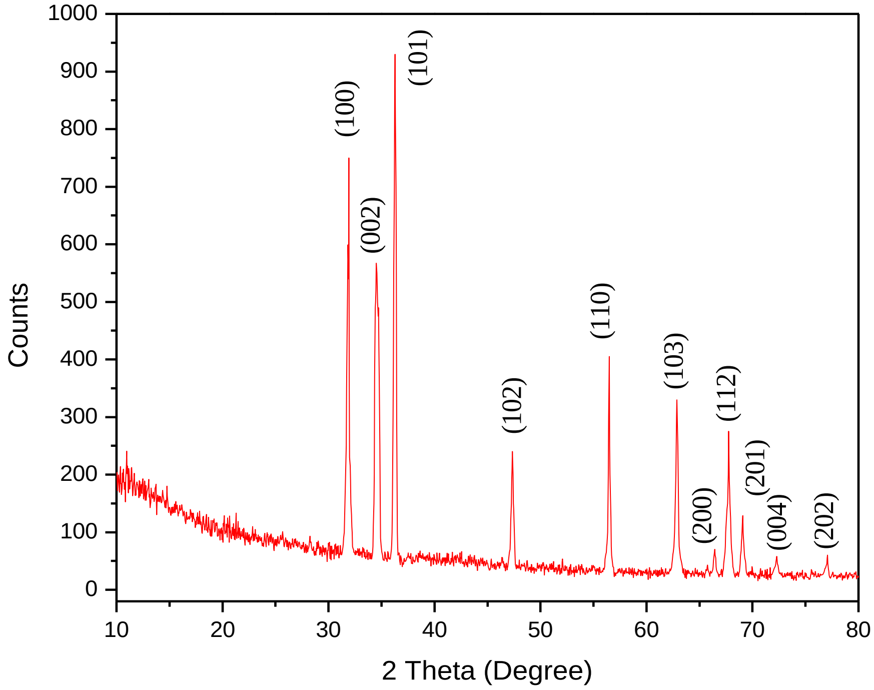


Fig. 3. XRD pattern of ZnO NPs

coordinated with JCPDS No. 36-1451. XRD pattern (Fig. 3) thus evidently elucidates the production of ZnO NPs.

3.4 Field Emission Scanning Electron Microscopy (FESEM)

The surface morphology and the approximate nanoparticle's size are revealed by the FESEM. The FESEM images (Figs. 4–6) demonstrate that the shape of the ZnO NPs is uneven. The size of the ZnO NPs produced by FESEM ranges between 20 and 60 nm.

3.5 Energy Dispersive X - ray Analysis (EDAX)

The EDAX spectrum of ZnO NPs is provided in Fig. 7. The atomic contributions of the components in the sample were calculated using energy dispersive X-ray analysis. Zn and O had weight percentages of 64.47 and 14.31 in the EDAX of ZnO NPs, respectively. As given in Table 1, the atomic (%) values for Zn and O are 52.43 and 47.57, respectively.

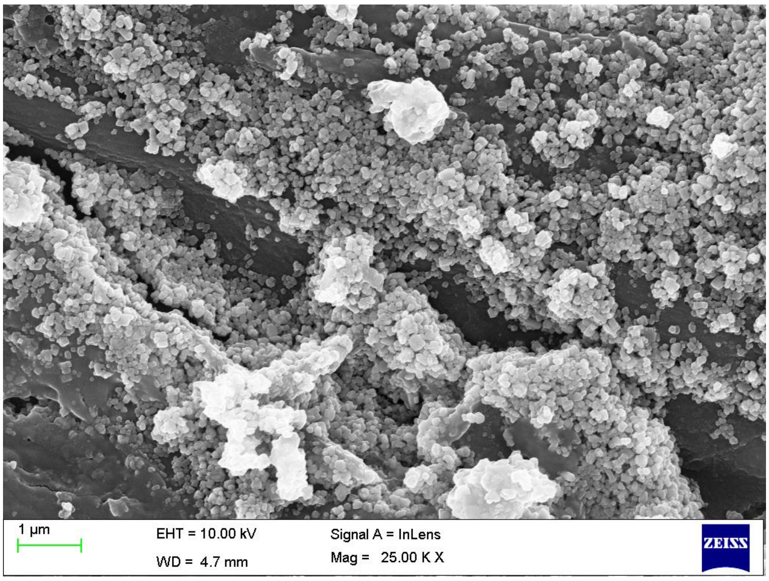


Fig. 4. FESEM image of ZnO NPs (1 µm scale)

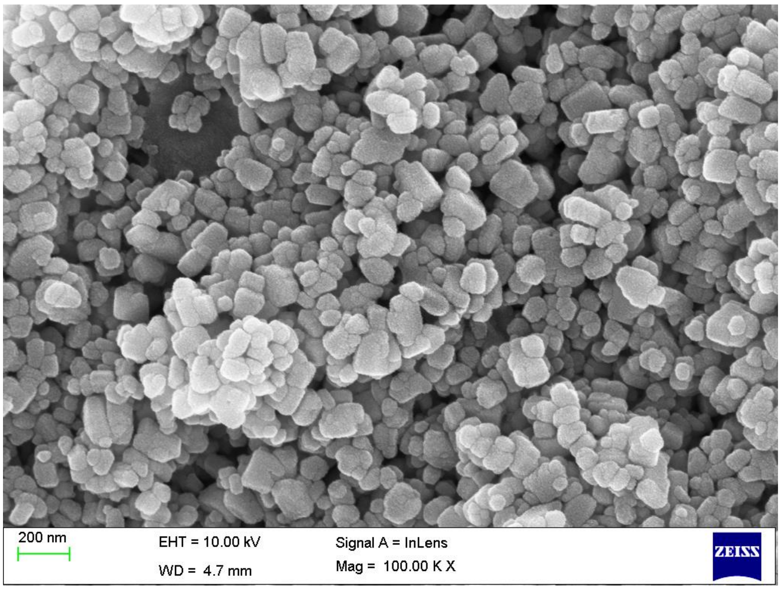


Fig. 5. FESEM image of ZnO NPs (200 nm scale)

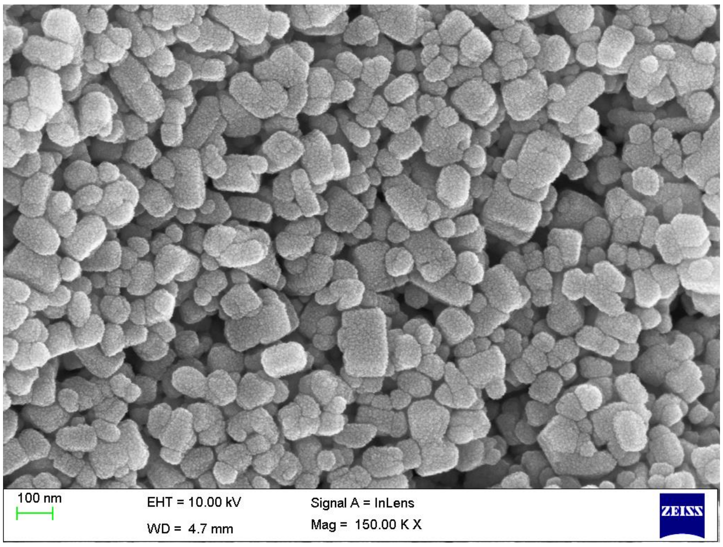


Fig. 6. FESEM image of ZnO NPs (100 nm scale)

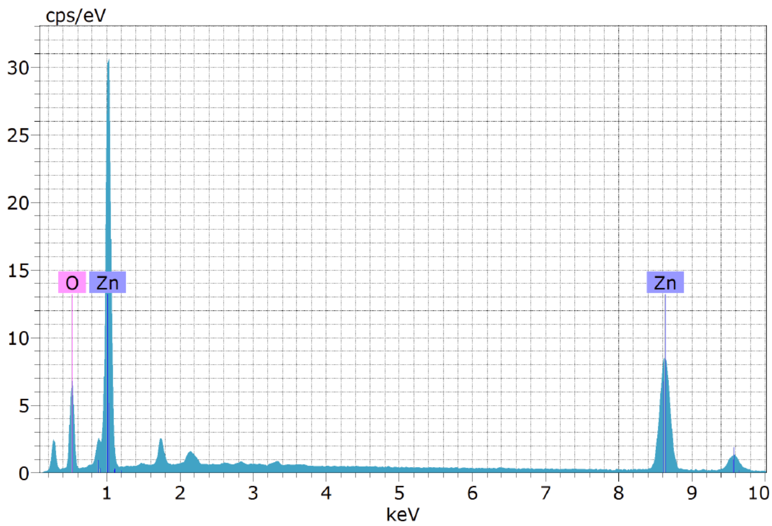


Fig. 7. EDAX spectrum of ZnO NPs

Table 1. EDAX data of ZnO NPs

Element	Weight %	Atomic %
Zn	64.47	52.43
O	14.31	47.57
TOTAL	100.00	100.00

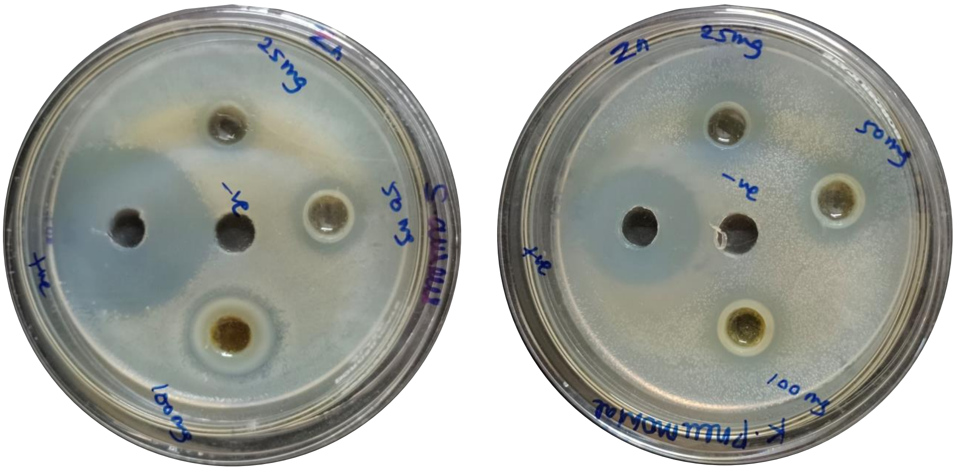
3.6 Anti-bacterial Activity

Using the agar - well diffusion technique, the antibacterial performance of nanoparticles against pathogenic bacterial types *Klebsiella pneumoniae* and *Staphylococcus aureus* was investigated. The conventional antibiotic chloramphenicol was used as the control group for the comparison. The antibacterial activity of ZnO NPs against *Klebsiella pneumoniae* and *Staphylococcus aureus* is shown in Fig. 8a and 8b, respectively.

Zone of Inhibition (ZOI) in mm is presented in Table 2. The percentage of Inhibitions of ZnO NPs against *Staphylococcus Aureus* and *Klebsiella pneumoniae* are given in Tables 3 and 4, respectively.

Table 2. Anti-bacterial activity of synthesized ZnO NPs

Test Pathogens	Zone of Inhibition (ZOI) in mm				
	100 mg/mL	**50 mg/mL**	**25 mg/mL**	**Positive Control**	**Negative Control**
Staphylococcus Aureus	9	7.5	5	15	No ZOI
Klebsiella pneumoniae	7.5	7.5	7.5	11	No ZOI



(a) (b)

Fig. 8. Anti-bacterial activity of ZnO NPs against (a) *Staphylococcus aureus (b) Klebsiella pneumoniae*

Table 3. Percentage of inhibition of ZnO NPs by using test pathogen as *Staphylococcus aureus*

Sample	25 mg/mL	50 mg/mL	100 mg/mL
ZnO NPs	33.33	50	60

Table 4. Percentage of Inhibition of ZnO NPs by using test pathogen as *Klebsiella pneumoniae*

Sample	25 mg/mL	50 mg/mL	100 mg/mL
ZnO NPs	68.18	68.18	68.18

The antibacterial activity data showed that all of the produced nanoparticles efficiently inhibited both *Klebsiella pneumoniae* and *Staphylococcus aureus* bacterial strains. ZnO NPs' antibacterial activity may be attributed to their ability to bind to the bacterial cell membrane and inhibit the active transport process, causing cell lysis [31,32].

4. CONCLUSION

The green synthesis of ZnO NPs by leaf extracts of *Momordica charantia* is revealed. *Momordica charantia* leaf extract was utilized as a reductant. The structure, composition, average particle size, and morphology of ZnO NPs were validated by XRD, FESEM, and EDAX techniques. UV-absorbing band centered at 352 nm. The production of ZnO NPs is confirmed by visible spectra. The existence of a Zn-O bond is established by a band at 630 cm^{-1} in the FT-IR spectra. Based on the XRD pattern, the average particle size of ZnO NPs is 33.81 nm. FESEM reveals that ZnO NPs have an uneven form with 20 - 60 nm range. The occurrence of zinc and oxygen in ZnO NPs is verified by EDAX. ZnO NPs have superior antibacterial action against both *Klebsiella pneumoniae* and *Staphylococcus aureus* bacterial strains.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Upadhyay A, Agrahari P, Singh D. A Review on Salient Pharmacological Features of Momordica charantia. International Journal of Pharmacology. 2015;11(5):405–413.
2. Grover J. Pharmacological actions and potential uses of Momordica charantia: A review. Journal of Ethnopharmacology; 2004.
3. Alwin David S, Vedhi C. Synthesis and Characterization of Co_3O_4 - CuO - ZrO_2 Ternary Nanoparticles, International Journal of Chem Tech Research. 2017;10:905–912.
4. Alwin David S, Revathi LU. Green synthesis of Fe_3O_4 nano particles using Camellia angustifolia leaf extract and their enhanced visible-light photocatalytic activity, International Journal of Advance Research in Science and Engineering. 2018;7(02):422-428.
5. Alwin David S, Ramkumar P, Revathi LU. Chemical synthesis of Fe_3O_4 nano particles for the degradation of Malachite Green and Methyl Orange under visible light irradiation, International Journal of Research and Analytical Reviews. 2018;1(02):1313-1316.
6. Alwin David S, Subramanian P. Biogenesis of Zirconium Oxide Nanoparticles by Momordica charantia (Bitter Gourd) Leaf Extract: Characterization and their Antimicrobial Activities, Journal of Pharmaceutical Research International. 2021;33(61B):354-362,
7. Alwin David S, Ram Kumar P. Biogenesis of MnO_2 Nanoparticles Using Momordica Charantia Leaf Extract, ECS Transactions. 2022;107:747.
8. Alwin David S, Asirvatham Doss, Rajamma Powel Praveen Pole, Thankappan Pushpabai Kumari Pushpa Rani, Raveendran Padma Latha Reshmi, Ramakrishnan Rajalakshmi. Synthesis and characterization of Cobalt Oxide nanoparticles using Momordica charantia and its photocatalytic activity, International Journal of Nano Dimension. 2022;13(3):335 -343.
9. Alwin David S, A Doss, RP Praveen Pole. Biosynthesis of chromium oxide nanoparticles by Momordica charantia leaf extract: Characterization and their Antibacterial Activities, Results in Surfaces and Interfaces. 2023;11:100120.
10. Amininia A, Pourshamsian K, Sadeghi B. Nano-ZnO Impregnated on Starch—A Highly Efficient Heterogeneous Bio-Based Catalyst for One-Pot Synthesis of Pyranopyrimidinone and Xanthene Derivativesas Potential Antibacterial Agents. Russian Journal of Organic Chemistry. 2020;56(7):1279–1288.
11. Ghane M, Sadeghi B, Jafari A, Paknejhad AR. Synthesis and characterization of a Bi-Oxide nanoparticle ZnO/CuO by thermal decomposition of oxalate precursor method. International Journal of Nano Dimension; 2010.
12. Al-Mohaimeed AM, Al-Onazi WA, El-Tohamy MF. Multifunctional Eco-Friendly Synthesis of ZnO Nanoparticles in Biomedical Applications. Molecules. 2022;27(2):579.
13. Akir S, Barras A, Coffinier Y, Bououdina M, Boukherroub R, Omrani AD. Eco-friendly synthesis of ZnO nanoparticles with different morphologies and their visible light photocatalytic performance for the degradation of Rhodamine B. Ceramics International. 2016;42(8):10259–10265.
14. Ren X, Du Y, Qu X, Li Y, Yin L, Shen K, Zhang J, Liu Y. Controllable Synthesis of ZnO Nanoparticles with Improved Photocatalytic Performance for the Degradation of Rhodamine B under Ultraviolet Light Irradiation. Molecules. 2023;28(13):5135.
15. Roshni A, Thambidurai S. Enhanced photocatalytic and antibacterial activity of ZnO with rice field crab chitosan and *Plectranthus amboinicus* extract. Materials Chemistry and Physics. 2022;291:126739.
16. Rai P, Yu YT. Citrate-assisted hydrothermal synthesis of single crystalline ZnO nanoparticles for gas sensor application. Sensors and Actuators B: Chemical. 2012;173:58–65.
17. Faisal S, Jan H, Shah SA, Shah S, Khan A, Akbar MT, et al. Green Synthesis of Zinc Oxide (ZnO) Nanoparticles Using Aqueous Fruit Extracts of Myristica fragrans: Their Characterizations and Biological and Environmental Applications. ACS Omega. 2021;6(14):9709–9722.
18. Primo JDO, Bittencourt C, Acosta S, Sierra-Castillo A, Colomer JF, Jaerger S, Teixeira VC, Anaissi FJ. Synthesis of Zinc

Oxide Nanoparticles by Ecofriendly Routes: Adsorbent for Copper Removal from Wastewater. Frontiers in Chemistry. 2020;8.

19. Naseer M, Aslam U, Khalid B, Chen B. Green route to synthesize Zinc Oxide Nanoparticles using leaf extracts of Cassia fistula and Melia azadarach and their antibacterial potential. Scientific Reports. 2020;10(1).
20. Shah T, Surendar S, Singh S. Green Synthesis of Zinc Oxide Nanoparticles Using Ananas comosus Extract: Preparation, Characterization, and Antimicrobial Efficacy. Cureus; 2023.
21. Iqbal J, Abbasi BA, Yaseen T, Zahra SA, Shahbaz A, Shah SA, et al. Green synthesis of zinc oxide nanoparticles using *Elaeagnus angustifolia* L. leaf extracts and their multiple in vitro biological applications. Scientific Reports. 2021; 11(1).
22. Bala N, Saha S, Chakraborty M, Maiti M, Das S, Basu R, Nandy P. Green synthesis of zinc oxide nanoparticles using *Hibiscus subdariffa* leaf extract: Effect of temperature on synthesis, anti-bacterial activity and anti-diabetic activity. RSC Advances. 2015;5(7):4993–5003.
23. Ukidav V, Ingale LT. Green Synthesis of Zinc Oxide Nanoparticles from *Coriandrum sativum* and Their Use as Fertilizer on Bengal Gram, Turkish Gram, and Green Gram Plant Growth. International Journal of Agronomy. 2022;2022:1–14.
24. Kebede Urge S, Tiruneh Dibaba S, Belay Gemta A. Green Synthesis Method of ZnO Nanoparticles using Extracts of Zingiber officinale and Garlic Bulb (*Allium sativum*) and Their Synergetic Effect for Antibacterial Activities. Journal of Nanomaterials. 2023;2023, 1–9.
25. Umamaheswari A, Prabu SL, John SA, Puratchikody A. Green synthesis of zinc oxide nanoparticles using leaf extracts of *Raphanus sativus* var. Longipinnatus and evaluation of their anticancer property in A549 cell lines. Biotechnology Reports. 2021;29:e00595.
26. Rajeshkumar S, Santhoshkumar J, Shamugam VK. Synthesis of zinc oxide nanoparticles using plant leaf extract against urinary tract infection pathogen. Resource-Efficient Technologies. 2017;4: 459–465.
27. Salih AM, Al-Qurainy F, Khan S, Tarroum M, Nadeem M, Shaikhaldein HO, Gaafar ARZ, Alfarraj NS. Biosynthesis of zinc oxide nanoparticles using *Phoenix dactylifera* and their effect on biomass and phytochemical compounds in Juniperus procera. Scientific Reports. 2021; 11(1).
28. Bhuyan T, Mishra K, Khanuja M, Prasad R, Varma A. Biosynthesis of zinc oxide nanoparticles from *Azadirachta indica* for antibacterial and photocatalytic applications. Materials Science in Semiconductor Processing. 2015;32:55–61.
29. Chaudhuri SK, Malodia L. Biosynthesis of zinc oxide nanoparticles using leaf extract of *Calotropis gigantea*: Characterization and its evaluation on tree seedling growth in nursery stage. Applied Nanoscience. 2017;7(8):501–512.
30. Muhammad W, Ullah N, Haroon M, Abbasi BH. Optical, morphological and biological analysis of zinc oxide nanoparticles (ZnO NPs) using *Papaver somniferum* L. RSC Advances. 2019;9(51):29541–29548.
31. Alwin David S, Subramanian P. Antibacterial Activity of CuO Nanoparticles Synthesized by *Justicia adhatoda* Leaf Extract, Journal of Pharmaceutical Research International. 2021;33(56B):160-170
32. Alwin David, Rajadurai SIS, Kumar V. Biosynthesis of copper oxide nanoparticles using *Momordica charantia* leaf extract and their characterization, International Journal of Advance Research in Science and Engineering. 2017;6(03):313-320.
33. Silva BBM da, Silva BBM da, Souza GP de, Arruda EF de, Muniz AB, Carvalho CM, Rodrigues, RA. Antibacterial Activity of *Calycophyllum spruceanum* Leaf Extract against *Enterococcus faecalis* Strains "*In vitro*" for Endodontic Purposes. Journal of Advances in Biology & Biotechnology. 2023;26(1):33–41.
    Available:https://doi.org/10.9734/jabb/2023/v26i1615
34. Osagie EA, Imuentinyan EMN, Ekundayo IO. *In vitro* Study of *Moringa oleifera* Lam. Leaf Extract Fractions against Multidrug-resistant *Pseudomonas aeruginosa* Strains from Surgical Wound Infections. Journal of Advances in Microbiology. 2022;22(10): 120–131.

Available:https://doi.org/10.9734/jamb/2022/v22i10671

35. Sangeetha G, Rajeshwari S, Venckatesh R. Green synthesis of zinc oxide nanoparticles by *Aloe barbadensis* miller leaf extract: Structure and optical properties. Materials Research Bulletin. 2011 Dec 1;46(12):2560-6.

Peer-review history:
The peer review history for this paper can be accessed here:
https://prh.mbimph.com/review-history/3567