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Investigating the antibacterial activity of Zinc Oxide Nanoparticles against *staphylococcus aureus isolates*

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Abstract

In the current research, Zinc oxide nanoparticle solution was prepared at different concentrations. A minimum particle size between (52.31 - 134.3 nm) was obtained. The structural and optical properties were determined as functions of concentration. Results of the Field Emission Scanning Electron Microscopy (FE-SEM) indicated that as the concentration of zinc oxide nanoparticles decreased, their size also decreased. Antibacterial activity for Zinc Oxide nanoparticles was tested by the extent of killing *Staphylococcus aureus isolates,* and using a lower Zinc Oxide nanoparticle concentration revealed 50% more activity than higher concentrations.

Keywords: ZnO Nanoparticles; *Staphylococcus aureus isolates*; Antibacterial activity

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Introduction:

Nanotechnology is a trendy topic for research in today's scientific and technological breakthroughs because it has so many new applications in medical procedures, food processing, inventive fabric compounds, and agricultural production [1,2]. High levels of advancement are used to establish the features and goals of both natural and artificial systems [3]. This study is interested in materials that, because of their nanoscale size, produce novel and significantly enhanced physicochemical and biological properties, as well as unique phenomena and functionalities [4]. Nanoparticles (NPs) often have bigger surface areas than macro-sized particles due to their nanoscale size [5].

NPs are known as controlled or managed particles (1–100 nm) at the atomic level, and they differ significantly from bulk materials in terms of size-related properties [6,7]. Because of their special properties, NPs have larger structures than their counterparts despite their small size. Because of this, they may be helpful in applications involving

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nanomedicine and highly desirable biosensors [8]. The size, content, crystallinity, and shape of metal nanoparticles (NPs), such as silver, zinc oxide (ZnO), titanium dioxide (TiO2), as well as their chemical, mechanical, electrical, structural, morphological, and optical characteristics, are the main characteristics that can be altered by reducing their size to the nanoscale [9].

These changed characteristics allow the NPs to interact with cell biomolecules uniquely and facilitate their physical entry into cellular structures [8]. The increased fraction of atoms at the surface of nanostructured materials results in increased surface reactivity. As a result, nanomaterials have become more significant in bionanotechnology and the basic and applied sciences in recent years. Numerous scientists have examined ZnO's diverse forms and exceptional nanoscale antibacterial capabilities against a broad range of bacterial species [10–15]. The antibacterial properties of ZnO formulations at the micro and nanoscale are currently being studied. When reduced to the nanoscale region, ZnO particles show notable antibacterial properties. The zinc oxide nanoparticles can show several bactericidal processes after interacting with the bacteria's surface and/or core [14,16]. These rare compounds, mostly used in the food sector, are highly toxic and easily interact with microorganisms to act as antibacterial agents. Thankfully, some investigations have demonstrated that ZnO-NPs are safe to use as antibacterial agents, are toxic to microorganisms, and are highly compatible with human cells [17]. Additionally, they do not harm human cells [18,19]. They can be used safely as antibacterial agents, are poisonous to microorganisms, and have good biocompatibility with human cells [17]. The high specific surface area-to-volume ratios of nanoparticles are primarily responsible for their diverse antibacterial properties [20]. However, because of their peculiarities in physiochemistry, they have been the focus of more discussion. Understanding this important global health issue would be aided by research into ZnO-NPs as an antibacterial agent [21].

Experimental:

Staphylococcus aureus isolates were chosen and prepared for testing in the present bioactivity application. *Staphylococcus aureus* isolates were cultured in nutrient broth for 24 hours. Following an overnight incubation, the isolates were diluted to McFarland standard. A diluted bacterial suspension of 0.1 ml was placed on Muller Hinton agar plates and distributed evenly, as shown in Figure 1. Different concentrations of ZnO Nanoparticles solutions were prepared by dissolving powdered ZnO nanoparticles in deionized water to produce (9.0×10^{-1}) mol/L; which then reduced to (5.0, 1.0, 1.1 and 1.6) $\times 10^{-1}$ mol/L, by the dilution equation, homogenized by vortex mixer, and added to the plates. After a final 24-hour incubation period at 37 °C, the development of growth inhibitory zones was investigated.

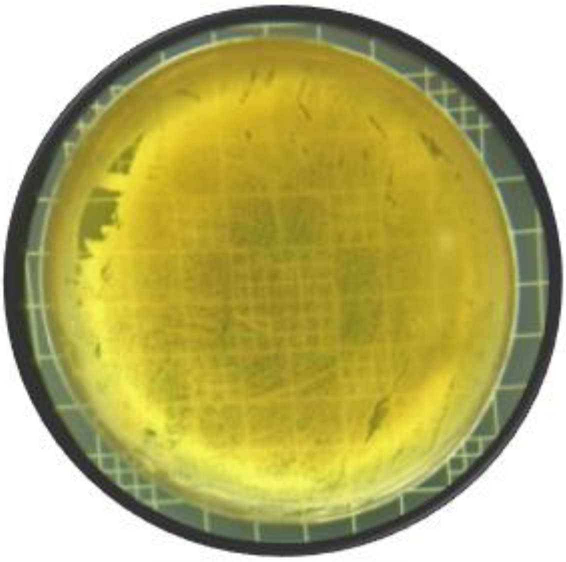


Figure1: *Staphylococcus aureus* bacteria isolates before treatment

Results and discussion:

To investigate the phase structure for the ZnO nanoparticles that were produced powder, XRD patterns of ZnO NPs were characterized as shown in figure 2. The X-ray pattern reveals a polycrystalline structure of the ZnO Nanoparticles with 11 peaks obtained at diffraction angles in the diffraction pattern: 31.8° (100), 34.4° (002), 36.2° (101), 47.6° (102), 56.6° (110), 63.3° (103), 66.6° (200),68.3° (112), 69.2° (004), 72.8° (201) and 77.2° (202). This refers to a hexagonal Structure [22-23].

concentration.

Field Emission-Scanning Electron Microscopy is a convenient method to investigate the samples' structure due to its important role in many applications. Figure 3 illustrates FE-SEM images of ZnO nanoparticle solutions, where the surface is covered with particles of different shapes (nanosheets, star shapes and rods) uniformly distributed over the entire surface.

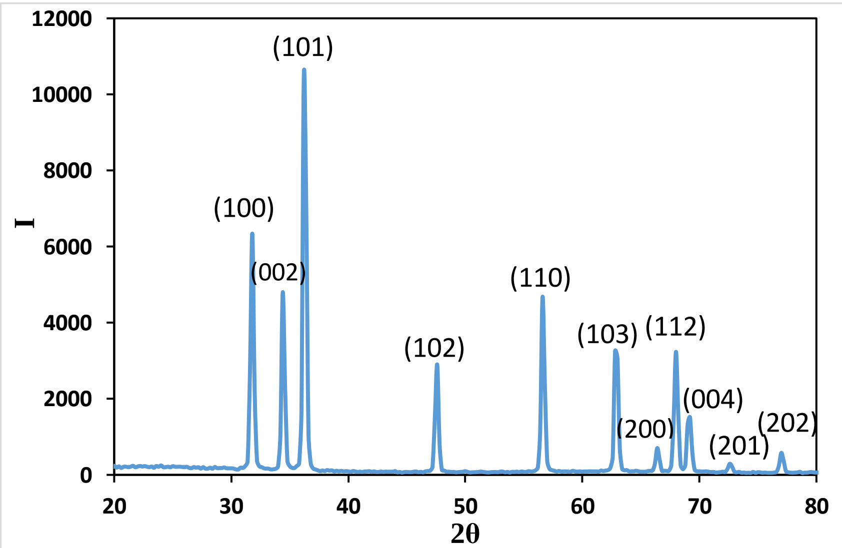


Figure 2: X-ray Diffraction of ZnO Nanoparticles at 9.0 × 10^{-1}M

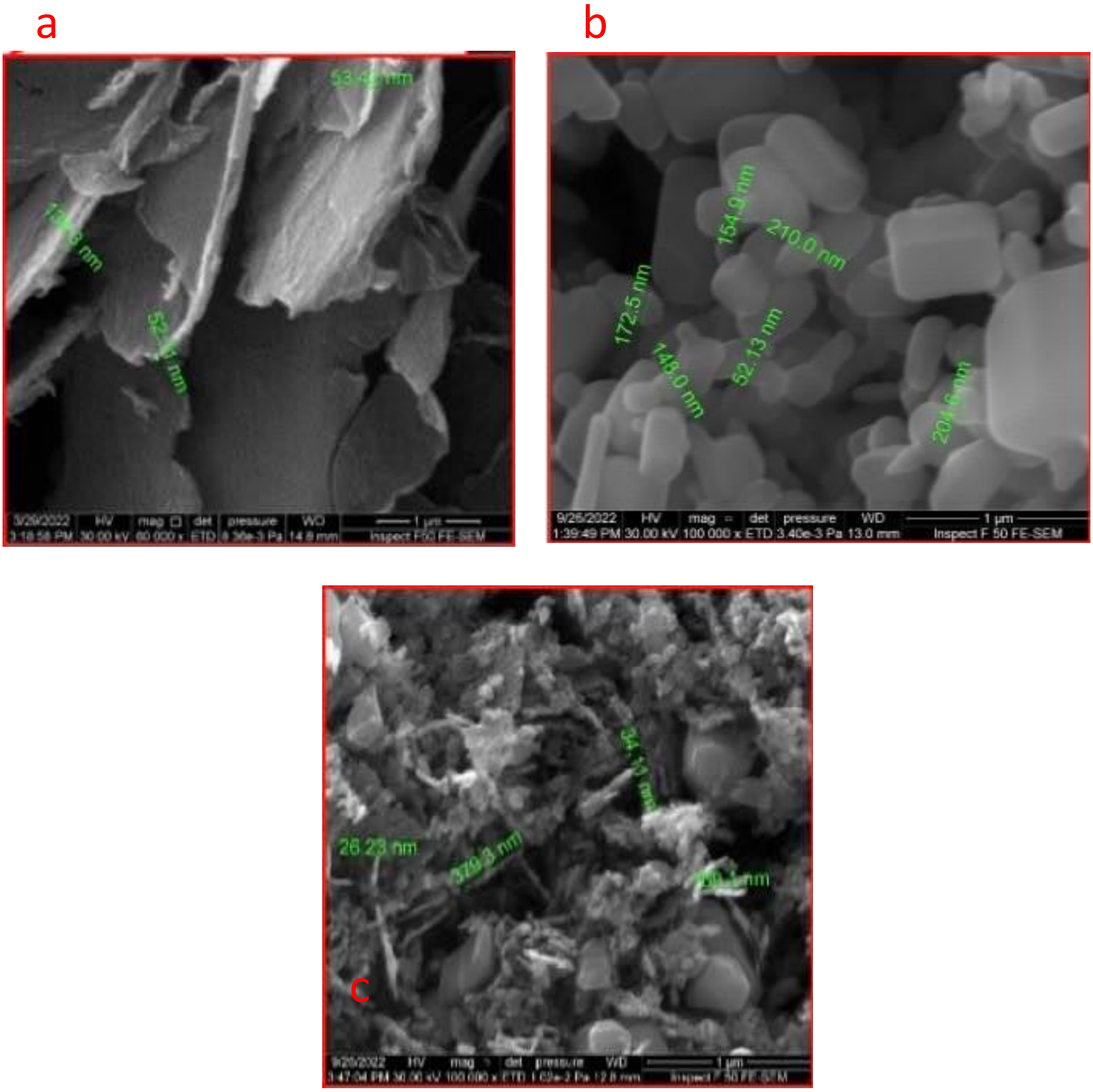


Figure 3: FE-SEM images of ZnO Nanoparticles at (a) 1.6 × 10^{-1} M, (b) 1.0 × 10^{-1} M & (C) 9.0 × 10^{-1}M concentrations.

By looking carefully at the images of Figure 3, it is possible to observe a regular distribution of nanoparticles at low concentrations. The particle size distribution gradually changes from small to larger clusters. The particle size value obtained from FE-SEM measurements indicates that at (1.6 × 10^{-1} M) concentration, the size is in the range of 52.31 to 134.3 nm (Figure 3-a). At higher concentration (1.0 × 10^{-1} M), it is in the range (52.13 - 210.0 nm) (Figure 3-b), and it is between (26.23 to 379.3 nm) at (9.0 × 10^{-1}M) as presented in Figure 3–c. These results confirm the fact that small particle size is obtained at low concentration. The figure also shows that there are some particle agglomerations. Our findings are in good agreement with research cited elsewhere [24].

The FTIR spectrum of ZnO Nanoparticles solution at 9.0 × 10^{-1} M concentration within the wavenumbers range (400 to 4000 cm^{-1}) is shown in Fig. 4. Peaks at 559,670 and 813 cm^{-1} are characteristic absorption of ZnO bonds and refer to hexagonal ZnO phase; corresponding with XRD. The peak at 3428 cm^{-1} is attributed to the O-H absorption. The range (2907 and 2936 cm^{-1}) is related to C-H, and the peak at 2359 cm^{-1} is attributed to the absorption of C=C. The peaks at 1659, 1458, and 1557 cm^{-1} are attributed to the absorption of C=O, and finally, the 1103 cm^{-1} peak is related to C-N [25].

The Energy Dispersive X-ray (EDX) spectra of ZnO Nanoparticles were recorded and analyzed, as shown in Figure (5). Elemental compositions for the final samples are shown in the tables and the figures. All of the investigated samples' EDX spectra verified the presence of oxygen and zinc atoms in the thin layers, indicating the thin film formation. For the final samples, the weight percentage of zinc, oxygen, and carbon was found to be 48.2%, 36.8%, and 14.9% at a concentration 9.0 × 10^{-1} mol/L, as shown in Figure (5-a). At a concentration 0.5 × 10^{-1} mol/L, the percentage of zinc, oxygen, and carbon was found to be 48.1%, 36%, and 16%, as shown in figure (5-b). However, at a concentration 1.0 × 10^{-1} mol/L, the percentage of zinc, oxygen, and carbon was found to be 39.5%, 41.4%, and 19%, as shown in figure (5-c). At a concentration 1.1 × 10^{-1} mol/L, the percentage of zinc, oxygen, and carbon was found to be 31.7%, 40.6.4%, and 26%, as shown in figure (5-d). At a concentration 1.6 × 10^{-1} mol/L, the percentage of zinc, oxygen, and carbon was found to be 31.5%, 42.4.4%, and 26%, as shown in figure (5-e).

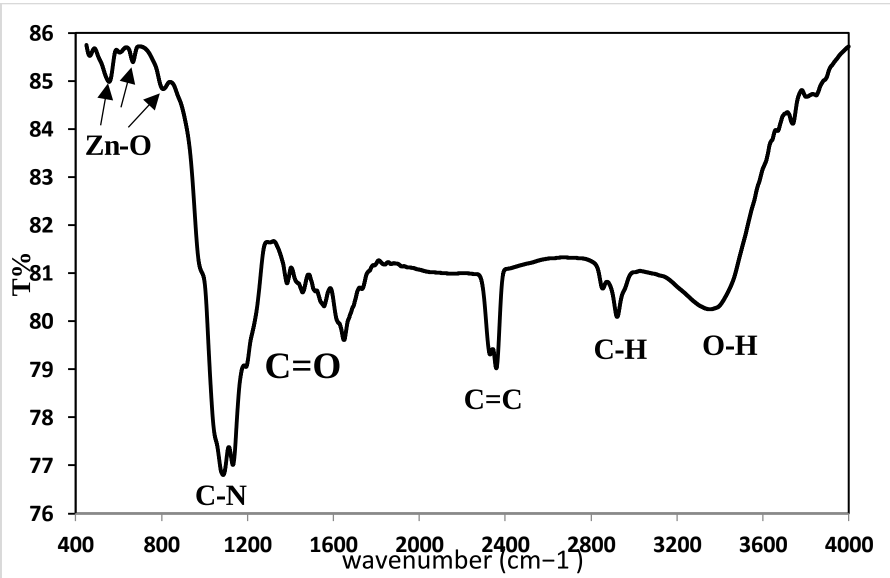
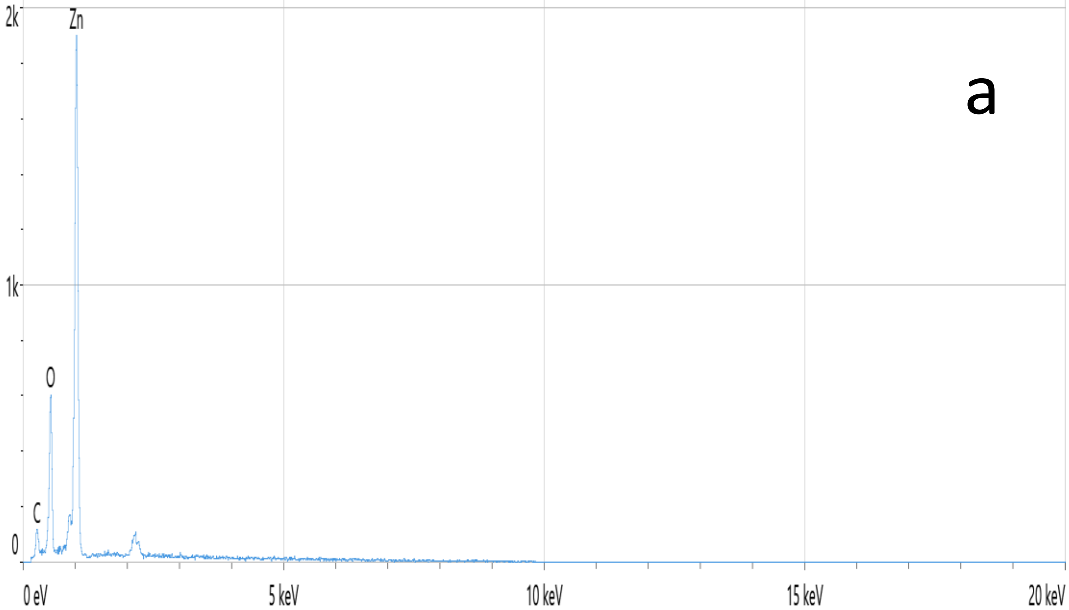
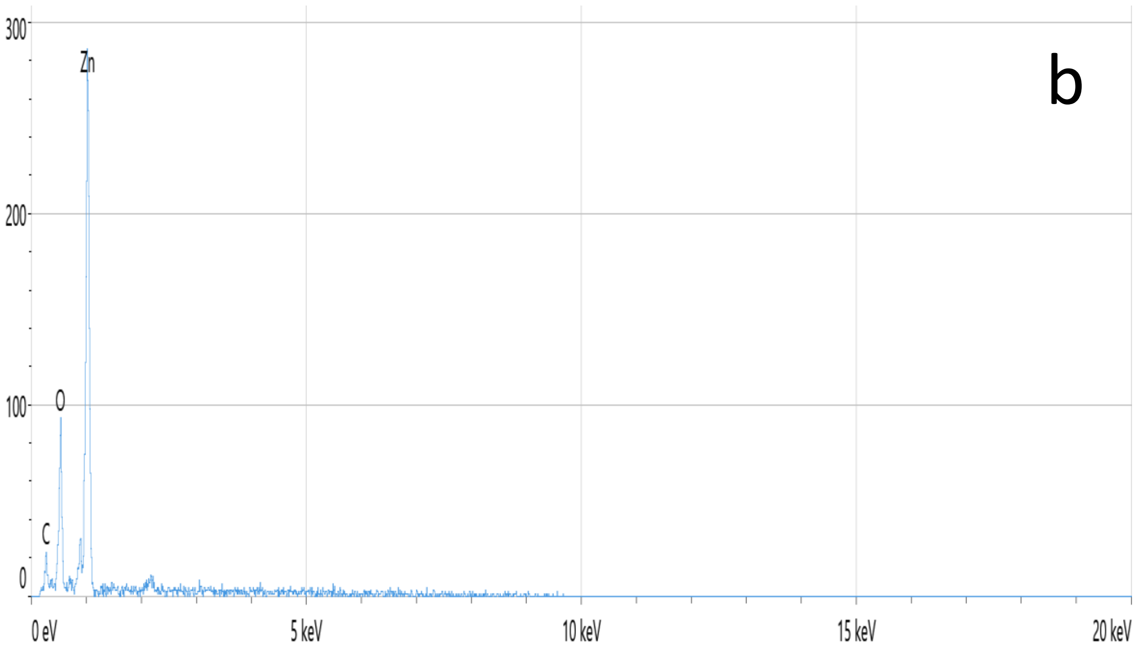


Figure 4: FTIR spectrum ZnO Nanoparticles at 9.0 × 10^{-1} M concentration.

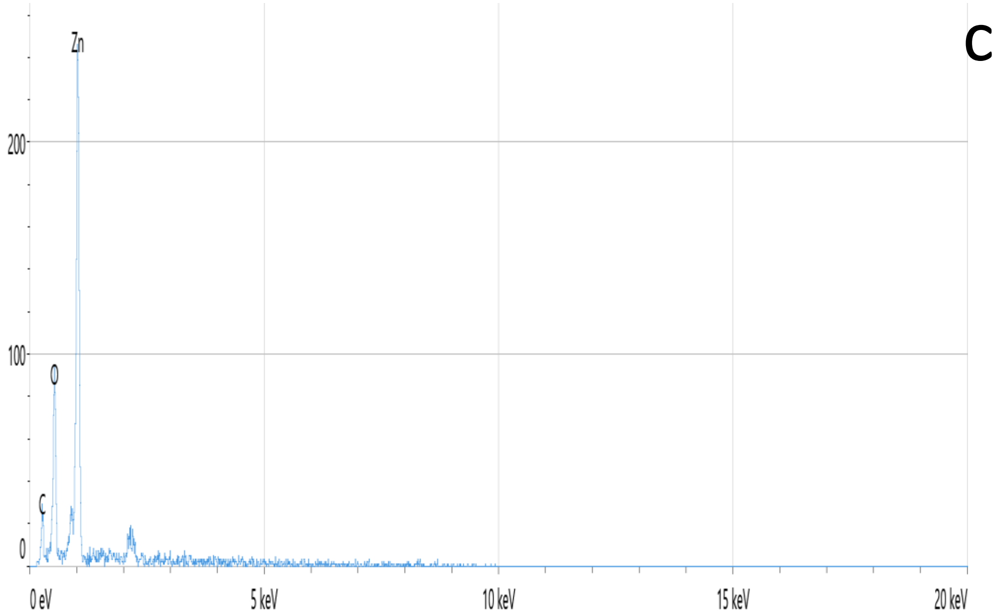
Element	Atomic %	Atomic % Error	Weight %	Weight % Error
C	14.9	0.7	4.6	0.2
O	36.9	0.7	15.1	0.3
Zn	48.2	0.5	80.4	0.8



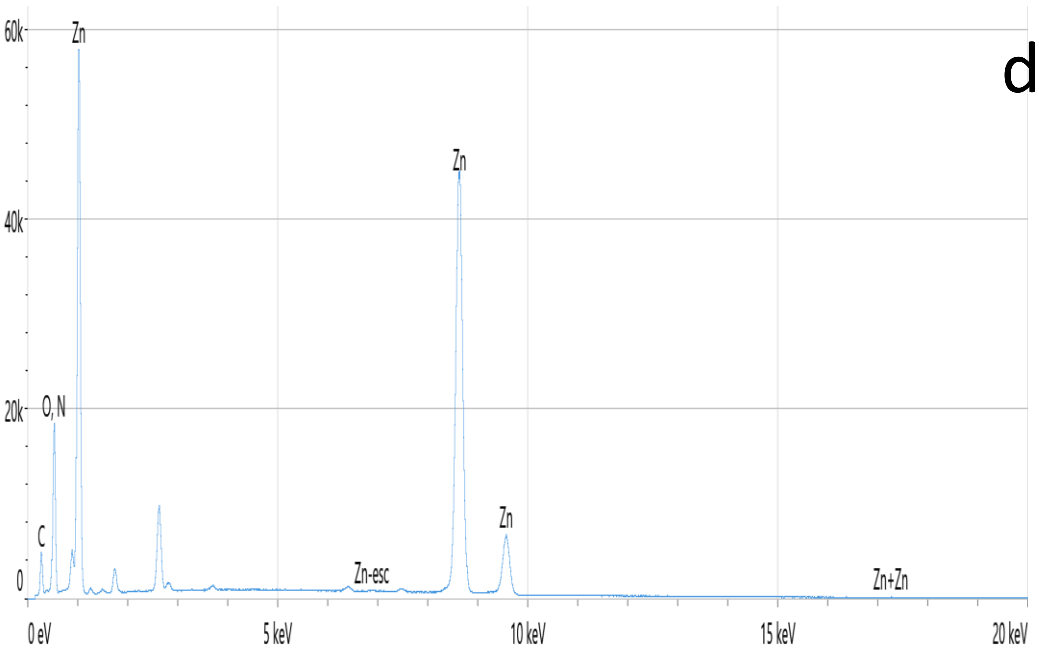
Element	Atomic %	Atomic % Error	Weight %	Weight % Error
C	16.0	1.5	4.9	0.5
O	36.0	1.9	14.7	0.8
Zn	48.1	1.1	80.4	1.8



Element	Atomic %	Atomic % Error	Weight %	Weight % Error
C	19.0	1.4	6.6	0.5
O	41.4	1.4	19.1	0.6
Zn	39.5	1.0	74.4	1.9



Element	Atomic 9%	Atomic % Error	Weight %	Weight % Error
C	26.0	0.3	10.2	0.1
O	40.6	0.3	21.2	0.1
Zn	31.7	0.1	67.7	0.2



Element	Atomic %	Atomic % Error	Weight %	Weight % Error
C	26.0	1.2	10.2	0.5
O	42.4	1.5	22.2	0.8
Zn	31.6	0.7	67.6	1.6

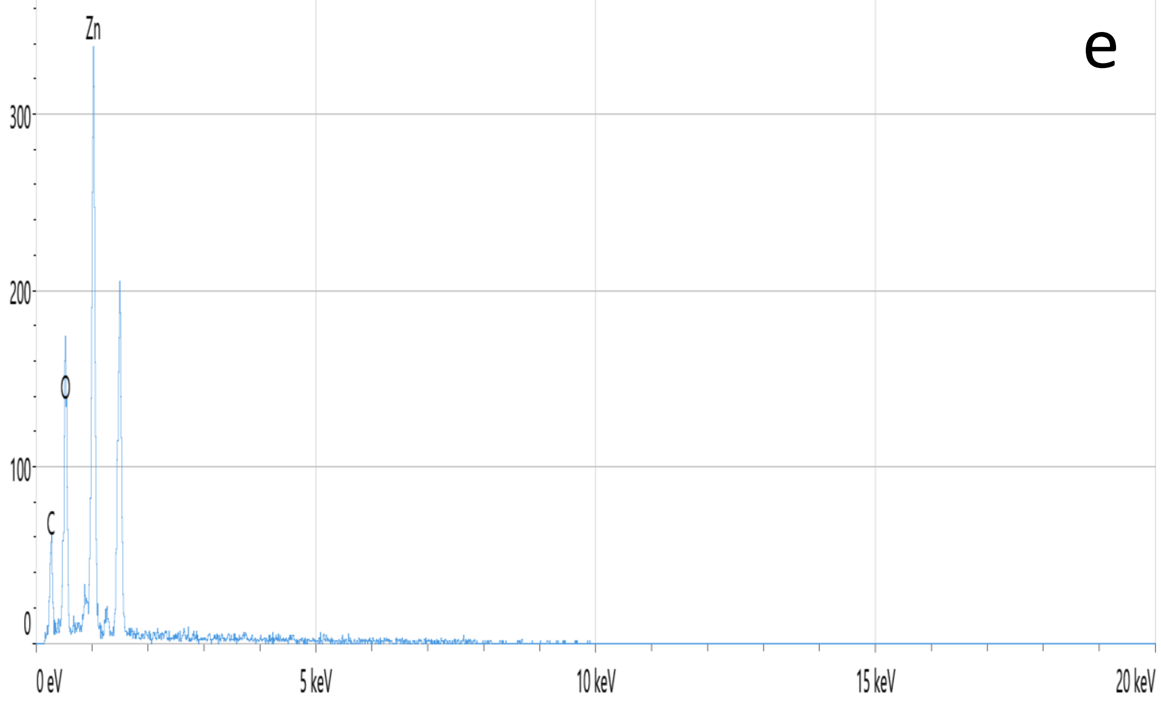


Figure 5: The EDX spectrum of ZnO Nanoparticles at different concentrations.

Figure (6) represents the absorbance of ZnO Nanoparticles colloidal was recorded as a function of ZnO Nanoparticles and it's increased with concentration; a result that agrees well with Beer-Lambert laws. It was observed that the peak in a range of (378 to 386) nm, and the absorption in this region is due to the formation of zinc oxide nanoparticles [26].

As concentrations increased, there was a red shift in the absorption peak. All of the samples showed an inclination to redshift, which was explained by the formation of shallow levels inside the band gap as a result of particle aggregation in the lattice [27].

The antibacterial activity of ZnO nanoparticles at various doses appears in Figure 7 (9.0, 5.0, 1.0, 1.1 and 1.6) × 10^{-1} M against *Staphylococcus aureus isolates*. The activity of ZnO Nanoparticles is related to the generation of free radicals induced by heat, which causes peroxidation when the radical interacts with the bacterial cell membrane. The resulting highly active oxygen molecules can oxidize organic compounds, leading to the disintegration and elimination of bacteria. The results showed that the concentration of ZnO Nanoparticles at 1.6 × 10^{-1} M had better antibacterial effects than other concentrations.

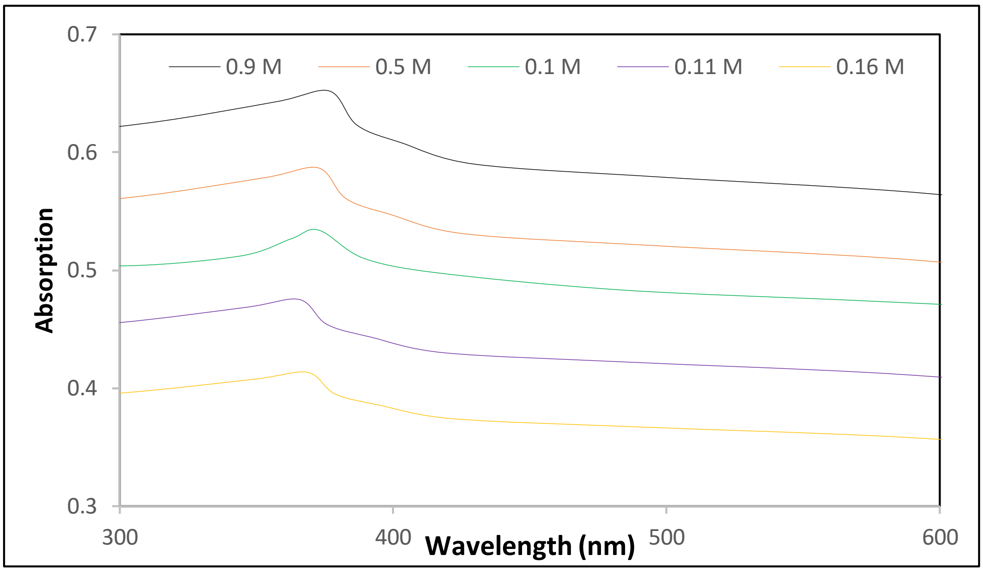


Figure 6: The absorption spectra of ZnO Nanoparticles at different concentrations

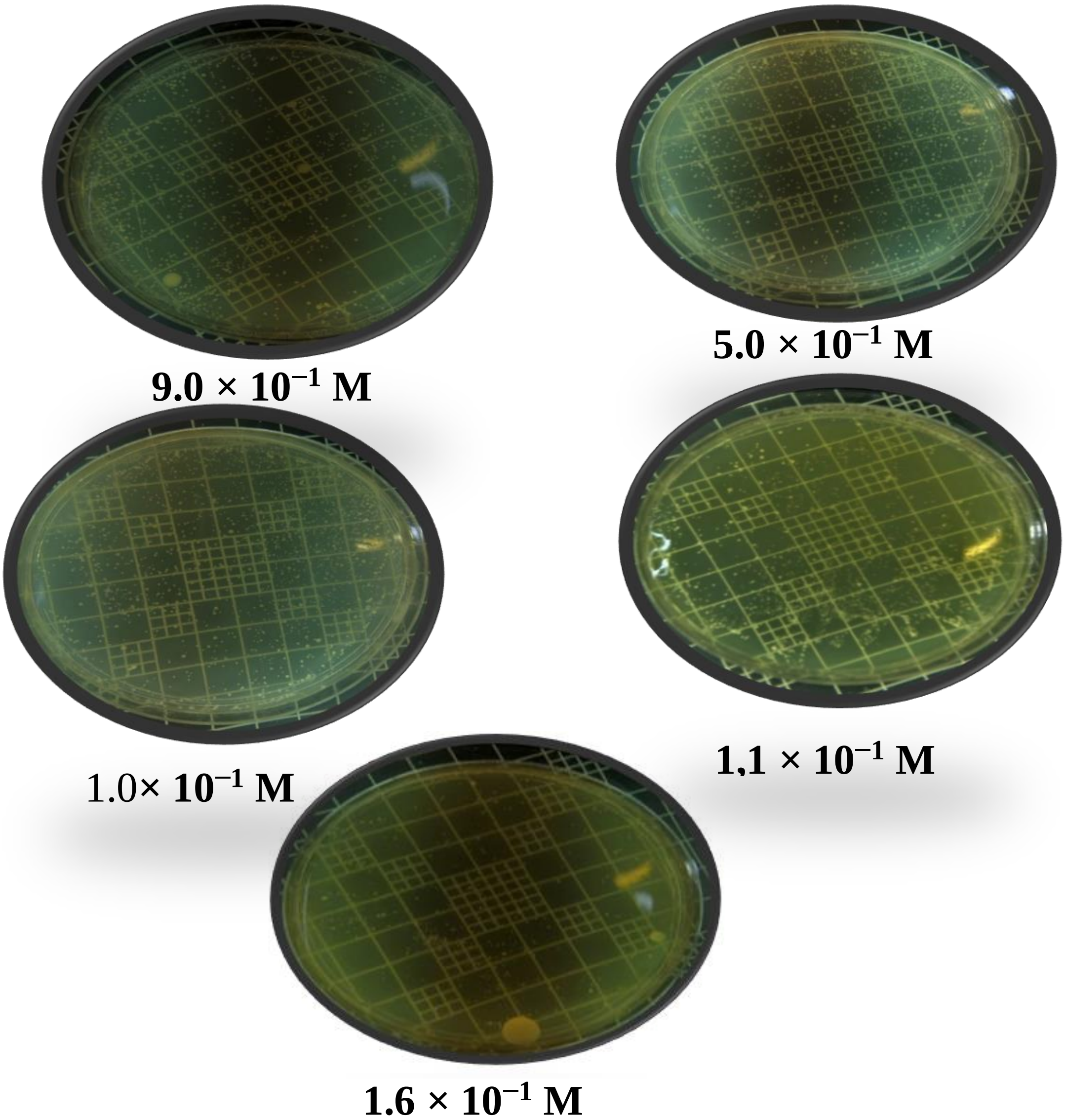


Figure 7: Antibacterial effect of Zinc oxide Nanoparticles at different concentrations

Conclusions:

The present research has indicated that a smaller and more uniform dispersion of nanoparticles of zinc oxide can be obtained at lower concentrations, in addition to the antibacterial effect of the lower concentration is 50% more than that of the other concentration.

Conflict of interest: NIL

Funding: NIL

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