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Biosynthesis and Characterization of Zinc Oxide Nanoparticles by Aloe vera Extract

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Abstract | The utilization of plant extracts in the synthesis of zinc oxide nanoparticles is recognized as one of the most environmentally friendly approaches. This study focuses on the green synthesis of zinc oxide nanoparticles (ZnO NPs) using Aloe vera leaf extract (ALE) as both the reducing and capping agents. The separation of this nanoparticle was performed by centrifugation while the identification was by UV-Visible spectroscopy, X-ray diffraction, Fourier Transmission Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM) and particle size, zeta potential analysis. White precipitate was formed after addition of zinc acetate to aloe vera extract indicating ZnO NPs formation. Particle size was recorded according to the data exhibited from the XRD results at 2θ around 41.3 nm which was calculated by using the Dubai-Scherrer equation. The ZnO NPs synthesized by the help of aloe vera extract were scanned using SEM. From the SEM image reveals that the ZnO NPs nanoparticle seems to be rod like in shape. From the results of the current study concluded that the green synthesis of ZnO-NPs using plant extract in a short time is cost-effective, safe and simple to obtain uniformed size ZnO-NPs crystal. The mean nanoparticles size was about 41.3 nm revealed the positive role of aloe vera extract as a capping agent and the size of the nanoparticles was also almost uniform. The XRD analysis revealed the purity of crystalline ZnO-NPs and the plant extract has the potential to contribute reduction reaction. The biosynthesis of zinc oxide nanoparticles (ZnO NPs) using aloe vera extract is a green and environmentally friendly method that relies on the plant's bioactive compounds as reducing and stabilizing agents. Aloe vera extract contains compounds such as phenols, flavonoids, and polysaccharides, which contribute to the reduction of zinc ions (Zn^{2+}) into stable nanoparticles.

Keywords | Nanoparticles, ZnO, Green synthesis, Aloe vera, FTIR, SEM, DLS

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INTRODUCTION

Nanotechnology is an arising area of research that deals with nano-scaled particles with dimensions mainly less than 100 nm1. Nanoscale dimensions provide the nanoparticle (NP) with a large surface area to volume ratio and thus highly unique properties including optical, electronic, and medicinal properties (Agarwal *et al*., 2017). Various methods can be used for the synthesis of NPs such as chemical, physical, and biological methods (Gupta *et al*., 2021). Zinc oxide (ZnO) is one of the important chemical compounds received much more attention in recent years due to its potential properties in various applications, such as photonics, cosmetics, pharmaceuticals and photocatalysis (Lee *et al*., 2019; Yusof *et al*., 2020) and as a potential antimicrobial property (Azizi *et al*., 2013). ZnO nanoparticles (ZnO-NPs) are synthesized using various methods, including sol–gel processes, ball milling, microemulsion and laser vaporization method (Abdolhoseinzadeh and Sheibani, 2020; Hernández *et al*., 2020). Generally, these nanoparticles preparation methods face several limitations, including, high-cost state of art experiments, man power, and required very large area for the installation of equipment's, and toxic chemicals, addition capping agent and stabilizers (Yusof *et al*., 2019). Almost all chemical methods are harmful to the environment because of the use of various chemicals for nanoparticles stabilization process and the chemicals bind with nanoparticles and affect various biological properties (Ielo *et al*., 2021). Whereas, nanoparticles synthesis by green-synthesis approach is generally considered as safe, bio-compatible and nontoxic to the organism or environment (Lakshmeesha *et al*., 2014). Additionally, green synthesis method has various advantages, such as simple method, less time consumed, low-cost and high purity. Several varieties of plant and fruit extracts were employed for the biosynthesis of ZnO-NPs, namely, Salvadora oleo ides, Echinacea spp, Boswellia ovalifoliolata, and Ocimum Americanum (Narendra *et al*., 2019). Additionally, many studies have proved that zinc oxide nanoparticles (ZnO-NPs) made using green synthesis processes have strong antibacterial and photocatalytic properties (Sedefoglu, 2023). It is well known that all bodily tissues, such as the brain, muscle, bone, and skin, contain significant amounts of zinc, an essential trace element. In addition to contributing to the body's metabolism, zinc serves as the primary component of numerous enzyme systems and is essential for the synthesis of proteins, nucleic acids, hematopoiesis, and neurogenesis (Saxena *et al*., 2010; Jo *et al*., 2015). Researchers have shown a huge interest towards the biosynthesis of NPs via the plant extracts 7. Plant-mediated synthesis of NPs exceeds other methods by several advantages as it is a reliable method, cost effective, simple, eco-friendly, reduced in toxic and hazardous products 8,9, and importantly, it needs no additional chemicals 10 (Duan and Li, 2013). The Aloe family Aloaceae, which includes about 400 distinct species, includes the genus Aloe one of the most prevalent and widespread species is aloe vera. 2009's Ombrella. The plant has thick leaves with long veins and a succulent form moving the water. When the green skin is removed, a clear mucus known as "gel" is left behind. This gel is packed with fiber, water, and vital components that help the paper retain moisture (Gurunathan, 2015). Aloe Vera used as general tonic for immune system, and its juice can potentially be used to treat various diseases (Jamdagni *et al*., 2018). It is effective in treating lung cancer, diabetes, rheumatic arthritis, ulcers, skin burns, and indigestion; it is used as an anti-inflammatory agent and has cosmological importance (Dobrucka and Długaszewska, 2016; Dar *et al*., 2021).

This study aimed to biosynthesis and characterization of ZnoNPs using Aloe vera extract.

MATERIALS AND METHODS

Plant sample collection

Fresh Aloe vera leaves were harvested from local garden in the city of Al-Hillah during the month of December 2024, and confirmation of their classification was done by the National Herbaceous, General Authority for Agricultural Research.

Preparation of plant aqueous extracts

The leaves were thoroughly washed with distilled water to remove dust and other impurities. After peeling the outer layer, 100 g of the leaf material was manually chopped into small pieces, then ground using an electric blender. The resulting mixture was transferred into a 250 ml glass conical flask containing 100 ml of distilled water. The mixture was placed in a water bath at 90 °C for one hour with continuous stirring using a hot plate with a magnetic stirrer to ensure proper homogenization. After heating, the solution was filtered using Whatman filter papers to remove solid residues and impurities. The obtained extract was collected and stored at room temperature for subsequent experimental applications (Wu *et al*., 2024; Mohammed *et al*., 2025).

Biosynthesis of zinc oxide nanoparticle's

Biosynthesis of ZnO NPs. In biosynthesis, 0.2 M zinc acetate precursor was prepared in 50 ml of deionized water. Aloe vera leaf extract (10 ml) was added drop wise under constant stirring using a magnetic stirrer for 10 minutes. In order to adjust the pH of the prepared solution, NaOH (2M) was added drop wise while stirring. The obtained white crystalline precipitate was washed well with distilled water, filtered and dried in an oven at 60 °C. The process ultimately ends with the formation of a white smooth powder (Prakash *et al*., 2022).

Zinc oxide nanoparticles biosynthesis

Zinc oxide Nanoparticles of the Aloe vera extract were prepared according to (Vidya *et al*., 2013; Bayda *et al*., 2019). Method by heating 50 ml of the water extract using a hot plate at a temperature of 40-45 °C. At a temperature of 45 °C, add 5 grams of zinc acetate to the solution and continue heating until it becomes a bright yellow paste. The dough was gathered in a glass petri dish and heated at 300° C for two hours to dry it out. The dried substance was then broken down into a light-yellow powder using a mortar, which was then carefully packaged for further characterization and treatment needs.

Separation and Identification of ZnONP

This work was done in Al-Qasim Green University, veterinary medicine Laboratories, Department of physiology, biochemistry and pharmacology.

Spectrophotometry: the stability and formation of Zno-NPs was monitored with ultraviolet -visible spectrophotometer apparatus in wavelength ranged from (300-600) nm. For the UV-Vis spectral analysis. Absorbance of this solution was measured at one hour interval for 1-24 hrs and the changing in the color was observed gradually as it turned deep brown at the end of 24 hrs (Awwad *et al.*, 2013).

X-Ray Diraction (XRD): Solution of the developed ZnoNPs was centrifuged at 10,000 rpm for 30 min. The solid residues of ZnoNPs were washed twice with deionized distilled water and then dried at 80 °C to obtain powder ZnoNPs used for X-ray powder diffraction measurements. The powder X-ray diffraction (XRD) patterns were recorded on (Shimadzu XRD-6000) with copper radiation (Cu Kα, 1.5406 Å) at 40 kV and 30 mA (Bykkam *et al.*, 2015).

Fourier Transform Infrared Spectroscopy (FTIR): FTIR measurements were carried out using (FTIR) Spectroscopy, ABB-Specro-Lab-MB3000, UK) the range from 3500 cm-1 to 500 cm-1. After complete reduction of ZnoNO ions by silymarin fruit extract, the mixture was centrifuged at 10000 rpm for 10 min to remove protein or other bioorganic compounds that were present in the solution. The zinc oxide nanoparticles pellet obtained was air dried. The dried nanoparticles were mixed with the potassium bromide (KBr) to made thin pellets and were used for FT-IR analysis in transmittance mode (Kalaiarasi *et al.*, 2013; Abdul-Ameer *et al.*, 2024).

Scanning electron microscopic analysis: There were no reported differences in the shapes and size of ZnoNPs at various initial biomaterial concentrations (Huang and Yang, 2004). So, the samples for electron microscopy were prepared from the extract and various time of reactions. The biomasses had settled in the base of the cone like carafes and the suspension over the accelerate was tested for scanning electron microscopy (SEM) perception. Scanning electron microscopy of the aqueous solution samples of ZnoNPs prepared by setting a one drop of the solution on the carbon-covered copper grids and the films on the SEM system permitted standing for two minutes, after that there is removing of the extra solution by using a blotting paper and drying the grid. The size appropriation of the subsequent nanoparticles evaluated on the basis of SEM micrographs (Naik *et al.*, 2013).

Particle size (PS) and Zeta potential (ZP) analysis

Dynamic light scattering (DLS) technique used to evaluate the particle size, zeta potential and polydispersity index. The dried ZnO NPs were reconstituted in distilled water. The size of NPs was determined by Zeta sizer based on DLS technique. Zeta potential, an indicator of surface charge, which determines particle stability in dispersion, was also measured using the principle of electrophoretic mobility in an electric field. The PDI which is a dimensionless number indicating the width of the size distribution, was also measured.

RESULTS AND DISCUSSION

A noticeable and verifiable change was observed in the bio reduction medium following the addition of Zinc acetate to Aloe vera (barbadensis Miller) extract. To regulate the particle size of ZnNPs, Zn (CH3COO)2 was incorporated into the aqueous leaf extract and incubated at 65 ˚C at a pH of 12. The synthesis was validated through visual observation of the Aloe vera extract and zinc acetate as shown in Figure 1.

Figure 1: Green synthesis of zinc oxide nanoparticles. Fresh Aloe vera leaves were collected. The leaves were washed and cut for preparing the extract. The extract was added to the zinc acetate solution. NaOH was slowly added to the mixture until the color of the mixture turned from clear bright yellow to a little turbid and a white precipitate began to form. White ZnO NPs were totally precipitated. White powder of ZnO NPs was obtained after calcination at 400 °C.

The combination of leaf extract and Zn (CH3COO)2 exhibited a color transformation over time, progressing from colorless to pale yellow after 30 minutes, followed by a slight turbidity and the formation of a white precipitate upon the addition of NaOH. No color change was observed during the incubation of Aloe vera extract in the absence of Zn (CH3COO)2. These findings aligned with previously published reports (Albarakaty *et al.*, 2023) also noted a white powder color while synthesizing zinc oxide nanoparticles from aloe vera peel extract. In another study, (Rajendran *et al.*, 2021). have reported the synthesis of pale white zinc oxide nanoparticles utilizing *Rubus fauxhemians* root extract.

To confirm the synthesis of ZnO NPs, UV/Vis spectrophotometry was performed in order to examine the optical characteristics of green synthesized ZnO NPs using Aloe vera extract. The UV–Vis absorption curve of ZnO nanoparticles is shown in Figure 2. Zinc oxide formation was confirmed as the absorption peak (lambda max) was found near 240.14 nm using a UV–Vis spectrometer Shimadzu-UV 1600 with distilled water as a reference.

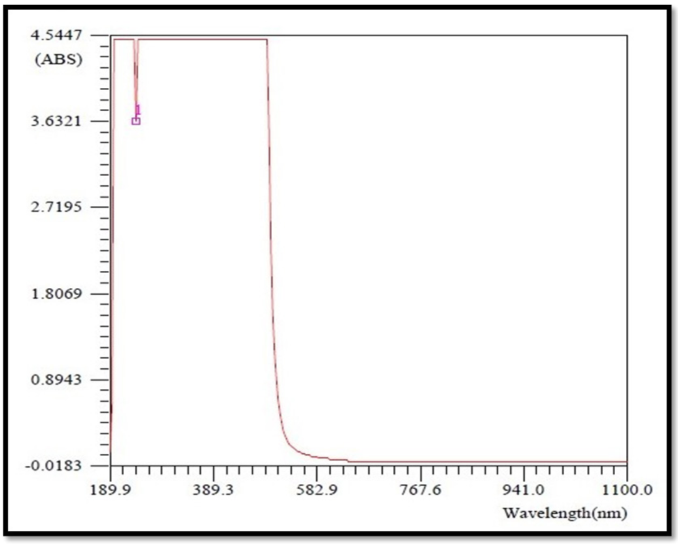

Figure 2: UV/Vis spectrum of ZnO NPs biosynthesized using Aloe vera extract.

The peak of absorption at 240.14 nm confirms the formation of ZnO NPs, because ZnO typically shows absorption in the UV spectrum due to the electronic transition from the valence band to the conduction band. The recorded value indicates that the particles are nano-sized and have a large energy gap, which reflects the small size and increased surface-to-volume ratio. The findings of the present study concur with those of the earlier research by Chaudhary *et al.* (2019) which synthesized zinc oxide nanoparticles from A. vera and demonstrated a maximum absorbance at 240 nm. Conversely, (Shekhawat *et al.*, 2014). Synthesised zinc oxide nanoparticles from *H. enneaspermus* and noted an absorption peak at 300 nm for the leaf extract, 290 nm for the stem extract, and 288 nm for the root extract.

The crystalline property of synthesized ZnONPs was analyzed by XRD. Figure 3 display the XRD diffractogram observed at 2θ showed numerous peaks including low, medium, and high-intensity peaks. Synthesized ZnO nanoparticles' XRD pattern revealed several peaks that corresponded to several crystal planes, indicating that the sample has a crystalline structure. The observed peaks at 2θ values of 18.75◦, 23.45◦, 26.28◦, 35.60◦, 39.64◦, 47.92◦, 52.38◦ and 65.91◦ can be assigned to the (100), (002), (101), (110), (103), (200), (004) and (203) crystal planes of ZnO nanoparticles. According to the data analysis and by using Debye-Scherrer formula; the average crystallite size is approximately 41.3 nm.

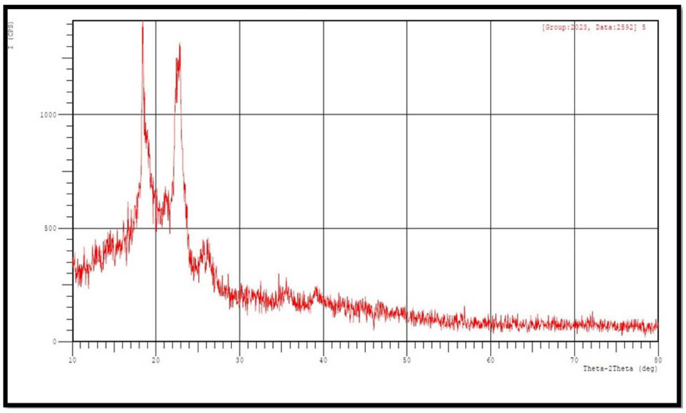

Figure 3: XRD patterns of green synthesized ZnO nanoparticles.

These strong peaks indicated the crystalline nature of zinc oxide nanoparticles. In previous study, (Rajiv *et al.*, 2013). synthesized zinc oxide nanoparticles from leaf extract of *P. hysterophorus* L. and reported Miller indices values at (100), (002), (101), (102), (110), (112) and (202), which also confirmed the crystalline nature of nanoparticles. In another study, (Jayarambabu *et al.*, 2015) reported crystalline nature of zinc oxide nanoparticles from green crops with XRD peaks and Miller indices values at 31.7° (100), 34.5° (002), 36.2° (101), 47.7° (102), 56.6° (110), 62.2° (103) and 68.4° (112).

FTIR is used to identify the different operative groups involved in the production of ZnONPs. The dual role of the plant extract, as a reducing as well as capping agent, and presence of some functional groups in both the aloe vera extract and ZnO Nps were investigated by FTIR analysis. The FTIR spectra of the powder samples are displayed in Figure 4. A series of absorption peaks in the range from 4000-400 cm^{-1} have been found. The absorption peaks at 1419, 1374 and 1341 cm^{-1} correspond to the C-N stretching vibration. The strong intensity peaks at 1240, 1143, 1080 and 1021 cm^{-1} are due to the C-O-C stretching bond. The bands at 964, 943, 842, 855, 761 cm^{-1} are attributed to –CH stretching vibration; while 668 cm^{-1} attributed to the alkynes (-C=C-H stretching vibration). The absorption band observed at 575, 547, 502, 518, 455, 474, 442 and 420 cm^{-1} confirmed the successful formation of Metal-Oxygen (ZnO).

A shift of peaks was observed between FTIR spectrum revealed the interaction of various functional groups of the phenols and flavonoids with the green synthesized ZnO-NPs. The peaks at 1374 cm^{-1}, 1341 cm^{-1} and 842 cm^{-1} correspond to C-O and CH vibrations, confirming the presence of organic compounds from Aloe vera extract. The most significant peak at 547 cm^{-1} corresponds to Zn-O stretching vibrations, providing direct evidence for the successful formation of ZnO nanoparticles. The available functional group from the plant extract donated electrons and these electrons reduced zinc ions (Zn2+) to Zn+1 and finally reduced as zinc NPs. The plant extract contains negative functional group and working as a stabilizing agent. The peak observed around 1000 cm1 and below is ascribed to the existence of metal oxide (Figure 4). In general, the meal oxides show very strong absorption bands with the wave number < 1000 cm^{-1} (Bhuyan *et al*., 2016; Ngom *et al*., 2016). The ZnO was formed from aloe vera leaf extract where the free carboxylic and the amino group of the plant extract acted as both reducing and capping agent (Agarwal *et al*., 2017). The results is was confirmed with the outcomes reported by another researchers when synthesize ZnO nanoparticles using aloe Vera extract (Rasli *et al*., 2020; Rehman *et al*., 2025).

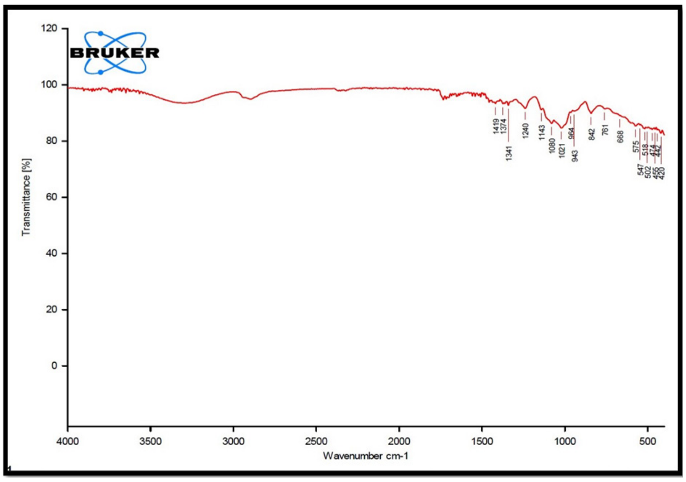

Figure 4: FTIR spectrum of ZnO nanoparticles.

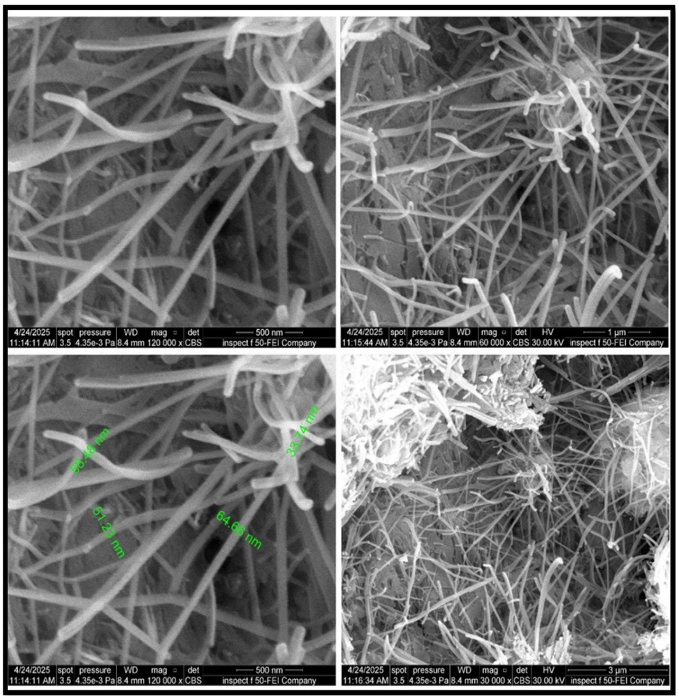

Figure 5: SEM images of rod-like green synthesized ZnO nanoparticles at different magnification power.

Scanning electron microscope is used to examine the surface morphology of the nanoparticles. The SEM images revealed the rod like morphology with good uniformity and some agglomerated particles at different magnification. The shape of the ZnO nanoparticles is rod-like with a particle size ranging from 33.7 to 64.6 nm, Figure 5.

The ZnO nanoparticles appear as randomly dispersed, rod-like, indicating minimal agglomeration and a stable synthesis process. This suggests that Aloe vera extract effectively acted as a reducing and stabilizing agent during nanoparticle formation. The agglomeration could be induced by the densification and microstructural changes resulting in the narrow space between particles and also decreased pore size and diameter (Sangeetha *et al*., 2011). The SEM results of Aloe Vera ZnO-NPs are similar to previous studies on green ZnO-NPs (Chaudhary *et al*., 2019; Patı *et al*., 2021).

Particle size diameter of the green synthesized ZnONPs were measured using the DLS technique. As shown in Figure 6, the measurements demonstrated that the average size (nm) of the ZnO NPs synthesized with Aloe vera extract was about 61.8 nm. The result obtained from the PSD profile of the ZnO nanoparticles revealed two notable peaks. Additionally, the ZnO NPs have a polydispersity index (PDI) of 0.286. The PDI value of <0.5 indicated homogeneous distribution and uniform particle sizes. The DLS result come true with (Kumar *et al*., 2014) who synthesize ZnO NPs by the aqueous peel extract of *C. paradise*. And recorded mean particle size of ZnO-NPs 76.5 nm.

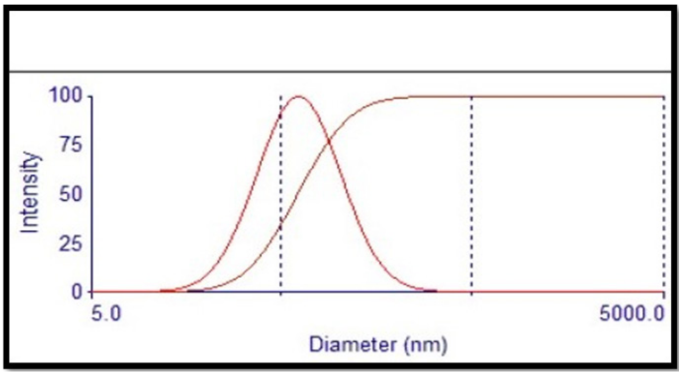

Figure 6: Particle size distribution using DLS for biosynthesized ZnO NPs.

Figure 7 illustrated the zeta potential of ZnO NPs. The amplitude of the zeta potential gives an indication of the colloidal system's potential stability. The stability degree according to zeta potential values was detected as follows: ±0-10 mV is very unstable; values in the ranges of ±10-20 mV are not very stable; ±20-30 mV is stable; and values greater than ± 30 mV are highly stable (Bhattacharjee *et al*., 2016). The zeta potential result for ZnO NPs synthesized

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in the current study indicates that the surface charge was negative. The zeta potential peak in the graph of this study around -28.7 mV, and most particles in the synthesized solution have a negative zeta potential value, and this provided high stability. The negative value could be related to the presence of the reducing agent's polyphenolic and flavonoid compounds in the plant extract, which displays electrostatic forces in green-synthesized NPs (Nasrollahzadeh *et al*., 2016). The effective stability of ZnO NPs and reduction of metal ions may be due to the extract's high protein and flavonoid content.

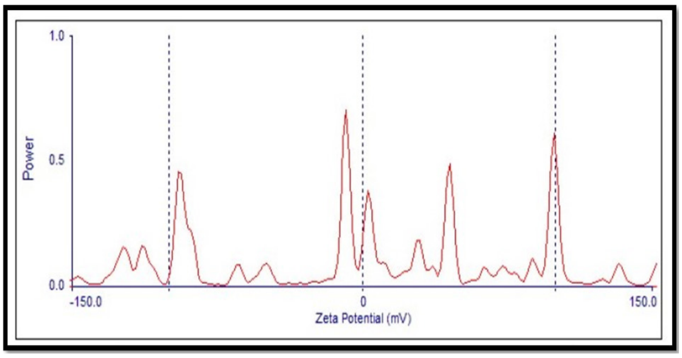

Figure 7: Zeta potential of green synthesized zinc oxide nanoparticle.

The present finding clearly revealed that the capping agent from the plant source is circled with ZnO-NPs. Naturally, the Aloe Vera contains rich of flavonoids and proteins and these phytochemical components involved in the reduction of metal ions and good stabilization of ZnO-NPs. Our findings were highly consistent with (Kaur *et al*., 2022).

CONCLUSIONS

Green synthesis of ZnO-NPs using plant extract in require a short time and it is cost-effective, safe and simple to obtain uniformed size ZnO-NPs crystal. The mean nanoparticles size was about 41.3 nm revealed the positive role of aloe vera extract as a capping agent and the size of the nanoparticles was also almost uniform. The biosynthesis of zinc oxide nanoparticles (ZnO NPs) using aloe vera extract is a green and environmentally friendly method that relies on the plant's bioactive compounds as reducing and stabilizing agents. Aloe vera extract contains compounds such as phenols, flavonoids, and polysaccharides, which contribute to the reduction of zinc ions (Zn^{2+}) into stable nanoparticles.

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NOVELTY STATEMENT

The present study reports the eco-friendly synthesis of zinc oxide nanoparticles (ZnO NPs) employing Aloe vera leaf extract as a dual reducing and capping agent. This biogenic approach offers a simple, cost-effective, and non-toxic alternative to conventional chemical synthesis routes, thereby aligning with sustainable nanotechnology practices. The phytochemicals present in Aloe vera, particularly phenolic compounds and flavonoids, facilitated the reduction of zinc ions and stabilized the resulting nanostructures. Unlike many previous studies, the current work successfully produced highly uniform, rod-shaped ZnO NPs with an average size of ~41.3 nm, exhibiting excellent crystallinity and stability. These characteristics underscore the effectiveness of Aloe vera-mediated synthesis in generating high-quality nanoparticles with potential for large-scale applications in biomedical, environmental, and industrial fields. Collectively, this green synthesis approach demonstrates that Aloe vera can serve as a sustainable bio factory for the scalable production of functional nanomaterials.

AUTHOR'S CONTRIBUTION

All authors worked equally for experimental design, writing, editing, and final version proofing of this research paper.

Generative AI and AI-assisted technology statement

AI doesn't used for writing of this research paper.

Conflict of interest

The authors have declared no conflict of interest.

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