

A Role of Biosynthesized Zinc Oxide Nanoparticles (ZnO NPs) For Enhancing Seed Quality: A Review

ABSTRACT

Abiotic and biotic stresses impact seed growth, resulting in economic losses. Seeds are being subjected to an increasing number of biotic and abiotic stress combinations as a result of global warming and climate change, which has adverse effects on their growth and production. Drought, flood, salinity, heavy mineral contamination, cold, and heat were all found to have a negative impact on seed germination. Because of their massive surface area-to-volume ratio, nanoparticles—microscopic pieces with a nanoscale dimension ranging from 1-100 nm—have exceptional thermal conductivity, catalytic reactivity, nonlinear optical performance, and chemical durability. There are various methods for creating nanoparticles, such as chemical, physical, and biological ones. However, because dangerous chemical compounds are used as reducing agents, the chemical and physical procedures are expensive, complicated, and might be hazardous to the environment. The synthesis of nanoparticles using green approaches may be easily scaled up, and they are also cost-effective. Because of their superior qualities, greenly coordinated nanoparticles are currently preferred over traditionally delivered NPs. Green synthesis approaches are particularly appealing due to their ability to reduce nanoparticle toxicity. The use of vitamins, amino acids, and plant extracts has increased as a result. While dangerous and extremely hazardous substances are used in the chemical and physical processes that may cause environmental issues, capping and reducing agents are essential to the synthesis of nanoparticles. The capping or reducing agent used in physical and chemical processes is expensive. When used as a seed treatment, nanoparticles can enhance germination as well as the length, vigor, viability, and quality of the seedlings. This paper aims to provide an overview and evaluation of zinc oxide nanoparticles (ZnO NPs) as a potential substitute for biosynthesised nanoparticles in seed quality improvement.

Keywords : Nanoparticles; zinc oxide nanoparticle; green synthesis; germination; vigour; viability and seed quality.

1. INTRODUCTION

Nanotechnology is a novel and rising area with the goal at creating unique materials at the nanoscale scale (Mohammad *et al.*, 2022). Nanomaterials, as compared to materials with indeterminate particle sizes, are made up of small particles having a large specific surface area, volume, quantum size, and macro tunneling effects. Considering for these qualities, nanomaterials have unique optical, mechanical, catalytic, and biological capabilities, giving them a wide range of applications (Guo *et al.*, 2018). Due to their high specific surface area, biocompatibility, ultraviolet light absorption and scattering, ZnO NPs are widely employed as a metallic nanomaterial in industries such as electrochemistry, medical devices, cosmetics, and the textile industry. It also demonstrates biological activity, such as antimicrobial features. (Ahmad *et al.*, 2017 and Saif *et al.*, 2019). ZnO NPs are typically synthesized using physical and chemical methods, both of which have problems such as high energy consumption, low purity, uneven particle size distribution, high cost, a huge quantity of secondary waste, and irreversible negative environmental effects. As the number of applications for ZnO NPs increases, there is growing concern over their synthesis using sustainable methods, owing

to the fact that the concept of environmental protection is profoundly established in the public's expectations (Xu *et al.*, 2021)

Green synthesis approaches involve the employment of microbes, enzymes, and plant extracts in the synthesis method. There are no toxic components required, and the process requires minimal resources. The benefits include environmental sustainability, eco-friendliness, and low cost, making it a desirable substitute to traditional physical and chemical procedures. (Bhalerao and Borkar, 2017, Khan *et al.*, 2018). Plants and their extracts are readily available, and the technique only requires a zinc salt solution as a metal precursor of zinc nitrate, zinc acetate and zinc chloride. ZnO NPs are manufactured by reacting plant extracts with zinc salt solution (zinc sulfate heptahydrate), which is an ecologically sound technique for synthesis of ZnO NPs. Several studies have demonstrated that extracts from plant leaves can act as both reducing and stabilizing agents in the synthesis of ZnO NPs (Anbuvannan *et al.*, 2015; Vijayakumar *et al.*, 2015; Gemachu and Birhanu, 2023). The green synthesis of ZnO NPs from plant extracts produces excellent antibacterial activity against a wide range of bacteria (Guy and Ozacar, 2016, Podasca *et al.*, 2016, and Sohrabnezhad and Seifi, 2016).

Abiotic and biotic stresses impact seed growth, resulting in economic losses. Seeds are being subjected to an increasing number of biotic and abiotic stress combinations as a result of global warming and climate change, which has adverse effects on their growth and production. Drought, flood, salinity, heavy mineral contamination, cold, and heat were all found to have a negative impact on seed germination. Bacteria, fungus, viruses, nematodes, insects, and other plant pathogens can contribute to biotic stress. Pathogen infection frequently causes changes in plant physiology, including reduced biomass, early blooming, decreased seed set, buildup of defensive chemicals, and a variety of other modifications (Chojake *et al.*, 2018). Researchers have found that a range of nanomaterials minimize biotic and abiotic stress and promote seed germination (Aslani *et al.*, 2014; Ansari *et al.*, 2020).

2. PROPERTIES OF ZINC OXIDE NANOPARTICLES (ZnO NPs)

Zinc oxide, a non-hygroscopic and nontoxic inorganic, polar, crystalline material, is very cheap, safe, and widely available, which has created significant interest in various organic transformations, sensors, transparent conductors, and surface Acoustic waves appliances (Gorla *et al.*, 1999; Tayeb *et al.*, 2010; and Bahrami *et al.*, 2011). ZnO NP is a unique material with semiconducting, piezoelectric, and pyroelectric properties that recognizes use in transparent electronics, ultraviolet (UV) emitters, sensor materials, spin technology, cosmetic products, catalysts, as well coatings, and paints (Akhtar *et al.*, 2011; Sasidharan *et al.*, 2013). ZnO NPs are used in antireflection coatings, electrodes with transparency in solar panels, UV light producers, diode lasers, variants, spin electronics, surface acoustic wave propagators (Tayeb *et al.*, 2010), as an antibacterial agent (Zhang *et al.*, 2008), as a photonic material (Xie *et al.*, 2006), and for gas sensing (Liewhiran and Phanichphant, 2007).

3. MECHANISM OF THE BIOSYNTHESIS OF ZnO NPs FROM PLANT EXTRACTS

The plant antioxidants, including polysaccharides, polyphenols, flavonoids, vitamins, amino acids, alkaloids, tannins, saponins, and terpenoids, are reductive. These plant extracts utilized as reducing and capping agents which reacting with zinc salt solution to produce ZnO NPs (Agarwal *et al.*, 2017, Liu *et al.*, 2019, and Yang *et al.*, 2023).

4. CHARACTERIZATION OF BIOSYNTHESISED ZnO NPs

Several techniques are used to characterize the synthesized nanoparticles, including FTIR (Fourier transform infrared spectroscopy), EDAX (energy dispersion analysis of X-ray), AFM (atomic force microscopy), XPS (X-ray photoelectron microscopy), ATR (attenuated total reflection), UV-DRS (UV-visible diffuse reflectance spectroscopy), XRD (X-ray diffractometer), TEM (transmission electron microscopy), TG-DTA (thermogravimetric-differential thermal analysis), DLS (dynamic light

scattering), FE-SEM (field emission scanning electron microscopy), PL (photoluminescence analysis), Raman spectroscopy, and SEM (scanning electron microscopy) (Arfat *et al.*, 2014, Yasmin *et al.*, 2014, Rajeshkumaret *et al.*, 2016, Ramesh *et al.*, 2021 and Urge *et al.*, 2023).

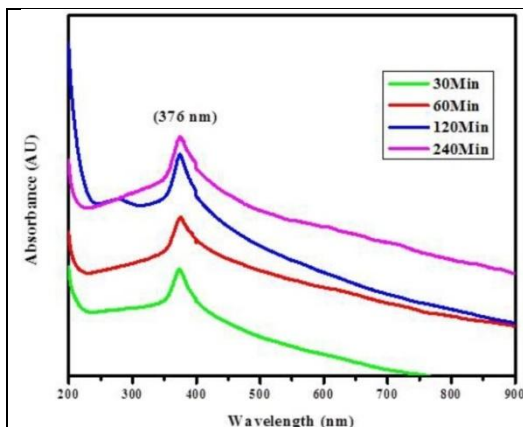


Fig. 1 UV-Vis spectra of biosynthesized ZnO NPs (Ramesh *et al.*, 2021).

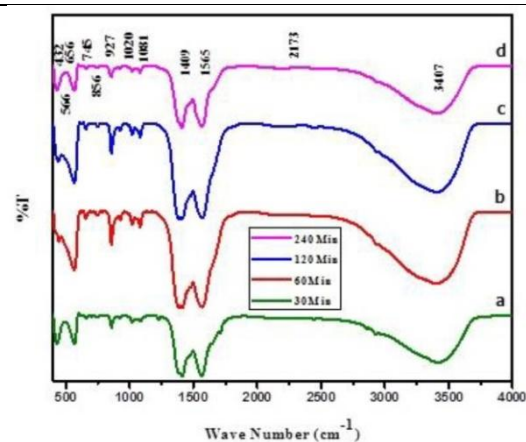


Fig. 2 FTIR spectra of biosynthesized ZnO NPs

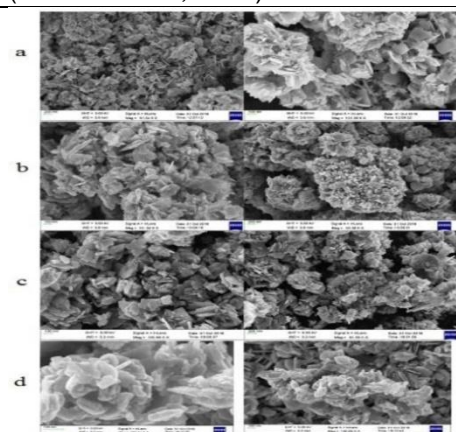


Fig. 3 FE-SEM Spectra of biosynthesized ZnO NPs(Ramesh *et al.*, 2021).

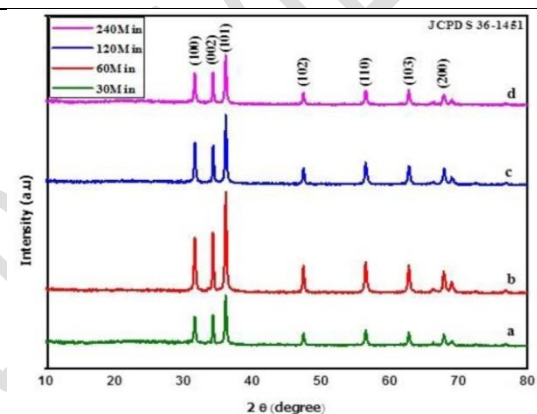


Fig. 5 XRD pattern of biosynthesized ZnO NPs (Ramesh *et al.*, 2021).

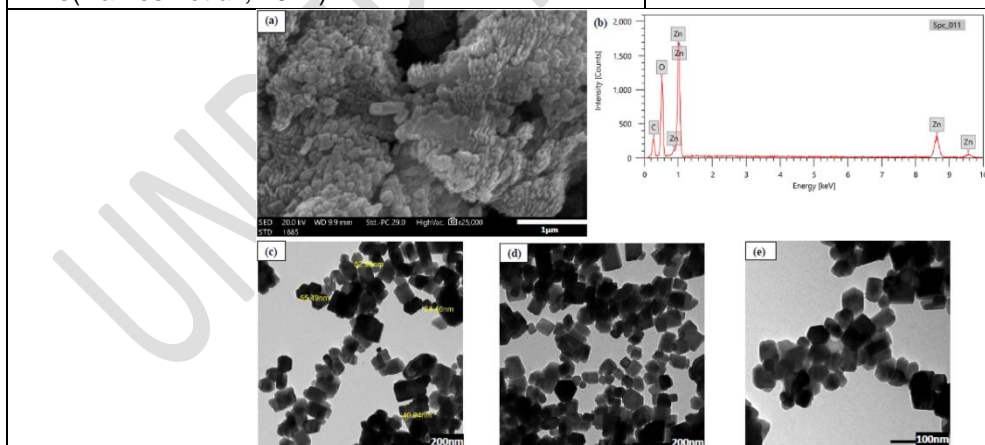


Fig. 6 (a) SEM; (b) EDX; (c–e) TEM images of the biosynthesized ZnO NPs. (Naiel *et al.*, 2022)

4. BIOSYNTHESIS OF ZINC OXIDE NANOPARTICLES (ZnO NPs) BY USING LEAVES EXTRACT:

The most common technique of preparing ZnO NPs from plant extracts involves thoroughly washing the plants with sterile or distilled water. Plant extracts are subsequently created by drying, grinding into powder, dissolving in solvent, or directly soaking. The extracts are then mixed with zinc

salt solution as a metallic precursor, producing a precipitate following reaction. Finally, calcining the precipitate produces ZnO NPs (Figure 7).

The synthesis of biological nanoparticles offers an alternative to physical and chemical methods of nanoparticle synthesis. Most of investigators worked on the green synthesis of nanoparticles to produce metal and oxide nanoparticles. Plant-based nanoparticle synthesis is a quick, low-cost, sustainable approach that is also safe for humans and agriculture (Raveendran *et al.*, 2003).

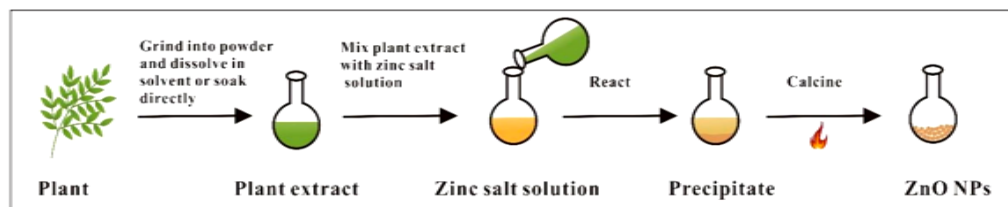


Figure 7. Process of green synthesis of ZnO NPs from leaves extracts. (Jun Xu *et al.*, 2021)

Several plant extracts have been reported to synthesize zinc oxide (ZnO) nanoparticles. For example, Raut *et al.*, (2013) used the leaf extract *OcimumTenuiflorum* as a reducing agent in the green synthesis of ZnO nanoparticles. The prepared ZnO nanoparticles were analyzed using X-Ray diffraction (XRD), scanning electron microscopy (SEM), and Fourier Transform Infrared Spectroscopy (FTIR). The average particle size is determined as 13.86 nm using Scherrer's formula.

Saravanak Kumar *et al.*, (2016) described a new sustainable approach for the production of ZnO nanoparticles at ambient temperature, in which Ajwain (Carom - *Trachyspermumammi*) seed extract acts as a zinc salt reduction agent. The UV-Vis spectrum of the produced ZnO nanoparticles displays strong absorption in the ultraviolet area at around 383.5 nm, indicating that the material is acceptable for UV filters. The scanning electron microscope analysis demonstrates that the crystal has a hexagonal shape. The average crystallite size is 34.27 nm.

Shah *et al.*, (2019) assess bioaugmented zinc oxide nanoparticles (ZnO NPs) derived from *Myristica fragrans* aqueous fruit extracts. UV-vis, XRD, FTIR, SEM, TEM, DLS, and TGA were all used to characterize ZnO NPs. The crystallites had a mean size of 41.23 nm assessed by XRD and were extremely pure, while SEM and TEM examinations of produced NPs confirmed their spherical or elliptical shape.

Yasotha *et al.*, (2020) reported the production and characterization of ZnO nanoparticles using the green synthesis approach. The capping agent from *Ocimumtenuiflorum* leaf extract was used as a reducing agent in the production of ZnO nanoparticles. The produced ZnO nanoparticles were analyzed by XRD, FT-IR, SEM, and EDAX. The obtained results show that the crystalline size is 18.53 nm, the morphology and content are consistent with the standard values, and it will be beneficial for antibacterial applications.

Elsamraet *et al.*, (2022) produced nano-ZnO by reducing Zn salt with an extract of *Ocimumtenuiflorum* leaves. The produced ZnO NPs were characterized by FT-IR, XRD, SEM, and EDX methods. The described XRD data indicated the production of a hexagonal wurtzite structure. SEM pictures revealed the spherical nature which had an average diameter of 19 nm.

Mishra *et al.*, (2023) attempted to biosynthesize zinc oxide nanoparticles (ZnO NPs) from *Catharanthus roseus* (L.) G. Don leaf extracts and use them as a nanoprimer agent to improve seed germination and seedling growth in *Eleusine coracana* (L.) (finger millet). UV-Vis., FTIR, FE-SEM, EDX, and TEM have been used for assessing biosynthesized nanoparticles (NPs). The peaks at 362

nm characterized the UV-Vis spectra of ZnO NPs. The FTIR absorption spectra of ZnO NPs revealed Zn-O bending at 547 cm^{-1} . The size (44.5 nm) and form (nonspherical) of ZnO NPs were determined by TEM image analysis. XRD revealed the hexagonal wurtzite phase of ZnO, with an average particle size of 35.19 nm.

Table 1 :Green synthesis of ZnO NPs using leaf extracts

SN	Name of plant	Size (nm)	Characterization	Structure/ shape	References
1	<i>Sedum alfredii</i>	53.7	UV-vis spectrophotometer, XRD and EDX.	Hexagonal and spherical	Qu <i>et al.</i> , 2011
2	<i>Ocimumtenuiflorum</i>	13.68	XRD, SEM and FTIR.	Hexagonal	Raut <i>et al.</i> , 2013
3	<i>Parthenium hysterophorus</i>	27–84	UV-Vis., XRD, FTIR, SEM, TEM and EDX.	Spherical and hexagonal	Rajiv <i>et al.</i> , 2013
4	<i>Olea europea</i>	18–30	UV-vis., SEM and XRD	Crystalline	Awwad <i>et al.</i> , 2014
5	<i>Sargassum muticum</i>	30–57	UV-vis., FTIR and FESEM	Hexagonal	Azizi <i>et al.</i> , 2014
6	<i>Hibiscus rosa-sinensis</i>	30–35	SEM and XRD	Crystal, spongy	Devi and Gayathri, 2014
7	<i>Eichhornia crassipes</i>	32-36	UV-vis., XRD, SEM and TEM	Spherical	Vanathiet <i>et al.</i> , 2014
8	<i>Ocimumbasilicum</i>	50	XRD, TEM and EDX analysis.	Hexagonal	Abdul <i>et al.</i> , 2014
9	<i>Catharanthus roseus</i>	23–57	XRD, SEM, EDAX, and FTRS.	Spherical	Savithramma and Bhumi, 2014
10	<i>Azadirachta indica</i>	50	FTIR and SEM	Spindle	Noorjahanet <i>et al.</i> , 2015
11	<i>Solanum nigrum</i>	20–30	UV-Vis DRS, PL, XRD, FTIR, FE-SEM, TEM, TG-DTA, and XPS	Hexagonal	Ramesh <i>et al.</i> , 2015
12	<i>Azadirachta indica</i>	25	SEM and XRD	Crystallin	Oudhiaet <i>et al.</i> , 2015
13	<i>Aloe vera</i>	22.18	UV-vis., XRD, SEM, PL, BET and TGA	Hexagonal	Varghese and George, 2015
14	<i>Senna auriculata</i>	22	UV-vis., FTIR, XRD and TEM	Spherical	Sindhuraet <i>et al.</i> , 2015
15	<i>Plectranthusamboinicus</i>	20–50	UV-Vis., FTIR, TEM and XRD	Crystalline	Vijayakumar <i>et al.</i> , 2015
16	<i>Azadiracta indica</i>	18	UV-Vis., PL, XRD, FTIR, SEM, EDAX, FESEM and AFM	Spherical	Elumalai and Velmurugan, 2015
17	<i>Phyllanthus niruri</i>	25.61	UV-Vis., UV-DRS, PL, XRD, FTIR, FE-SEM and TEM	Quasispherical	Anbuvarannanet <i>et al.</i> , 2015
18	<i>Anisochilus carnosus</i>	20–40	UV-DRS, PL, FT-IR, XRD, FE-SEM, and TEM.	Hexagonal wurtzite	Anbuvarannanet <i>et al.</i> , 2015a
19	<i>Pongamia pinnata</i>	26	XRD, UV-vis, DLS, SEM, TEM and FT-IR.	Spherical, hexagonal and nano rod	Sundrarajanet <i>et al.</i> , 2015
20	<i>Limoniaacidissima</i>	12–53	UV-Vis., FTIR, and XRD	Spherical	Patil and Taranath, 2016
21	<i>Aloe Vera</i>	8-20	UV-Vis., XRD, FTIR, SEM, EDX and TEM	Spherical, oval and hexagonal	Ali <i>et al.</i> , 2016
22	<i>Ceropegia candelabrum</i>	12–35	FT-IR, SEM, XRD	Hexagonal	Murali <i>et al.</i> , 2017
23	<i>Celosia argentea</i>	25	UV-Vis., SEM, DLS,TGA, XRD and FT-IR	Spherical	Vaishnav <i>et al.</i> , 2017
24	<i>Couroupitaguianensis</i>	57	UV-Vis., SPR and XRD	Hexagonal	Sathishkumar <i>et al.</i> , 2017
25	<i>Calotropis gigantea</i>	1.5–8.5	UV-Vis., DLS, XRD, FTIR, SEM, EDX and AFM	Spherical	Chaudhuri and Malodia, 2017
26	<i>Moringa oleifera</i>	15-20	CV, XRD, HRTEM, SEAD, DSC,TGA, FTIR and UV- vis.	--	Matiniseet <i>et al.</i> , 2017

27	<i>Camellia sinensis</i>	19	EDS, FESEM, FTIR, and UV-vis.	Spherical	Akbarian <i>et al.</i> , 2019
28	<i>Tecomacastanifolia</i>	70–75	UV-Vis., TEM, EDX, XRD and FTIR.	Spherical	Sharmila <i>et al.</i> , 2019
29	<i>Hibiscus sabdariffa</i>	20–40	FTIR, XRD, XPS, TEM, HRTEM and EDS	Spherical	Soto <i>et al.</i> , 2019
30	<i>C. halicacabum</i>	62	UV-vis., DLSA, ZP, XRD, FTIR and SEM	Hexagonal	Nithya and Kalyanasundharam, 2019
31	<i>Musa acuminata</i>	30- 80	UV-Vis., XRD, FTIR and SEM	Granular shaped	Abdullah <i>et al.</i> , 2020
32	<i>Eucalyptus globules</i>	20 - 25	UV-Vis., FT IR, XRD and SEM	Spherical and elongated	Ahmad and Kalra, 2020
33	<i>Veronica multifida</i>	11.5	UV-Vis., XRD, FTIR and TEM	Hexagonal and spherical	Dogan and Kocabas, 2020
34	<i>Ocimumgratissimum</i>	14 - 29	UV-Vis,SEM, XRD and FTIR	Spherical	Mfonet <i>et al.</i> , 2020
35	<i>Aloe vera</i>	63	UV-Vis., XRD, TEM and SEM	Spherical	Sharma <i>et al.</i> , 2020
36	<i>Aloe vera</i>	18	SEM, EDX, FTIR and XRD	Flaky and rod	Rasliet <i>et al.</i> , 2020
38	<i>Cassia auriculata</i>	20–30	UV-Vis., XRD and SEM	Rod shape	Ramesh <i>et al.</i> , 2021
39	<i>Artemisia pallens</i>	50–100	XRD, SEM and TEM	Hexagonal	Gomathi and Suhana, 2021
40	<i>Cayratiapedata</i>	52.24	UV-vis., FESEM, XRD, EDX and FT-IR	Spherical	Jayachandran <i>et al.</i> , 2021

5. EFFECT OF BIOSYNTHESIZED ZINC OXIDE NANOPARTICLES (ZnO NPs) ON SEED QUALITY PARAMETER

Food security is currently a pressing concern for the world's growing population due to limited resources and global climate change. Progressive climate change refers to changes in the climate's baseline over time, such as temperature, water scarcity, cold, salt, alkalinity, and toxic metal pollution. As a consequence, the main aim is to help plants to adapt more quickly while not damaging existing ecological systems as they must deal with climate change (Vermeulen *et al.*, 2012).

Nanoscience is a new scientific innovation platform that involves developing approaches to multiple types of low-cost nanotech applications that enhance seed germination, plant growth, development, and adaptation to the environment. Seed germination is an important stage in the plant's life cycle that promotes seedling development, survival, and population dynamics. However, environmental conditions, genetic traits, moisture availability, and soil fertility all have a significant impact on seed germination (Manjaiah *et al.*, 2019). In this regard, numerous studies have shown that the use of nanomaterials has a positive effect on germination as well as plant growth and development. As an example,

Prasad *et al.*, (2012) investigated the effects of nanoscale zinc oxide on peanut germination, growth, and yield. Peanut seeds were treated with various concentrations of nanoscale zinc oxide (ZnO) and chelated bulk zinc sulfate (ZnSO₄) suspensions (a common zinc supplement), and the treatment improved seed germination, seedling vigour, plant growth, flowering, chlorophyll content, pod yield, and root development. Treatment with ZnO NPs (25 nm mean particle size) at 1000 ppm concentration enhanced seed germination and seedling vigor, early flowering, and higher leaf chlorophyll content. As a result, a field trial using nanoscale ZnO particles at a 15-fold lower dose than chelated ZnSO₄ suggested yielded 29.5% and 26.3% higher pod yields, respectively, than chelated ZnSO₄.

According to Patra *et al.*, (2013), a foliar spray of ZnO NPs at a concentration of 10 mg/L significantly increased plant biomass, shoot and root length, and root area in cluster bean. Furthermore, they significantly increased the levels of chlorophyll (276.2%), whole soluble leaf protein (27.1%), and enzyme activities such as acid phosphatase (73.5%), alkaline phosphatase (48.7%), and phytase (72.4%) when compared to the control. The total lipids, proteins, amino acids, thiols, and chlorophyll concentrations were significantly increased after being treated with different concentration of sulfur and zinc oxide NPs as compared to the untreated control.

Ramy and Osama (2013) investigated the antifungal activity of zinc oxide nanoparticles (ZnO NPs) against two pathogenic fungus species, *Fusarium oxysporum* and *Penicillium expansum*. The antifungal action of ZnO NPs was shown to be concentration dependent, such the highest experimental concentration (12 mgL⁻¹) resulted in the greatest inhibition of Mycelial growth, with 77 and 100% growth inhibition for *F. oxysporum* and *P. expansum*, respectively.

Laware and Raskar (2014) investigated the effect of zinc oxide nanoparticles (ZnO NPs) on onions. Plants treated with ZnO NPs at 20 and 30 µg ml⁻¹ exhibited improved growth and bloomed 12-14 days faster than the control. Treated plants had considerably higher values for seeded fruit per umbel, seed weight per umbel, and 1000 seed weight than control plants. This finding suggests that ZnO NPs can shorten onion flowering time by 12-14 days and even produce viable seeds.

Raskar and Laware (2014) studied the effect of different concentrations (0.0, 10, 20, 30, and 40 g ml⁻¹) of ZnO NPs in onion seeds on cell division, seed germination, and early seedling growth. In higher concentrations of zinc oxide nanoparticles, they observed a reduction in the Mitotic Index (MI) and an increase in chromosomal abnormalities. Seed germination rates increased at lower concentrations but decreased at higher doses.

Shyla and Natarajan (2014) synthesized zinc oxide (ZnO), silver (Ag), and titanium dioxide (TiO₂) nanoparticles using a template-free aqueous solution and a simple chemical method. Groundnut seeds were dry-dressed with synthesised nanoparticles at doses of 500, 750, 1000, and 1250 mg kg⁻¹. The dose of ZnO NPs 1000 mg kg⁻¹ outperformed on the basis of germination (75%), shoot length (20.97 cm), root length (17.98), and thus vigour index (2949) compared to the control (55%, 16.92, 15.21, and 1759) respectively.

Korishettare *et al.*, (2016) investigated the effects of seed polymer coating with Zn and Fe nanoparticles (NPs) at different concentrations (10, 25, 50, 100, 250, 500, 750, and 1000 ppm) in pigeon pea. Seed polymer coating with Zn NPs at 750 ppm resulted in significantly higher seed germination (96.00%), seedling length (26.63 cm), seedling dry weight (85.00 mg), speed of germination (32.95), field emergence (89.67%), seedling vigour index (2556), dehydrogenase activity (0.975 OD value), and α-amylase activity (25.67 mm), as well as the lowest abnormal seedlings (2.50%) compared to bulk forms and control, followed by Fe and Zn NPs at 500 ppm. Despite their positive effects, these NPs inhibited germination and seedling growth at higher concentrations (nano Zn >750 ppm and nano Fe > 500 ppm). Thus, it is concluded that Zn NPs at 750 ppm can be used to improve the quality of pigeonpea seeds.

Raliya *et al.*, (2016) observed that the foliar application of zinc nanoparticles to mung beans resulted in an extension of the rhizosphere zone, with root volume increasing by 58.9%. They also found that rhizosphere enzyme activity increased, including acid phosphatase (98.07%), alkaline phosphatase (93.02%), and phytase (108%). These factors play a vital part in plant nitrogen and phosphorus absorption.

Shyla and Natarajan (2016) described the usage of inorganic nanoparticles to improve seed quality in groundnut. They used inorganic nanoparticles (NPs) such as zinc oxide (ZnO), silver (Ag), and titanium dioxide (TiO₂), which were generated chemically and analyzed using a scanning electron microscope (SEM) and a transmission electron microscope (TEM). Groundnut seeds were treated with ZnO, Ag, and TiO₂ nanoparticles at concentrations of 750, 1000, and 1250 mg kg⁻¹ of seed,

respectively, and stored for 12 months at ambient temperature. Seeds treated with ZnO NPs at 1000 mg kg⁻¹ showed improved germination (77%), vigour index (3067), electrical conductivity (0.347 dS m⁻¹), catalase enzyme activity (0.421 µg H₂O₂ mg⁻¹ min⁻¹), and lower lipid peroxidation activity (0.089 OD value) compared to the control.

According to Anandaraj and Natarajan (2017) evaluated zinc oxide (ZnO), silver (Ag), copper oxide (CuO), and titanium oxide (TiO₂) nanoparticles by using a simple chemical method. Onion seeds were dry dressed with synthesized nanoparticles at concentrations of 750, 1000, 1250, and 1500 mg/kg, with the 1,000 mg/kg dose compared to the control on the basis of germination (72%), shoot length (7.5 cm), root length (6.4 cm), and vigour index (998) compared to the control (60%, 6.0 cm, 5.4 cm, and 692), respectively.

Lakshmi *et al.*, (2017) synthesized zinc oxide nanoparticles from spinach (*Spinacia oleracea*) leaves. ZnO NPs at various concentrations (0, 25, 50, 75, 100, 125, 150, 175 and 200 ppm) were used to treat green gram (*Vigna radiata*) seeds to examine the effect on seed germination, early seedling growth, and green gram growth characteristics. The greatest findings for seed quality parameters were obtained at 125 ppm of zinc oxide nanoparticles compared to untreated seeds.

Syed and Chaurasia (2017) studied the effect of zinc oxide nanoparticles on seed germination and vigour in chilli. Different concentrations (0.0, 0.25, 0.50, and 0.75g) of ZnO NPs were prepared in distilled water and applied to chilli seeds to study the effect on seed germination, root length, shoot length, and seedling growth. The results demonstrated that ZnO nanoparticles had significant effects on germination, root, and shoot length. Seed germination increased in higher concentrations, but decreased in lower concentrations. The root, shoot, and seedling lengths were also greatest in higher concentrations, whereas in lower concentrations they decreased.

Latef *et al.*, (2017) found that seed priming with various concentrations (20 ppm, 40 ppm, and 60 ppm) of ZnONPs increased root and shoot length, fresh and dry weight of the root, and shoot of lupine (*Lupinus termis*). Seeds primed with 60 ppm concentration showed highest growth. In addition, nano-priming increased SOD, CAT, POD, and APX enzyme activity while decreasing malondialdehyde (MDA) and sodium (Na) levels as compared to salt-stressed plants without nano-priming. Priming the seeds with ZnO NPs enhanced salt tolerance in *Lupinus termis* plants.

Upadhyaya *et al.*, (2017) observed that exposure to Zn NP (0, 5, 10, 15, 20, and 50 mg/L) induced significant changes in radicle and plumule length, mass (fresh and dry mass), and seed moisture content in rice. ZnNP treatment improved the activity of antioxidant enzymes such as guaiacol peroxidase (GPX), catalase (CAT), superoxide dismutase (SOD), and glutathione reductase (GR). They reasoned that Zn NP may significantly alter antioxidant metabolism during rice seed germination and concluded that Zn NP protects rice plants from ROS damage by increasing antioxidant enzyme activity during germination.

Alaghemand *et al.*, (2018) developed zinc oxide nanoparticles from *Nigella sativa* L. seed extract and investigated plant height and number of branches. They found that spraying caused significant variations in plant height and number of branches. The best results for number of branches and height were obtained with 2 per thousand Zn nanoparticles.

Munir *et al.*, (2018) assessed the effect of ZnO NPs seed priming (0.25, 50, 75, and 100 ppm) on wheat growth and Zn uptake. Seed priming with ZnO NPs improved wheat growth, photosynthesis, and biomass linearly. ZnO NPs significantly raised the content of zinc in wheat roots, shoots, and grains. Studies showed these particles might be utilized as a supply of zinc in order to deal with Zn shortage in plants.

Rawat *et al.*, (2018) observed that seed treatment with nanoparticles at a 50 ppm concentration increases seedling length, shoot dry weight, seedling dry weight, seedling vigour index I, and seedling vigour index II when compared to seed soaking at 300 ppm concentration. They found

that seed soaking for up to 4 hours was more effective than 6 and 8 hours. Seed soaking with nanoparticles, specifically ZnO, TiO₂, and chitosan, improved wheat germination and seedling growth indices.

Lorenzo *et al.*, (2019) carried out a study in order to clarify the effects of foliar application of zinc sulfate (ZnSO₄) and zinc nano-fertilizer (ZnO NPs) on plant physiology in *Coffea arabica* L. One-year-old coffee plants were cultivated in greenhouse conditions and treated with two foliar treatments of 10 mg/L of Zn, either as zinc sulfate monohydrate (ZnSO₄·H₂O) or zinc oxide nanoparticles (20% w/t). ZnO NPs increased the fresh weight (FW) and dry weight (DW) of roots and leaves by 37% and 95%, respectively. The DW increased by 28%, 85%, and 20% in the roots, stems, and leaves, respectively. The net photosynthetic rate rose by 55% in response to ZnO NPs. ZnO NP-treated leaves contained significantly higher concentration. Overall, ZnO NPs had a greater positive impact on coffee growth and physiology than traditional Zn salts, most likely due to their higher potential to be absorbed by the leaf. These findings suggest that using ZnO NPs in coffee systems could help increase fruit set and quality, particularly in locations where Zn deficiency is high.

Choudhary and Khandelwal (2020) synthesized ZnO nanoparticles utilizing the zinc oxalate decomposition technique. Seeds were soaked in solutions containing 100, 500, 1000 ppm ZnO NPs, as well as 100 ppm ZnO supplement solution. After 5 days, it was shown that the seeds of lentil (*Lens esculentum* Linn) and chickpea (*Cicer arietinum* Linn) had the highest seed germination and vigour index at 100 ppm.

Mahmood *et al.*, (2020) biosynthesised zinc nanoparticles (Zn NPs) from licorice (*Glycyrrhiza glabra*) root extract and applied them to *Sorghum bicolor* at concentrations of 25%, 50%, and 75%. Exposure to Zn NPs had a significant impact on seed germination and other growth parameters of sorghum seedling. The low concentration of 25% Zn NPs produced the longest shoot length when compared to the high concentrations (50% and 75%). The two concentrations of 50% and 75% showed the presence of hairy roots due to the short size of roots. Thus, a low concentration (25%) of Zn NPs can be used in the field for *Sorghum bicolor* seed priming with no adverse effects to the plant.

Meher *et al.*, (2020) investigated the influence of green synthesised ZnO NPs on wheat seed germination using different nanoparticle concentrations (20, 50, and 100 ppm). Seedling length, seedling dry weight, and seed vigour index (I and II) all increased significantly at 50 ppm. At a concentration of 100 ppm for ZnO NPs, a decrease in all parameters was observed, indicating that the toxicity of nanoparticles increased with concentration.

Gunes *et al.*, (2021) also examined the effect of ZnO NPs green synthesised from *Nigella sativa* L. on the germination and seedling growth of medicinal sage. The application of ZnO NPs in various doses (0, 0.5, 1.5, and 2.5 mg ZnO NP kg⁻¹) altered the germination of *Salvia officinalis* L. seeds from 90-94%, stem length from 1.86-2.92 cm, and shoot length from 1.01-1.98 cm. The highest root and shoot lengths were obtained by applying 2.5 mg ZnO NPs kg⁻¹.

Rai-Kalal and Jajoo (2021) demonstrated the positive effects of seed priming with ZnO NPs in wheat. The primed seeds outperformed the unprimed and hydroprimed seeds in terms of germination and vigour index. It increased water absorption, which enhanced α- amylase activity. The nanoprimered plants showed a significant decrease in enzymes such as peroxidase (POD), catalase (CAD), and superoxide dismutase (SOD), which can be attributed to reduced levels of reactive oxygen species (ROS). ZnO NPs have been shown to be a promising seed priming agent for improving germination and photosynthetic efficiency in wheat seedlings.

In their 2020 study, Mahaweret *et al.*, investigated the effect of nano zinc oxide (250, 500, 750, and 1000 µg ml⁻¹) and nano copper oxide (50, 100, 200, and 400 µg ml⁻¹) on barley seed germination and growth. Nano ZnO at a concentration of 500 µg ml⁻¹ improved shoot length and germination. The maximum germination rate for micro CuO was observed at 100 µg ml⁻¹. The treatment with 200 µg ml⁻¹

¹ nano CuO increased spikelet number and shoot fresh weight, while macro CuSO₄ increased stem dry weight, shoot length, and root fresh weight.

Rawdeshdehet *et al.*, (2020) investigated the influence of moderately polydisperse ZnO nanoparticles at two concentrations (25 and 50 ppm) on lettuce seed quality. The results revealed that the treated seeds not only retained seed viability, but also showed a detectable increase in germination over the control.

Aklilu and Aderaw (2022) used khat (*Catha edulis*) leaf extract as reducing and stabilizing agent for the production of zinc oxide nanoparticles. The antibacterial activity of green-produced zinc oxide nanoparticles against Gram-positive and Gram-negative bacteria was evaluated. It has the largest inhibition zone (23 mm) against *E. coli*, but the lowest activity against *S. pneumoniae* (15 mm).

Alabdallahet *et al.*, (2022) assessed the effect of six salinity concentrations (0, 10, 25, 50, 75, and 100% of seawater) on the growth of two crop species, cowpea (*Vigna unguiculata* L.) and okra (*Abelmoschus esculentus* L.), in the presence or absence of (10 mg/L) of green synthesized zinc oxide nanoparticles (ZnO NPs) or zinc oxide (bulk ZnO) as a foliar spray after (20, 40, and 60 days) of sowing. The results showed that when seawater concentrations increased, shoot and root lengths, fresh and dry shoot weights, leaf area, and relative growth rate (RGR) decreased in both plants. The use of ZnO enhanced growth parameters when compared to the control plants, but the plants treated with (ZnONPs) performed better. Thus, (ZnO) nanoparticles are ecologically sound, low-cost, and have the potential to mitigate the effects of salt stress on plants.

Lopez *et al.*, (2022) evaluated the effects of ZnO nanoparticles on germination and plant growth, chlorophyll content, antioxidative enzyme activity, and root morphological impacts in maize (*Zea mays*) and bean (*Phaseolus vulgaris*) to see whether they may be used as an alternative to increase agricultural productivity. The seedlings were grown under hydroponic conditions for 15 days with various concentrations of ZnO nanoparticles (0, 1, 5, 10, 50, 100, and 500 mg/L). The results showed that each species responds differently to the presence of ZnO nanoparticles in the hydroponic medium, with bean seeds showing higher germination than corn seeds, with 5 mg/L being the most favourable concentration, resulting in 25% and 18% germination, respectively. Seedling development, as shown by root and stem expansion, showed differences between species, with beans responding best at 10 mg/L and corn at 100 mg/L. In this case of biomass production, concentrations of 100 and 500 mg/L resulted in a decrease in biomass results in bean seedlings, but corn seedlings produced the highest amounts of biomass at these concentrations.

Marek *et al.*, (2022) studied the effects of 1 mgL⁻¹ ZnO-NP on lentil yield, seed nutritional quality, and stress response under field settings. ZnO-NPs exposure increased yield, thousand-seed weight, and the number of pods per plant; however, there was no significant change in nutrient and anti-nutrient content between treated and untreated plant seeds. Significant differences in stomatal conductance, crop water stress index, and plant temperature were also observed. Foliar treatment of low ZnO-NP concentrations has therefore shown promising in increasing crop productivity.

Tondoyet *et al.*, (2022) evaluated the effectiveness of seed invigoration treatments with bulk zinc and ZnO nanoparticles (ZnO NPs). Seed priming with ZnO NPs at doses of 0, 20, and 40 mg L⁻¹ increased total chlorophyll content. The treatment with ZnO NPs increased yield-contributing variables such as the number of seeds per cob and the 1000-grain weight. Coating seeds with ZnO NPs (40 mg L⁻¹) significantly increased cob weight, starch, total soluble protein, and soil nutrient content (N, P, K, and Zn).

Zewde and Geremew (2022) green synthesized ZnO NPs with an aqueous solution of *Hageniaabyssinica* leaf extract, which works as a reducing and stabilizing agent for zinc acetate dihydrate. The disc diffusion method was used to test antibacterial activity against gram-positive (*S.*

aureus and *S. epidermidis*) and gram-negative (*E. coli* and *K. pneumoniae*) microorganisms. The biosynthesised ZnO nanoparticles were extremely efficient against *S. epidermidis* with inhibition zones of 21 ± 1.0 mm at 30 mg/mL, but less effective against *E. coli* with inhibition zones of 16 ± 1.0 mm at 10 mg/ml.

Abhilash *et al.*, (2023) treated onion seeds with various concentrations of ZnO nanoparticles (10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140 ppm) and stored in two types of packaging materials: aluminum foil pouch (P_1) and tin container (P_2). The seeds treated with ZnO @ 50 ppm (T_5) for 2 hours of soaking in distilled water produce better results in seed quality measurements.

Mishra *et al.*, (2023) biosynthesised zinc oxide nanoparticles (ZnO NPs) from *Catharanthus roseus* (L.) G. Don leaf extracts and used them as a nanoprimer agent to improve seed germination and seedling growth in *Eleusine coracana* (L.) (finger millet). The ZnO-nano primed seeds at 500 mg/L significantly increased all seed germination parameters, including plumule length (23.4%), radicle length (55%), vigour index (41.94%), and dry matter production (54.6%).

Ukidave and Ingale (2022) green synthesized zinc oxide nanoparticles from *Coriandrum sativum* leaves extract and evaluated their effect on plant development, specifically Bengal gram, Turkish gram, and green gram. Zinc oxide nanoparticles growth stimulatory effects were evaluated in various media by measuring seed germination rate, root and shoot length, fresh weight, dry weight, protein and chlorophyll content. Zinc oxide nanoparticles have been shown to promote seed germination and plant growth, as well as increase protein and chlorophyll content. In contrast, a lack of zinc reduces germination rate, plant development, chlorophyll, and protein content.

Singh *et al.*, (2023) examined the effects of nanoparticles on rice seed germination and physiological-biochemical phenomena under salt stress conditions. Seed treatment with ZnO-NPs at 50 mg/L enhances germination and lowers the malondialdehyde (MAD) level, which then improves the amount of photosynthetic components (such as carotenoids and chlorophyll) and the degree of germination.

Sowjanya and Prasad (2023) treated fresh pigeonpea seed with green zinc oxide (500, 750, 1000, and 1250 ppm), green silica (250, 500, 750, and 1000 ppm), chemical zinc (250 and 500 ppm), and chemical silica (250 and 500 ppm), as well as spinosad at 4.4 mg/kg seed. Green zinc oxide @ 1250 ppm had the highest seed germination, speed of germination, mean seedling length, seedling dry weight, seedling vigour index-I, seedling vigour index-II, field emergence, total dehydrogenase activity, and lowest electrical conductivity compared to the control, followed by chemical zinc oxide nanoparticles @ 500 ppm. They concluded that, seed treatment with green zinc oxide @ 1250 ppm and chemical zinc oxide @ 500 ppm nanoparticles was useful in maintaining seed quality in pigeonpea.

7. CONCLUSIONS

The ZnO NPs have become most significant and adaptable materials due to their different properties, functions, advantages, and agricultural applications. The green sources serve as a stabilizing and reducing agent in the synthesis of nanoparticles with precise size and shape. Overall, applying ZnO NPs to crops improves crop growth and yield. Zinc oxide nanoparticles at low concentrations have a positive influence on seed quality parameters such as germination, root and shoot length, dry matter content, and vigour index, but large concentrations have adverse effects.

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