

RESEARCH PAPER

Effect of Zinc Doping on the Structural, Optical, and Antibacterial Properties of Nickel Oxide Nanoparticles Synthesized via Sol-Gel Technique

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ABSTRACT

Sol-gel technique was used to create nickel oxide nanoparticles (NiO-NPs) and modified with zinc ion to enhance the optical, structural and antibacterial properties of the prepared nanoparticles. X-ray diffraction (XRD) analysis was used to examine the structure of each nanoparticle. A highly crystalline cubic structure was identified by XRD measurements. The size and structure of the nanoparticles were investigated morphologically using scanning electron microscopy (FESEM). The Zn-NiO doped sample showed a spherical structure, but the synthesized sample showed a lamellar form according to FESEM examination. Crystal size was evaluated using the Scherrer method and X-ray peak amplitude analysis. The average size of the nickel oxide nanoparticles, as measured by scanning electron microscopy (FESEM), was found to be strongly correlated with the Scherrer equation. Together with UV-Vis absorption spectroscopy results, the photon energy was plotted using the Tauc equation to determine the energy gap. The antibacterial activity of *Staphylococcus aureus* and *Escherichia coli* bacteria was tested against the nanoparticles. According to the findings, pure nickel nanoparticles had less antibacterial activity than grafted (Zn-NiO) nanoparticles.

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INTRODUCTION

One of the most important fields of study in modern science is nanoscience [1]. The term "nanomaterials" refers to materials whose dimensions fall inside the nanoscopic range, which is defined as atoms with sizes between 1 and 100 nm [2]. Scientists can now modify matter at the atomic and molecular levels thanks to nanotechnology, which has significantly advanced the life sciences and healthcare [3, 4]. There are numerous applications for metal oxide

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nanoparticles (NiO NPs) in the fields of biological sciences, energy, agriculture, health, and the environment. Scientific groups have recently become more interested in these applications [5]. Large surface area, enhanced solubility, reactivity, and unique optical, electronic, and magnetic properties are among the size-dependent features of NiO NPs [6]. They also have remarkable chemical and physical properties such increased surface reactivity, high thermal conductivity, and mechanical solid stability [7]. Certain chemicals



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have qualities that make them highly promising for use in the biological sciences [8]. Furthermore, NiO NPs' application in nanobiotechnology is improved by their ability to generate a significant amount of reactive oxygen species (ROS) with a high selectivity for certain targets and less susceptibility to healthy cells [9]. Unique features that were distinct from the bulk materials were produced via the synthesis of inorganic or organic compounds [10]. Due to the unique properties of magnesium oxide nanoparticles, nickel oxide is an important inorganic substance that has found widespread uses in many industries, making it useful in industry, medicine, and various research projects [11]. With many applications, nickel oxide is a very efficient drying agent [12]. It catalyzes chemical reactions, especially those involving oxidation-reduction and the synthesis of fuels and hydrogen [13]. Additionally, it is utilized in the production of electrical charges and batteries, particularly some rechargeable battery kinds [14]. Because of its semiconductor qualities, it is utilized in solar cells and electronic gadgets. Also, it is used as a coloring agent and to give some characteristics to glass and ceramics [15]. Nickel oxide nanoparticles, which have uses in energy, medicine, and the environment, are created using it in nanotechnology research [16]. It is used as a conductive material in the electrodes of super capacitors and energy storage devices due to their high conductivity and electrochemical activity [17]. Moreover, NiO also has good bacteria performance due to its ability to generate superoxide [18]. Nickel oxide nanoparticles are created using a variety of physical and chemical techniques, such as sol-gel [19]. This is a technique that can be applied to produce different types of oxides and materials at extremely low temperatures, which is useful for heat sensitive materials, and can lower the energy consumption [20]. Moreover, it provides a good control of the homogeneity and organization of the manufactured components, thus good uniformity of properties throughout the material [21]. It is also suitable for producing materials of amazing purity. Also, it is easily available for both research institutes and business applications because it is relatively simple and inexpensive as compared with other methodologies [22]. Besides this, the nanoparticles generated by sol gel technique have special structural characteristics that make them more active against bacteria, such as: high surface area, nanometer size and

nanometer porosity [23]. The aim of this work was to investigate the effect of the zinc acetate doped nickel oxide and NiO prepared by sol-gel method on the activity of bacteria *Escherichia coli* and *Staphylococcus aureus*.

MATERIALS AND METHODS

Synthesis of NiO NPs

The sol-gel technique was used to create NiO NPs. in accordance with the subsequent process. 400 milliliters of deionized distilled water were mixed with 43.3 grams of nickel nitrate hexahydrate. After then, 3.3 grams of zinc acetate were added. Next, add 21.04 grams of citric acid. Lastly, 25 milliliters of ethylene glycol were added. One ingredient should be added every 30 minutes. After the mixture was heated to 80°C for three to five hours at a medium speed on an electric stirrer, the gel was formed. After that, the precipitate was kept at 125°C for the entire night in an electric oven. The final product was obtained by calcining the resultant solid material for three hours at 500°C. Commercial sources provided all of the materials used in this method, and no additional technical assistance was required.

Characterization of NiO powder

Phase and structural identification of our synthesized NiO samples was done with the "PanAlytical Aeris Benchtop X-ray Diffractometer 40 kW, 7.5 mA" from Cu-K α radiation source (0.154 nm). The data were analysed with the use of X'pert High Score Plus software. Global parameters such as scale factor and background were refined at the beginning of the Rietveld refinement. The process relies on fitting using a fourth-degree polynomial. Subsequently, the unit-cell parameters, profile half-widths (u, v, w), site occupancy factor, peak shape parameters, and preferred orientation were refined. Field emission scanning electron microscopy (FESEM, Zeiss Supra 35) was used to investigate the NiO samples morphology. The UV-visible and photoluminescence spectroscopy were performed with UV-visible spectroscopy (Varian-Cary Win UV 100) and photoluminescence spectroscopy (Jobin Yvon HR 800 UV), respectively. From the characterization results, we chose the best growth properties sample.

Antibacterial activity

The antibacterial activity of pure NiO NPs and doped Zn-doped NiO NPs was evaluated

by employing the agar well diffusion method against selected bacterial isolates: Gram-positive *Staphylococcus aureus* (*S. aureus*) and Gram-negative *Escherichia coli* (*E. coli*). This method was applied at concentration of 10 mg/mL, with minor modifications. In short, the bacterial isolates were initially grown in nutrient broth and incubated at 37 °C for 18–24 h to harvest log-phase cells. Following incubation, 0.1 mL of each bacterial suspension was plated on nutrient agar plates, which were then briefly incubated at 37 °C for 24

h to ensure appropriate growth. Then a single, well-isolated colony was transferred to a test tube containing 5 mL of sterile normal saline, and the suspension's turbidity was adjusted to match that of the standard turbidity solution, corresponding to approximately 1.5×10^8 CFU/mL. The prepared bacterial suspension was evenly and gently spread over Mueller–Hinton agar plates using a sterile cotton swab, then left to stand for 10 minutes to allow absorption. Well of about 5 mm in diameter were subsequently punched aseptically

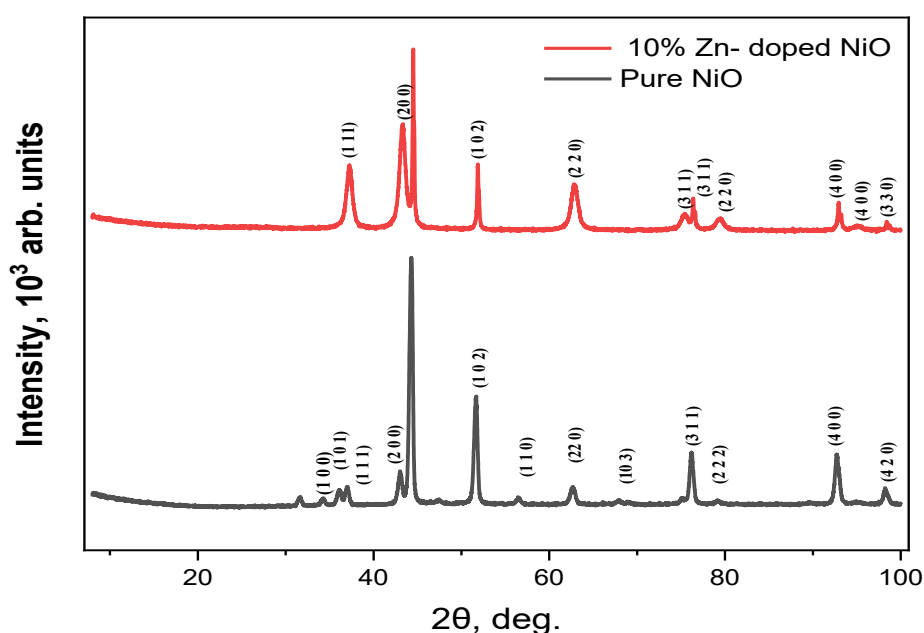


Fig. 1. X-ray diffraction pattern of the prepared nickel oxide nanoparticles.

Table. 1. XRD analysis of structural parameters of pure NiO nanoparticles.

No.	2θ (deg)	h k l	Crystallite Size (nm)	FWHM (degree)
1	31.531	Not match	12.1	0.683
2	34.260	(100)	14.6	0.568
3	36.062	(101)	12.2	0.683
4	37.018	(111)	14.7	0.568
5	43.025	(200)	18.9	0.453
6	44.393	Not match	25.4	0.338
7	51.709	(102)	26.1	0.338
8	56.440	(110)	13.2	0.683
9	62.626	(220)	11.7	0.798
10	67.896	(103)	12.0	0.798
11	76.179	(311)	19.8	0.51
12	79.100	(222)	11.3	0.913
13	92.724	(400)	22.6	0.51
14	98.254	(331)/(420)	30.7	0.395
NiO				
Ni(OH) ₂		FCC cubic		

into the agar (two well per plate). To prepare for the experimentation phase, the agar plugs were removed from their respective plates, and 50 μ L of pure NiO and Zn-doped NiO NPs suspensions EPS were introduced in their designated well via micropipette; distilled water was added to the central well as a negative control. Inoculated plates were incubated for 18 h at 37 °C, and antibacterial activity was determined by measuring the diameters of inhibition zones around each well. This allowed us to comparatively analyze the EPS effect in Gram-positive and Gram-negative oral pathogens.

RESULTS AND DISCUSSION

Structural studies

Fig. 1 shows the X-ray diffraction (XRD) pattern of the pure and zinc-doped nickel oxide nanostructure. The diffraction peaks show a decrease in width with increasing temperature. The analysis indicated the presence of both the pure and doped structures.

The crystal structure and phase composition of the synthesized pure and Zn-doped NiO nanoparticles were examined using X-ray diffraction (XRD), and the obtained diffraction patterns are presented in Fig. 1.

Fig. 1 shows XRD pattern of pure NiO nanoparticles showing the characteristic diffraction peaks corresponding to the cubic FCC NiO structure and XRD pattern of Zn-doped

NiO nanoparticles the preservation of the cubic FCC NiO structure after Zn incorporation. The corresponding structural parameters extracted from the diffraction data are summarized in s 1 and 2. The XRD results showed that the successful formation of the crystalline NiO nanoparticles with a cubic face centered cubic (FCC) structure was achieved.

UV-visible spectroscopy

Fig. 2a-c shows the optical behaviour of the pure and synthetic nickel oxide nanoparticles. The properties were measured using absorption spectra in the range of ultraviolet and visible wavelengths (200 - 1100 nm). The formula for the calculation of the band gap of NiO nanoparticles is given as: For the pure NiO sample, the diffraction peaks observed at approximately 37.01°, 43.02°, 62.62°, 76.17°, 79.10°, 92.72°, and 98.25° were indexed to the crystallographic planes (111), (200), (220), (311), (222), (400), and (331)/(420). The peaks are typical of cubic rock-salt structure of NiO and match the previously reported standard diffraction data. The diffraction peaks were sharp and well-defined, suggesting the formation of highly crystalline nanoparticles of the observed reflections, the strongest reflection was from the (200) plane, which appeared close to 43° indicating that a preferred crystal orientation occurred along this direction. This preferential growth phenomenon is frequently seen in chemically

Table. 2. Structural parameters of XRD analysis of NiO-doped Zn NPs.

No.	2 θ (deg)	h k l	Crystallite Size (nm)	FWHM (degree)
1	37.200	(111)	0.98	0.856
2	43.300	(200)	12.5	0.683
3	44.527	Not match	63.1	0.136
4	51.880	(102)	96.0	0.092
5	62.834	(220)	0.91	1.028
6	75.328	(311)	12.6	0.798
7	76.389	(311)	74.3	0.136
8	79.469	(222)	0.90	1.142
9	92.939	(400)	84.8	0.136
10	95.156	(400)	10.3	1.144
11	98.471	(331)/(420)	74.2	0.164
Average crystallite size = 414Å(41.4 nm)				

synthesized NiO nanostructures, and it has been correlated to the improvement of crystal ordering during the calcination process. In addition to the major NiO reflections, a few weak peaks were detected at approximately 34.26° , 36.06° , 51.70° , 56.44° , and 67.89° . Such weak reflections might be attributed to the presence of small amounts of nickel hydroxide phases, such as $\text{Ni}(\text{OH})_2$, which may be due to incomplete conversion of hydroxide to oxide during the synthesis. It is found that the intensity of these peaks is relatively low, which means that the prepared samples are in the main NiO phase.

The lattice parameter of the pure NiO sample was determined to be about $a=4.20 \text{ \AA}$, which is in good agreement with the standard value of the lattice parameter of cubic NiO. The average crystallite size was calculated by Debye-Scherrer equation:

$$D = K\lambda / (\beta \cos \theta) \quad (1)$$

The above equation is derived from where

D is the crystallite size, K is the shape factor, λ is the wavelength of the Cu $K\alpha$ radiation, β is the full width at half maximum (FWHM), and θ is the Bragg angle. The average crystallite size of the pure NiO nanoparticles was measured to be around $175 \text{ \AA}$ (17.5 nm) which confirms the nano crystalline nature of synthesized material.

The overall diffraction pattern of the Zn doped sample was still composed of the characteristic peaks of cubic NiO, suggesting that the FCC structure was also s after the incorporation of the Zn ions. The main diffraction peaks located at approximately 37.2° , 43.3° , 62.83° , 75.32° , 79.46° , 92.93° , and 98.47° were assigned to the planes (111), (200), (220), (311), (222), (400), and (331)/(420), respectively. However, the improvement in the peak-shape and diffraction behavior was observed in Zn incorporated. Several additional weak reflections appeared near 44.52° , 51.88° , 76.38° , and 95.15° . These peaks could be related to either lattice distortion due to Zn or to slight structural changes in the NiO lattice in the presence of Zn. The change in peak shape

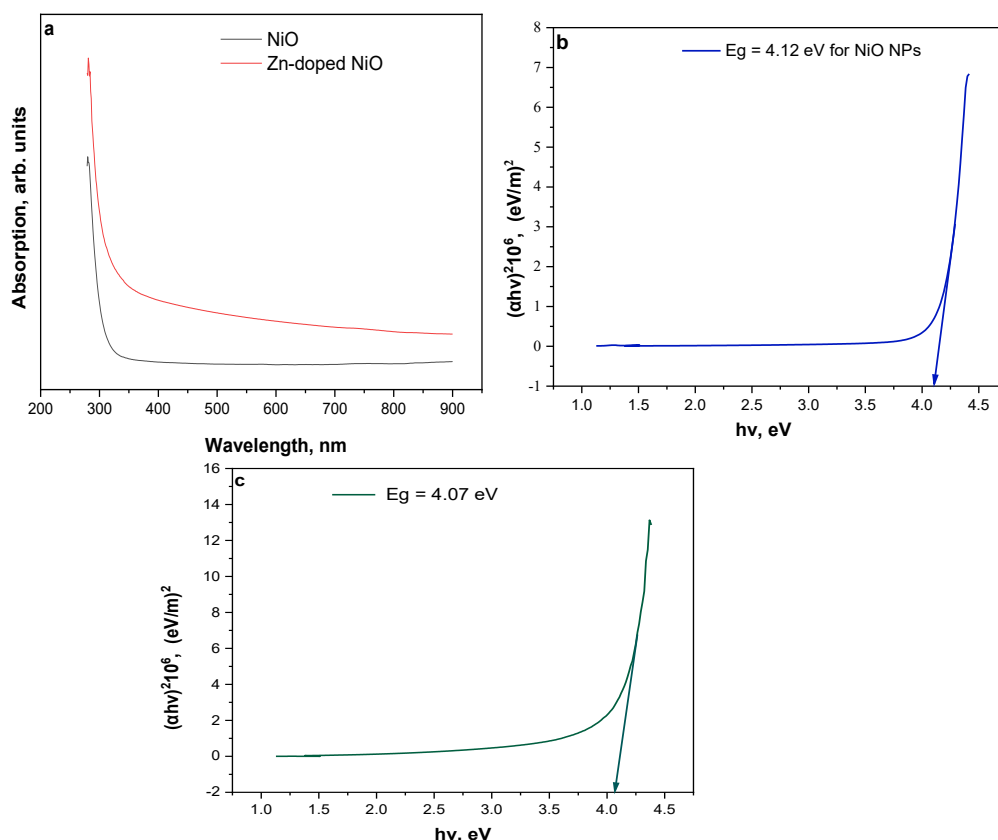


Fig. 2. (a) Absorption of NiO and Zn-doped NiO NPs and Energy gap of (b) NiO NPs, and (c) Zn-doped NiO NPs.

and intensity could be due to the insertion of lattice defects and local strain caused by the larger ionic radius of Zn^{2+} compared to NiO^{2+} .

The diffraction peaks of the Zn-doped NiO nanoparticles were sharper than the pure NiO sample and the average crystallite size of the doped sample was about $414\text{\AA}$ (41.4 nm), as compared to the pure NiO sample. The increase in crystallite size indicates that the addition of Zn has led to crystal growth and enhanced the crystallinity of the prepared nanoparticles. The increase in the crystallinity could be attributed to the role of Zn ions in the nucleation and grain growth phenomena during the synthesis process.

In general, the XRD data indicates that doping of NiO nanoparticles with Zn has been successful to alter the structural properties of NiO nanoparticles without changing the crystal structure from cubic FCC. The observed growth in crystallite size, appearance of extra weak reflections and minor changes in the peak shape suggest that the Zn ions had some control over the crystal growth behaviour and some perturbation of the local structure of NiO.

$$\alpha = A (h\nu - E_g)^{1/2} / h\nu \quad (2)$$

with α , E_g and A representing the band gap, constant and absorption coefficient, respectively. [31].

In the above equation, α denotes the band gap, E_g denotes the constant and A denotes the

absorption coefficient [24]. The NiO NPs optical band gap energy, and Zn-doped NiO NPs was found to be 4.12 eV and 4.07 eV, respectively. The values obtained are comparable to previous reported work, finding that NiO oxide is a semiconductor having a large and direct energy gap [25]. The synthesized NiO NPs are found to have a band gap of 4.12 eV, and when the NiO NPs are doped with Zn, a slight red shift of the band gap is observed to be 4.07 eV. This transition is explained from the point of view of the larger particle sizes and morphology closer to a spherical shape of the Zn-doped NiO NPs resulting in a lesser quantum confinement effect and surface defect [26]. Thus, the surface-to-volume ratio is lower for the NiO NPs doped with Zn so that there are less number of surface states that can trap charge carriers, and hence the band gap energy is affected by the presence of these surface states [27].

Morphological studies

Nanoflakes, nanoflowers and nanoparticles can be formed by some of the above mentioned factors such as careful choice of precursors, control of the response parameters, controlled synthesis procedures, and the use of post-synthesis treatments, etc. [29]. The fabrication of nanoflakes, nanoflowers and nanoparticles with different properties and applications in various fields such as electronics, catalysis, sensing and medicine can be achieved by precise control of these parameters [30]. Response parameters that

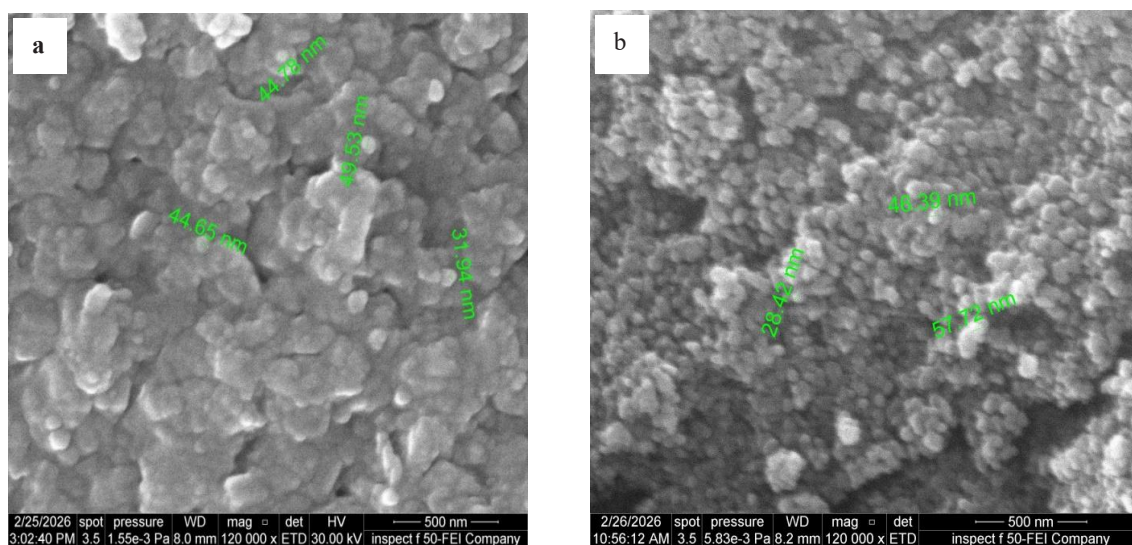


Fig. 3. FSEM images of (a) pure NiO nanoparticles and (b) Zn-doped NiO nanoparticles.

affect the rate of the reaction and formation of the nanostructure have significant influence over the morphology and dimensions of the nanostructures [31]. The parameters that are covered are precursor concentrations, reaction time, pH, temperature, and pressure. One way to improve response time is to enhance the likelihood of achieving improved nanostructure growth [32]. Thus, the formation rate of the nanostructure directly depends on these parameters and thus affects the size and form of the nanostructure [33]. The Fig. 3 are the scanning electron microscope (FSEM) images of Pure and impure Nickel oxide (NiO) nanoparticles. The results indicate a nanoparticles morphology of the particles and smooth surfaces. There were some particles that were heavily.

Scattered, with most concentrated in a group. Average size of Pure NiO nanoparticles was 42.71 nm with size range between 31.94 nm and 49.53 nm, while in the case of Zn-doped NiO nanoparticles, the average size was 44.17 nm, ranging from 28.42 to 57.72 nm. Observed nanostructures are created by rapid room-temperature reduction, assembly and sintering of nanoparticles.

Energy dispersive X-ray spectroscopy (EDX)

Using X-ray scattering and a FESEM examination, the materials and percentages of each substance or element within the sample are determined. The elements in each sample, which were prepared using the sol-gel method, are shown in Fig. 4. After X-rays are emitted and dispersed at particular wavelengths, an EDX analysis is performed to determine the amounts of each material and the chemical makeup of the different compounds.

The sample was discovered to include Nickel oxide nanoparticles. The components present in the sample were found to be 85.5% NiO, 6.4% O, 8.3% Zn, [34]. Chemical composition of nickel and oxygen nanoparticles, in addition to some other elements O Atomic % (20.2) Weight % (6.4), Ni Atomic % (73.6) Weight % (85.5), Zn Atomic % (6.2) Weight % (8.0).

Antibacterial activity

The antibacterial activity of synthesized pure NiO NPs and Zn-doped NiO NPs was evaluated against Gram-positive *S. aureus* and Gram-negative *E. coli*. The findings indicate that both the pure and doped samples demonstrated an inhibitory effect; however, the Zn-doped NiO NPs displayed notably greater antibacterial activity than the NiO NPs. Overall, Pure NiO and doped samples show higher inhibition efficiency against *S. aureus* than *E. coli*. These differences are due to variations in bacterial cell wall structure and sensitivity to NPs. The difference between these two groups lies in their respective cell wall structures: peptidoglycan and the absence of an outer membrane characterize Gram-positive bacteria, whereas a thin layer of peptidoglycan sandwiched between an inner and outer membrane defines Gram-negative bacteria [35]. Thus, Gram-positive bacteria, in general, are more sensitive to membrane disruption [36]. The quantities and diameters of the inhibitory zones utilized with each bacterial species used in the paper are Concentration (10 mg/mL) of the *S. aureus* and Inhibitory Zone Diameter (20 mm) Pure NiO NPs, Zn-doped NiO NPs, while *E. coli* concentration was (10 mg/mL) and Inhibitory

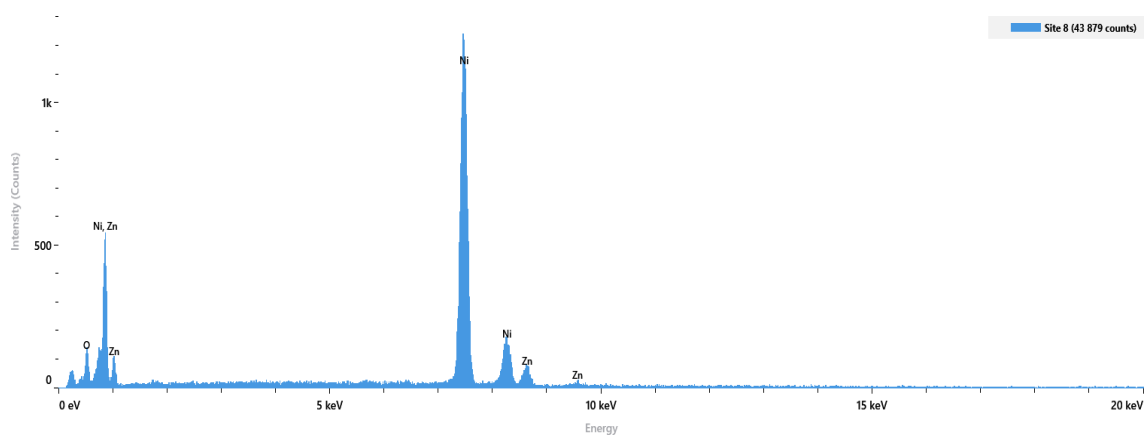
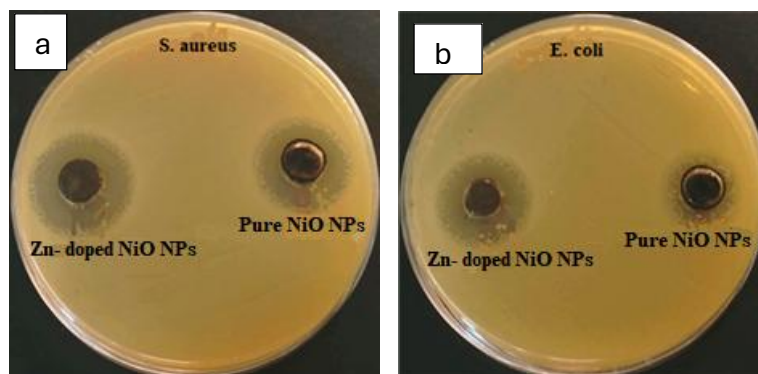


Fig. 4. EDX analysis of NiO nanoparticles and certain impurities (other chemical elements).

Fig. 5. Antibacterial test (a) *S. aureus* and (b) *E. coli*.

Zone Diameter (14 mm) of Pure NiO NPs, Zn-doped NiO NPs (21mm). The antibacterial activity of both pure NiO NPs and Zn-doped NiO NPs has been tested by the agar well diffusion method as shown in Fig. 7. The concentration of pure and doped was 10 mg/mL in each well; two wells were present in each Petri dish. The activity is clearly seen to be concentration-dependent wherein the NiO: Zn-doped NiO NPs exhibited the highest inhibition zone against *S. aureus* and *E. coli* (Fig. 5a, b). This improved antibacterial performance cannot be attributed to the surface area or the morphological items. The persistence of enhanced antibacterial activity may indicate the dominance of another mechanism, since the dopant invariably leads to larger crystallite sizes, which would otherwise reduce surface reactivity. The most salient reason ascribed to this improvement is Zn-doping-induced defect engineering. These defects act as charge-trapping sites, which in turn enhance electron-hole separation and promote the generation of ROS, such as $\bullet\text{OH}$ and $\text{O}_2^{\bullet-}$. The increased production of ROS stems from oxidative stress, which subsequently causes significant harm to cellular structures and triggers cell death. Furthermore, the reduced band gap observed in Zn-doped samples facilitates electron excitation and surface redox reactions, thereby promoting ROS generation. The antibacterial activity of Zn-doped NiO NPs is mainly due to changes in their electronic structure. ROS are particularly noticeable when there are defects, rather than changes in the particle's size or shape.

CONCLUSION

Pure nickel oxide nanoparticles (NiO NPs) and zinc acetate-doped nickel oxide nanoparticles

(Zn-doped NiO NPs) were prepared using the sol-gel method. Their average sizes were 31.94 nm for the pure nanoparticles and 28.42 nm for the zinc acetate-doped nickel oxide nanoparticles. The samples were then analyzed using field-emission scanning electron microscopy (FESEM), X-ray diffraction (XRD), and UV-Vis spectroscopy. Surface morphology studies showed that the size distribution of the pure and doped nickel oxide nanoparticles ranged from 31.94 to 49.75 nm and from 28.42 to 57.72 nm, respectively. Furthermore, X-ray diffraction analysis confirmed that the nanoparticle samples had a monoclinic structure. Untreated nickel oxide nanoparticles exhibited lower antibacterial activity than treated nickel oxide nanoparticles. Furthermore, absorption spectra in the UV-Vis and visible ranges revealed an energy gap of 4.12 eV for untreated nickel oxide nanoparticles, significantly higher than the previously reported value of 4.07 eV for zinc acetate-treated nickel oxide nanoparticles. The narrowing of the energy gap in the doped nanoparticles reflects greater control over their production, leading to improved optical properties. The enhanced antibacterial activity of zinc acetate-doped nickel oxide nanoparticles at all concentrations, compared to undoped nickel oxide, is attributed to the defect engineering resulting from zinc incorporation. This leads to a marked improvement in antibacterial properties, despite the increased crystal size. This seemingly paradoxical observation can be explained by the formation of oxygen voids and defect states in the nickel oxide lattice. This occurs when divalent nickel ions are replaced by zinc ions or displaced. These voids act as charge traps, helping to reduce electron and hole recombination. As a result, this

process effective charge separation. This process then leads to a significant increase in ROS levels including superoxide anions and hydroxyl radicals. The enhanced generation of ROS predominates over the negative effect of the reduced specific surface area associated with larger crystallite dimensions, resulting in greater damage to bacterial cells. Thus, electrical modifications caused by defects are the main factors affecting the antibacterial activity rather than morphological parameters. A detailed investigation can be obtained by measuring other characterization parameters including Density derived from X-ray diffraction, dislocation density, specific surface area, stacking fault probability, and lattice strain. The data obtained will provide insight into the structural and physical properties of the nanoparticles, thereby confirming their functional efficiency and potential application areas.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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