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Review Article

Role of Nanotechnology in Plant Fungal Diseases: An Emerging Need to Control Fungal Diseases

Subhan Ali^{1*}, Rabia Rasheed¹, Rana Mubasher Hasan², Muhammad Usman Afzal³, Nadia Liaqat⁴, Hira Akhtar¹, Muzammil Qazi¹, Uswa Maryam¹, Sara Ijaz¹

¹ Institute of Plant Protection, MNS- University of Agriculture Multan, 61000, Pakistan.

² Agriculture Department Punjab, Pakistan.

³ ViviGro Sustainable Solutions, Canada.

⁴ Department of Plant Pathology, University of Agriculture Faisalabad 38000, Pakistan.

ABSTRACT

Fungal diseases significantly impact crop production, causing considerable economic losses and environmental harm. Nanotechnology, particularly nanoparticles (NPs) has emerged as a pivotal tool for advanced pathogen diagnostics and management, exhibiting novel antifungal properties such as cell damage, lipid peroxidation and DNA destruction. This review highlights specific NPs (e.g., silver, copper oxide) with outstanding antifungal activities and their mechanisms of action. It quantifies successes in agricultural applications demonstrating NPs' potential in enhancing productivity and sustainability through nanocomposites, nanohybrid antifungals and nanobiosensors. Additionally, we address the environmental safety and scalability challenges, emphasizing the need for comprehensive field studies and the development of safe, effective nanoparticle delivery systems. The promising results underline NPs as a sustainable solution to combat phytopathogenic fungi, setting the stage for future innovations in agricultural disease management.

Keywords: Plant pathogen diagnostics, Antifungal activities, Cell wall damage, Lipid peroxidation, Protein denaturation.

INTRODUCTION

Agriculture, a critical pillar of the global economy has seen remarkable advancements in yield and productivity partly due to the increased use of pesticides and insecticides (Kalia and Gosal, 2011; Meena et al., 2020). Despite these gains, plant diseases and pests account for approximately 20% to 40% of annual crop losses, with fungal pathogens, including Ascomycetes and Basidiomycetes, significantly inhibiting plant development and production, thereby imposing considerable economic burdens on the agricultural sector (Shuping and Eloff, 2017; Kutawa et al 2021). The resilience of fungi capable of thriving in any environment with low moisture. Fruits, vegetables, and crops like ginger, soybeans, and other rhizomes (Pawar et al., 2008; Ghuffar et al., 2021) are susceptible to fungal spores from air, water and soil. The phytopathogenic fungus like *Aspergillus niger*, *A. carbonarius*, *A. flavus*, *A. parasiticus*, and *A. ochraceus* can cause diseases in many crop in fields and storage spaces (Perrone et al., 2007; Ingle et al., 2023). Ochratoxins are produced by *A. niger* and *A. ochraceus* poses health risk to human health and farm animals when ingested alongside the contaminated meals. Other human infections from the *Aspergillus* genus include *A. flavus* and *A. fumigatus*, causes severe fungal asthma and invasive aspergillosis in immunocompromised people (Boyer et al., 2023; Stewart et al.,



Correspondence

Subhan Ali
sa99344@gmail.com

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2023). Human infections have been treated with a variety of antifungal drugs (Tóth et al., 2023; Ingle et al., 2023). Maize, grapes, garlic, onion, peanut and many crop are particularly vulnerable to *Aspergillus* infestations (Pimienta et al., 2022; Gilbert et al., 2023). The farm level management of fungi is crucial for controlling plant diseases. Advanced technologies, such as smart nanoparticles (NPs) with biological components are needed for long-term efficiency (Alghuthaymi et al., 2021).

Nanotechnology has revolutionized agriculture offering new innovative solutions for plant disease management. It has made considerable progress in pharmacology and medicine but agricultural use of nanotechnologies is relatively less developed (Balaure et al., 2017; Sinha et al., 2017). Material scientists engineer NPs with desired properties for protection and active delivery through adsorption, encapsulation, and conjugation, ensuring effective quantities are precisely applied (Khandelwal et al., 2016). NPs can accurately detect and quantify food safety, quality, nutritional characteristics (Zhang et al., 2018; Wang et al., 2020a); gases and vapours (Tang et al., 2019; Xiao et al., 2020); small organic molecules (colourants, vitamins, pesticides, fungal toxins etc) (Yang et al., 2019); biomolecules such as proteins (Wang et al., 2020b) and polynucleotides (Xiong et al., 2020); microorganisms (Rippa et al., 2020); and environmental stimuli (temperature, light, moisture, pH) by targeting inorganic chemicals and heavy metal ions (Ehgartner et al., 2016; Gupta et al., 2019; Yang and Duncan, 2021). The potential emergence of new fungicides, insecticides and other actives for plant diseases management could significantly contribute to the advancement of agricultural nanotechnology (Worrall et al., 2018).

NPs can be used to protect plants in two different ways: 1) Direct protection of crops by spray; 2) alter seed or plant functions by coating on seeds and dipping foliar tissue or roots (Hayles et al., 2017). Nanotechnology has shown promising results in the development of self-assembling systems and organic entities, impacting medicine, pharmacology and agriculture (Yadav et al., 2020). They are also essential for medication delivery, diagnostics, imaging, antimicrobial agents and sensors (Patra et al., 2018). Nanotechnology is also being used in the agricultural industry for plant hormone delivery, nano-barcoding, and nano-sensor systems for disease, pest, and nutritional deficiency diagnosis. It can also improve pesticides by decreasing toxicity, extending shelf life, and increasing solubility of poorly soluble pesticides (Shang et al., 2019). These benefits may also have a positive impact on the environment (Worrall et al., 2018). Nanotechnology can also help control fungal diseases by reducing adverse effects of fungicides, increasing solubility of low water-soluble fungicides, and minimizing toxic impact (Kutawa et al., 2021). Nanotechnology can reveal biological features of biological systems, microbes, and antimicrobial chemicals by altering molecular sizes (Singh et al., 2011). Silver particles on graphene oxide were found to reduce tomato bacterial spots in vitro; however, nanoparticles comprised of zinc (Zn), silver (Ag) and copper (Cu) have previously been reported to control plant diseases (Ocoy et al., 2013). CuNPs inhibited the *Fusarium verticillioides* growth due to their dependence on phloem (Giannousi et al., 2013). ZnNPs inhibit fungi of *Aspergillus. sp*, *Fusarium. sp* and *Rhizopus. sp*, as well as bactericide such as *Pseudomonas* (Servin et al., 2015). However, rhizobacteria in biotechnology has limitations such as allergies, displacement, and pathogenic effects on non-target organisms.

Carbon, silver metal oxides or alumino-silicates and silica (Si) are the utmost used nanoparticles materials in plant disease management. These NPs effectively control a range of plant diseases such as *Aspergillus spp.*, *Botrytis cinerea* and *Fusarium spp.*, and the promote growth and development in plants. SiNPs induces resistances in maize against phytopathogens such as *F. oxysporum* and *A. niger* (Worrall et al., 2018). However, nano-hybrids or composites can increase their action spectrum by using NPs in the formulation of fungicides (Paek et al., 2011; Spadola et al., 2020). Furthermore, the use of synthesized, alumino-silicate nanoplates have been reported with twin benefits of enhanced biological activity and improved environmental security (Iavicoli et al., 2017).

TYPES OF NPs FOR PLANT FUNGAL DISEASE MANAGEMENT

Nanoparticles (NPs) have been incorporated into disease control plans as bactericides and fungicides and as nano-fertilizers to improve plant health. Some types of NPs used for fungal plant disease management (PDM) are graphically represented (Figure 1).

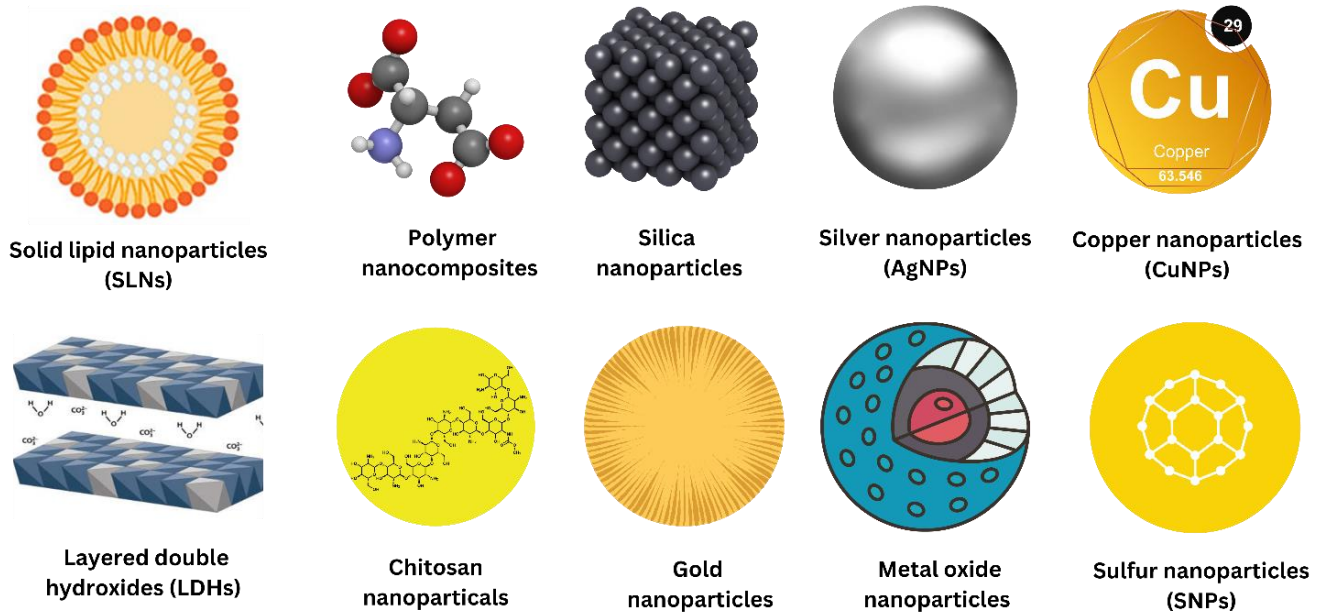


Figure 1. Graphical representation of different types of nanoparticles used for plant fungal disease management.

Silver nanoparticles (AgNPs) and their role in PDM underscores the significant potential of nanotechnology in agriculture. AgNPs have been recognized for their multifunctional nature, antimicrobial activity against a broad range of microorganisms, which makes them a very potential and safe substitute for chemical fungicides. The potential to eradicate the mycelial growth and spore germination of numerous phytopathogenic fungi such as *B. cinerea*, *Bipolaris* sp., and *Fusarium* sp. stresses the development of new effective treatment methods in plant pathology and disease control (Li et al., 2022). The manner in which AgNPs cause adherence to the fungal cell wall and inhibit cell metabolism to have its antifungal effects shows that this action is selective and thereby, may not induce extra undesirable effects compared to broad spectrum antifungal agents, as well as contribute to the development of resistance among pathogenic fungi (Wen et al., 2023). In addition, the mutual enhancement of antifungal effects when AgNPs are acted together with *A. alternata* and fluconazole against the strains like *Phoma herbarum*, *P. glomerata*, *Fusarium semitectum*, and *Trichoderma* sp. paves way for genetic based management practices and tools for disease control (Gaibhiye et al., 2009). Interestingly, the observed variation in the inhibition rates of these fungi indicate that AgNPs could be modified or used in combination with other compounds that charge the wanted pathogen more efficiently. Earlier studies have shown that AgNPs can control *Sclerotinia minor*, *S. sclerotiorum* and *R. solani* effectively (Min et al., 2009).

Polymer nanocomposites are a significant enhancement in agricultural usage, particularly in combating plant diseases that are resulted from fungal infections. These are multiphase composites of polymers and nanoparticles that work within the scale of one to one hundred nanometers. They make it quite possible to have direct intervention on the fungal pathogen which in turn makes it possible to have direct approach in handling the diseases (Ntow-Boahene, et al, 2021). These are materials that can be used in antifungal agents or in nanofertilization offering several ways to improve the quality and the yield of crops. They also serve as weed control, nanopesticides and biostats that enhances crop defense and yield (Adisa et al., 2019). The utilization of these nanocomposites in farming ensures that sustainable farming practices are adopted, disease control is effective and crops perform well. These nanoparticles have properties that make their catabolism more environmental friendly as opposes to other forms of particles.

Sulfur nanoparticles (SNPs) effectiveness for antifungal activity has been studied, acknowledging their ability for surface transformation and nanoscale synthesis of elemental sulfur. Most of fungus and bacteria resist the sulfur compounds which are available in the market. The powdery mildew of okra caused by *Erysiphe cichoracearum*, was evaluated against compounds containing SNPs and a considerable negative impact on the germination of *E. cichoracearum* conidia were observed (Gogoi et al., 2013). The effectiveness of SNPs against *F. solani* and *Venturia*

inaequalis were exhibited (Rao and Paria, 2013). It was shown that smaller SNPs (less than 35 nm) have more antifungal activity.

Copper nanoparticles (CuNPs) are identified as effective antifungal agents against many phytopathogenic fungi. CuNPs produced synthetically from the endophytic actinomycete *Streptomyces capillispiralis* have been shown to exhibit encouraging antifungal action against phytopathogenic fungi. CuNPs were found to be effective against *Alternaria* spp., *A. niger*, *Fusarium* spp., and *Pythium* spp. (Hassan et al., 2019). Citrus juice (*Citrus medica* Linn.) was effectively used in bio-synthesized CuNPs by Kirby-Bauer disk diffusion method against phytopathogenic fungi such as *F. culmorum*, *F. graminearum* (Shende et al., 2015). In another study, *in-vitro* efficacy of CuNPs was evaluated by Banik and Pérez-de-Luque (2017). *Phytophthora cinnamomi* was 76% inhibited by CuNPs in conjunction with copper oxychloride at 50 mg/ml. Reduction of *A. alternata* mycelium and sporulation was also noticed using CuNPs and copper oxychloride composite.

Solid lipid nanoparticles (SLNs) are made up of lipids that are solid at normal temperature and resemble emulsions. SLNs have the benefit of offering a matrix that can ensnare lipophilic active molecules without requiring the use of organic solvents (Ekambaram, 2012). Additionally, SLNs can potentially offer regulated release of different lipophilic components because the active in the solid matrix is less mobile (Ekambaram, 2012; Borel and Sabliov, 2014). In an experiment SLNs were evaluated against *A. fumigatus*, the inhibition zone increased by 8 mm as in contrast to 18 to 26 mm when natamycin was used alone. When natamycin was added in SLNs, the MIC ascertained by the broth dilution experiment decreased by 2.5 times (Khames et al., 2019).

Silica based nanoparticles (SiO₂NPs) are significant due to a relatively simple synthesis and a well-tuned nanoscopic size, shape, and composition (Kashyap et al., 2015). The impact of SiO₂-NPs on *A. dauci* was showed potential antifungal activity and minimized the fungus's effect on carrots (Ahamad and Siddiqui, 2021). The study assessed the efficacy of biosynthesized SiO₂NPs as an alternative to pesticides against early eggplant blight disease, using *A. niger* for environmentally safe silica nanoparticles with large surface areas and compact dimensions (Albalawi et al., 2022).

Metal oxide nanoparticles have been widely applied as novel fungicides for plant disease management. The antifungal characteristics of zinc oxide (ZnO), copper oxides (CuO), iron oxide (Fe₃O₄) and other metal oxide nanoparticles have been shown to be useful against phytopathogenic fungi (Cruz-Luna et al., 2023). Size-dependent antifungal efficacy against a variety of plant pathogenic fungi has been confirmed for ZnONPs that are biologically produced utilizing *Parthenium hysterophorus* L. The first report on the green manufacture of Fe₃O₄ NPs from *Platanus orientalis* leaf extract was reported (Devi et al., (2019). These Fe₃O₄ NPs with Tannic acid exhibited significant inhibitory effect towards *M. piriformis* and *A. niger*. (Parveen et al., 2018).

Chitosan is a nontoxic, bio-degradable and bio-compatible polymer that has been broadly accepted as one of the important organic compounds that can be used as the natural carrier for the active agent to control plant diseases (Rubina et al., 2017). The polymers had such an antimicrobial trait that it had an effect on the fungal virulence factors like the mycelia growth, sporulation viability and germination (Xu et al, 2007). Being reactive amine and hydroxyl group, chitosan can be modified and its properties gain improvement by ionic interactions, grafting and modifying (Li et al., 2011). Higher efficiency of the chitosan based Ag NPs related with 4(E)-2-(3-hydroxynaphthalene-2-yl) diazenyl-1-benzoic acid was reported by the researchers with 20.2 mm and 27.0 mm inhibitory diameters against *A. flavus* and *A. niger*, respectively (Mathew et al., 2013). Likewise, with concentration 800 mg/mL of chitosan hydrogel incorporating cinnamic acid encapsulated *Mentha piperita* essential oil, the growth of *A. flavus* mycelia is highly retarded (Beyki et al., 2014).

Layered double hydroxides (LDHs) are clays that take the shape of hexagonal sheets and contain layers of active molecules trapped in between layers (Xu et al., 2006; Worrall et al., 2018). LDH nanoparticles break down whenever moisture and atmospheric carbon dioxide are added, or when the environment becomes acidic (Mitter et al., 2017). Positive charged delaminated LDH lactate NPs have been exhibit to increase the permeability of physiologically active materials across the wall of plant cell (Bao et al., 2016). LDH nanosheets improve fungicide adherence to leaves and antifungal efficacy (Zhi et al., 2019). The charge distribution in LDHs promotes effective pesticide dispersion, improves adherence and the antifungal activity.

NANOTECHNOLOGY IN FUNGAL DISEASE MANAGEMENT

Nanoparticles (NPs) exhibit increased antifungal activity and interact with fungal diseases in plants. The application of NPs results in the disruption of cell metabolism, cell wall damage, substrate damage, as well as the accumulation

of the cytoplasm. NPs are biosynthesized from fungi via intracellular and extracellular pathways so that the pathogen growth is inhibited and oxidative reaction is stimulated. The NPs in plants are also involved in hormonal activities that plays an important role defense against diseases.

Protectants

Nanoparticles (NPs) as protectants pose a significant in controlling fungal plant diseases by protecting phytopathogens from plant foliage, roots and seeds (Kutawa et al., 2021). Metal oxide NPs such as ZnO, CuO and ferrous compounds have potential nano-fungicidal agents against PDM (Cruz-Luna et al., 2023). Furthermore, the relation of fungal polymer based nanoparticles has explored for protecting tomato plants against wilt disease caused by *Fusarium oxysporum* f. sp. *Lycopersici* (Ram et al., 2023). NPs such as silver, zinc, and copper can alter antimicrobial activity by altering host defenses. They can be combined with other antifungal agents, such as fungicides, for better effectiveness. For example, biosynthesized AgNPs have been shown synergistic antifungal activity when combined with fungicides (Worrall et al., 2018).

Nanoparticles are sized from 10 to 100 nm and synthesized with a desirable unique physical, biological, or chemical properties that are differentiable from the molecule and bulk counterparts (Yang et al., 2008; Worrall et al., 2018). NPs can be sprayed directly to plant seeds, leaves, or roots to protect them from pests and diseases. Metal Nps such as ZnO, CuO, SiO and titanium dioxide NPs have thoroughly been studied for antibacterial and antifungal properties (Kah and Hofmann, 2014; Kim et al., 2018). Recent green-synthesis of SNPs through plants, fungus, or yeast, gained attention (Rafique et al., 2017). Nanotechnology exposed the imaginable to generate a tough footmark for emerging actual preparations (Figure 2).

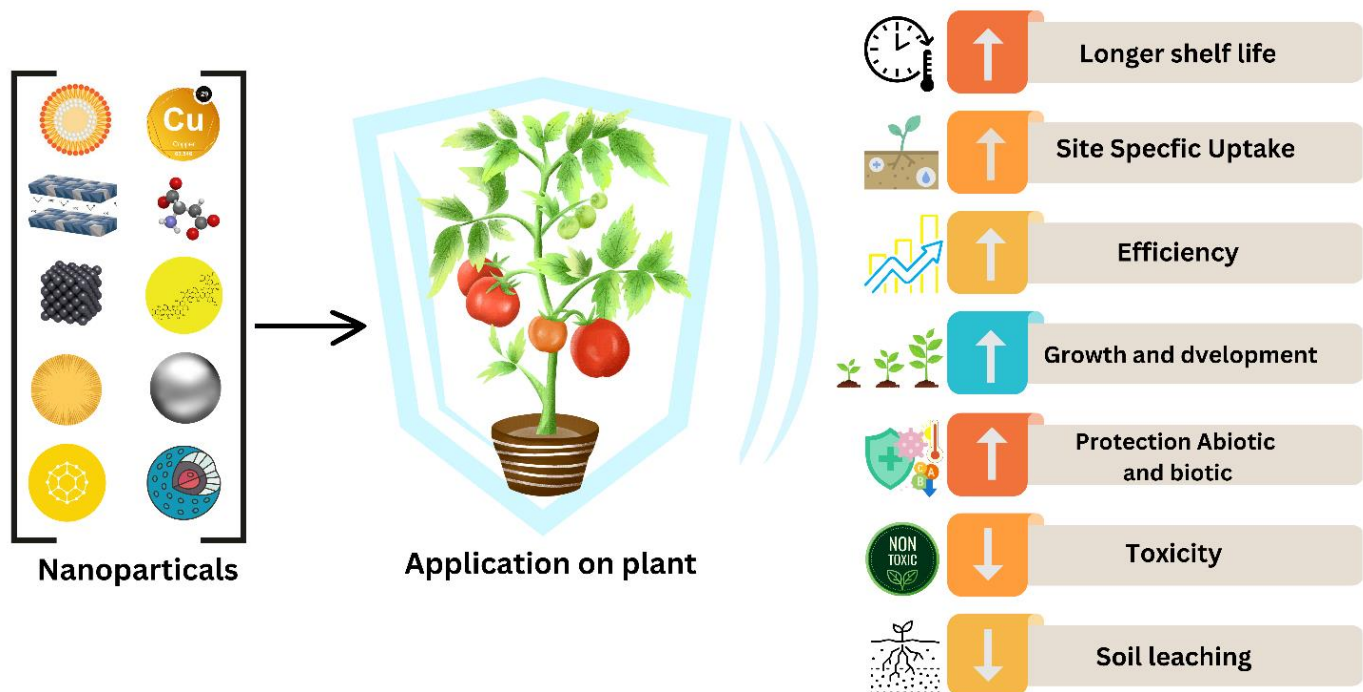


Figure 2. Different nanoparticles Act as protectants

SNPs have been shown antifungal inhibition of *A. alternata*, *B. cinerea*, *Curvularia lunata*, *Macrophomina phaseolina*, *R. solani* and *S. sclerotiorum*, by well diffusion assay (Krishnaraj et al., 2012). It has been noted that some chitosan NPs had antifungal activity against Fusarium crown, root rot on tomato, Botrytis bunch rot on grape and *Phyricularia grisea* in rice (Kashyap et al., 2015).

Fungicide Carriers

NPs are carriers, as they allow for the directed delivery of active substances and cause minimal chemical contamination (Worrall et al., 2018). The use of nanohybrid materials in managing fungal diseases is gaining importance due to their unique physicochemical properties. These nanoparticles can be used as carriers of fungicides, making them an environmentally-friendly approach to disease management (Worrall et al., 2018). These nano-materials have been exhibit effective potential in managing plant diseases with eco-friendly compared to

traditional agrochemicals, making them best alternative (Servin et al., 2015; Matras et al., 2022; Cruz-Luna et al., 2023). Silver and gold nanoparticles has revealed shape and size dependent antimicrobial activities and potential materials as compare to fungicides in agricultural applications (Matras et al., 2022). Surveys reports in many regions of Mexico has revealed the metal oxide nanoparticles as best alternative for fungicides in controlling the plant diseases (Cruz-Luna et al., 2023).

Nanoparticles and Rnai for Plant Protection

Plant protection strategies have been formulated using nanoparticles and RNA interference (RNAi), especially the control of fungal disease. Recent studies show nanoparticles delivered RNAi particles can sustain disease protection for longer periods, providing potential for plant defense against phytopathogenic fungi (Ray et al., 2022). In addition, polymer-based nanoparticles have been used against wilt disease caused by *F. oxysporum* f. sp. *Lycopersici* in tomato plants (Ram et al., 2023). Application of NPs disrupts the activity of important membrane proteins, interferes with cell permeability, and inhibits the transcription and replication processes (Cruz-Luna et al., 2021; Arshad et al., 2023). By raising the amounts of gene transcription in response to oxidative stress (ROS), they impact the potential of the mitochondrial membrane. Moreover, the NPs genotoxic activity damages DNA, which results in cell death. ROS are produced when certain metallic NPs increase oxidation processes. This results in significant damage to proteins, membranes, and DNA, as well as disruptions to the absorption of nutrients (Huerta-García et al., 2014; Kumari et al., 2017; Zhao et al., 2017; Mikhailov, 2020; Rana et al., 2020; Cruz-Luna et al., 2021; Arshad et al., 2023). Effects of applying various nanoparticle types containing miRNA on organelles and cellular components in phytopathogenic fungus cells are currently being explored in Figure 3.

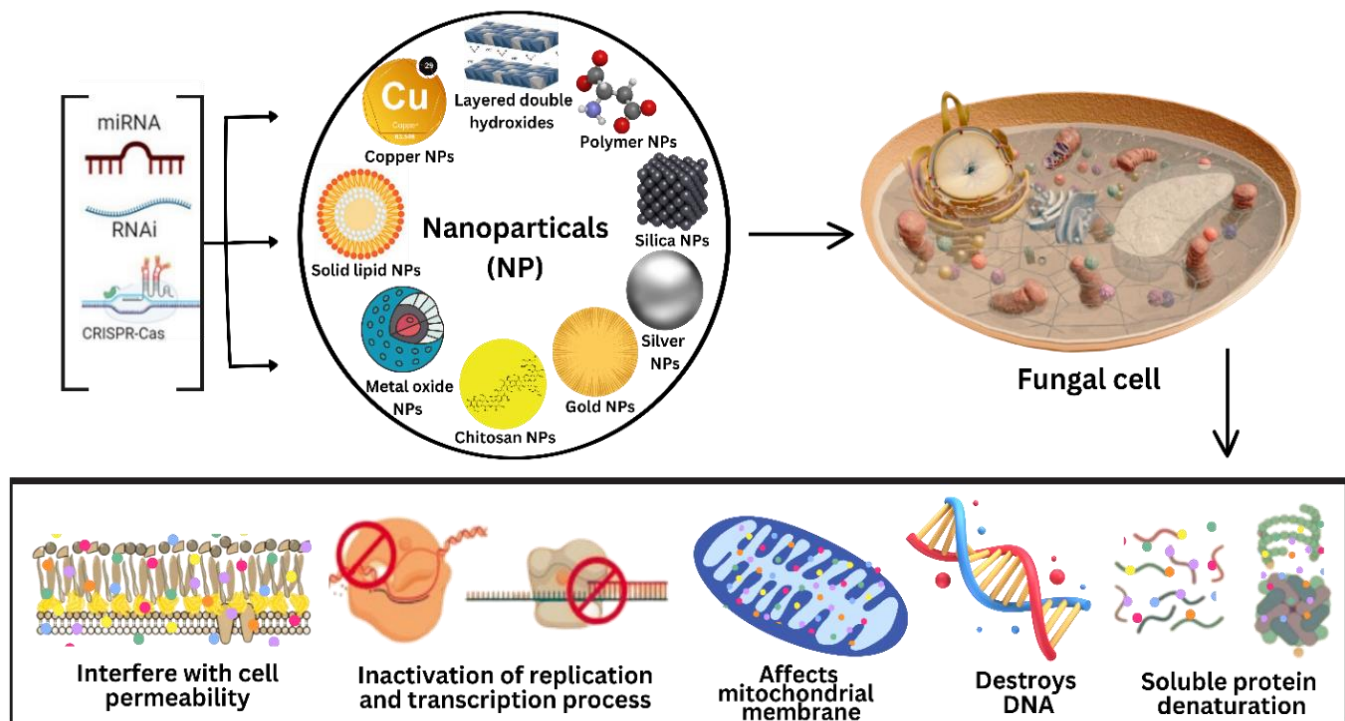


Figure 3. Influence of applying multiple types of NPs with miRNA on cellular components and organelles in phytopathogenic fungal cell

In plants, the initiation of RNAi with dsRNA being treated into small-interfering RNA (siRNA) is through Dicer-like (DCL) enzymes. Incorporation of these siRNAs into RNA-induced silencing complex (RISC) leads to base-pairing-induced degradation in pathogen RNA triggered by RISCs because the pathogen RNA cannot be used as a template for translation (Baulcombe, 2004). Currently, genetically synthesized RNAi is a dominant strategy against plant diseases and pests. However, research into novel dsRNA delivery strategies is ongoing due to contentious regulations and topical use of RNAi with NPs is also explored against many fungal diseases (Zhang et al., 2010, 2015). External use of RNAi still has challenges, even though exogenous administration of RNAi-inducing chemicals for crop protection is more appealing than pesticides because of its lower toxicity (Mitter et al., 2017b).

ADVANCEMENTS IN NANOTECHNOLOGY FOR IMPROVED PATHOGEN DIAGNOSIS

Advancements in nanotechnology have led to the development of novel approaches for improved pathogen diagnosis, particularly in the management of fungal diseases. Nanohybrid antifungals are a possible approach to controlling plant diseases due their advanced performance against phytopathogens as compared with traditional forms of methods used for management the disease (Alghuthaymi *et al.*, 2021). The techniques employed in nanohybrid synthesis may be expensive due to the use of costly reagents and physical energy but are highly effective, ecofriendly and long term resistance against phytopathogen microorganism. Fungal biomass produces mycogenic NPs that protect plants from fungal pathogens (Yadav *et al.*, 2023). These NPs can be combined with other nano-strategies to improve detection and management of pathogens. Silver and gold NPs have been investigated for fungal disease management and can also be used to diagnose disorders (Ray *et al.*, 2023). Nanobiosensors are developed to diagnose and target specific pathogens due to having diagnostic agents like enzymes and antibodies to identify and target plant sites, making pathogen detection more efficient (Mansoor *et al.*, 2021). Some fungi synthesize bioactive compounds that can act as prototypes of antifungal nanoparticles which in turn can be used to control fungal plant pathogens and diagnosis (Yadav *et al.*, 2023).

The nanotechnology in diagnosis of pathogens is experiencing popularity because of the optical property and input control capacity (Wang *et al.*, 2016). Protein conjugation to NPs, DNA, and other targeting biomolecules enables rapid, selective, and specific detection of infections (Kashyap *et al.*, 2017). Gold nanoparticles possessing optical or electrochemical functions have been widely adopted in diagnostics because of their method recovery of pathogen diagnosis. Schwenkbier *et al.*, (2015) designed a chip hybridization method with SNPs for *Phytophthora* spp. The diagnosis of *Aspergillus niger* in agricultural crops is now achievable with the combined coated application of CNPs and CuO. Another approach included the use of transportable technology to identify bacterial and fungal species within cereal grains in stored conditions (Alghuthaymi *et al.*, 2021). These innovations in nanotechnology can significantly leads to the development of better fungal diseases management for crops.

NANOCOMPOSITES DEVELOPMENT FOR INTEGRATED MANAGEMENT OF FUNGAL DISEASES

Nanocomposites have been found to effectively manage fungal diseases in crops due to antifungal properties through engineering. Chitosan-loaded CuONPS have been examined for their antifungal properties against Fusarium wilt disease in tomato plants (Mosa and El-Abeid, 2023). Therefore metal oxide nanoparticles can replace fungicides in PDM (Cruz-Luna *et al.*, 2023). SNPs effectively control phytopathogens such as *R. solani*, *S. sclerotiorum* and *Spilocaea minor* caused significant injury by rupturing hyphal wall membrane and destroy interior hyphae (Min *et al.*, 2009). The AgTNTs nanocomposites functionalized using SNPs was tested against *B. cinerea* using a photo-inactivation method, causing conidia destruction and cell death (Rodríguez-González *et al.*, 2016). The synthesized CNPs were effectively exhibited antifungal activity against *Fusarium poae* and *F. graminearum* (Wang *et al.*, 2014).

Nanocomposite materials have multi-phase components and co-synthesized by different inorganic and organic elements together to perform the specified action has triggered them (Adnan *et al.*, 2018; Ashfaq *et al.*, 2020). *Nyctanthes arbor-tristis* mediated ZnONPs has testified to contain antifungal properties against some phytopathogen; *Alternaria* sp, *Aspergillus* sp, *Botrytis* sp, *Fusarium* sp, and *Penicillium* sp (Ingle *et al.*, 2020). The effect of metal ion chelators, particularly Fe-chelator on the antifungal activity of metal-oxide NPs was described (Zabrieski *et al.*, 2015). Comparing the results obtained for susceptibility of both pathogens to CuONPs, it is seen that *P. ultimum* was discovered to be further susceptible to CuONPs than *Pythium aphanidermatum*. As expected, when *Pythium* sp. was grown on media which did not contain ZnONPs, its growth was again limited by their presence in the original substrate. A study focuses on the use of nanotechnology in plant fungal diseases control, for instance, in a recent study, it was shown that Graphene oxide and Iron oxide nanocomposite can inhibit spore germination of *Plasmopara viticola*, thus reducing disease effects on grape plants (Wang *et al.*, 2017; Akhtar *et al.*, 2024). Rasheed *et al.*, (2023) proved that 1 percent potassium silicate and potassium monophosphate cut back powdery mildew of pumpkin vines by 49 percent while Bravo and Score fungicides lessen the disease impact by 61 percent making up prime mixture management strategies. Polymers may require slower or faster release and biodegradation rates, earning them further application as vehicles for agrochemicals and medicines (Sampathkumar *et al.*, 2020). Drugs and agrochemicals are frequently delivered using polymers such polylactic acid, poly-butadiene, poly-lactic-glycolic acid, polyacrylamide, polyvinyl alcohol (PVA), cellulose acetate phthalate, gelatin, chitosan, gum Arabic, polyhydroxyalkanoates, and polystyrene (Spadola *et al.*, 2020).

Table 1. Different nanoparticles with their potential applications against Fungal Diseases

NPs	Vegetable	Disease Causing Agent	Disease	Mechanism	Sources
AgNPs	Potato	<i>P. infestans</i>	Late blight of potato	Antifungal activity	(Worrall et al., 2018)
	Tomato	<i>F. oxysporum f. sp. Lycopersici</i>	Fusarium wilt	AgNPs (25 to 200 ppm) reduced fungal growth	(Ansari, 2023)
	Cucumber	<i>Colletotrichum orbiculare</i>	Anthracnose	Antifungal activity and reduced disease severity	(Worrall et al., 2018)
Chitosan NPs	Pepper	<i>Phytophthora capsici</i>	Phytophthora blight in peppers	Antifungal activity, inhibition of mycelial growth and sporangia production	(Pabón-Mora et al., 2023)
CuONPs	Cucumber	<i>F. oxysporum f. sp. Cucumerinum</i>	Fusarium wilt	Reduced fungal growth and disease incidence	(Worrall et al., 2018)
	Tomato	<i>F. oxysporum f. sp. Lycopersici</i>	Fusarium wilt		(Worrall et al., 2018)
Fe ₃ O ₄ NPs	Tomato	<i>F. oxysporum f. sp. Lycopersici</i>	Fusarium wilt		(Ansari, 2023)
SiO ₂ NPs	Tomato	<i>F. oxysporum f. sp. Lycopersici</i>	Fusarium wilt	Reduced fungal growth and disease incidence	(Rajani et al., 2022)
		<i>R. solani</i>	Black rot of tomato	Inhibited the growth of <i>R. solani</i>	(Alghuthaymi et al., 2021)
TiO ₂ NPs	Cucumber	<i>Colletotrichum orbiculare</i>	Anthracnose	Antifungal activity and reduced disease severity	(Kutawa et al., 2021)
	Tomato	<i>Alternaria solani</i>	Early blight		(Dutta et al., 2022)
ZnONPs	Tomato	<i>Alternaria solani</i>	Early blight		(Pabón-Mora et al., 2023)
	Cucumber	<i>F. oxysporum f. sp. Cucumerinum</i>	Fusarium wilt	Reduced fungal growth and disease incidence	(Worrall et al., 2018)

NANOTECHNOLOGY IN FOOD AND PLANT SCIENCE

Nanotechnology plays a pivotal role in food and plant sciences, offering innovative solutions for the control of fungal diseases, development of nanopesticides and improvement of food safety. Utilization of nanomaterials (NMs) including nanometals, nanorods and nano-films contribute to plant growth enhancement, genome modification and increased stress tolerance. Nanotechnology is also being explored for food pathogen detection and sensors. It is susceptible for solving the issues like climate change and sustainable agriculture (Sanzari et al., 2019; Ansari, 2023). Nanotechnology being a critical component of food security it helps to increase agricultural productivity by stopping microbial diseases and insects, and provides nutritional value and safety. It aids in cooking, packaging, and long-term food storage, improving taste and texture. NMs and nanosensors provide consumers with information on food condition and nutritional value, and nanotechnology-based delivery technologies enable targeted distribution of food bio-actives and enhanced bioavailability (Nile et al., 2020). Humans and food have both been shown to bioaccumulate some NMs, such as AgNPs produced by nano-packaging (Jovanović, 2015).

Increased amount of NPs can be very harmful because they denature the body's protein and enzymes through processes that increase free radical generation which is thought to be among major factors causing oxidative stress in humans (Li et al., 2011; Fu, 2014). Once being released into the agro-environment, NPs go through complex transformations that can pose inherent risk to the health of human and plants (Shi et al., 2013; Berekaa, 2015).

AgNPs exposition brings phytotoxicity in aquatic plants such as *Egeria densa* and *Juncus effusus* by promoting lipid peroxidation, physiological trait changes and causing extra stress among submerged macrophytes (Cubadda *et al.*, 2013). To protect the environment, plants, people, animals, and aquatic life, green nanofillers are required for nanocomposite research and the safer use in the food industry necessitates the establishment of legislative legislation, rules, and regulations.

Regulations from the European Union declare that before being authorized for use of any food ingredient based on nanotechnology must undergo a safety evaluation conducted by the Food and Drug Administration (FDA) (Cubadda *et al.*, 2013). Toxicological screening is essential for the use of nanotechnology, nevertheless, as many countries do not have appropriate laws governing, the synthesis and processing of NMs (Nile *et al.*, 2020). Plant genetic engineering working to produced well-established CRISPR-DNA that NPs can transport CRISPR chemicals to plant mitochondria and plastids for genome alteration (Demirer *et al.*, 2021; Ansari, 2023).

CHALLENGES AND POTENTIAL OF NANO-BIOTECHNOLOGY IN FUNGAL DISEASES MANAGEMENT

Nanobiotechnology has proven to have a high level of effectiveness when used on managing fungal disease in crops. It was noted that biological NPs produces from bacteria and fungi have shown potential antifungal activities against diverse pathogenic fungus strain demonstrating the application of cost effective eco-friendly disease management strategies (Murali *et al.*, 2021). Table 2 summarizes the details about biological NPs used against some phytopathogenic fungi. Further, nanohybrid antifungals have been synthesized from the combination of NMs with biopolymers and are known to control fungal pathogens in plants under changing climatic situations as well as widespread diseases (Alghuthaymi *et al.*, 2021). Nanotechnology, including inorganic nanoparticles and mycogenic nanoparticles, has the potential to reduce plant diseases and protect against pests and pathogens in sustainable agriculture (Yadav *et al.*, 2023). These developments are reflecting the potential of nano-biotechnology as an advanced technology for proper control fungal diseases in crops that bring novel and eco-friendly ways to agricultural industries.

Table 2. Biosynthesized nanoparticles used against phytopathogenic fungi.

NPs	Synthesis Used	Against Phytopathogenic fungi	Against Disease	Sources
AgNPs	<i>A. alternate</i> + fluconazole	<i>Phoma glomerata</i> ,	-	(Gajbhiye <i>et al.</i> , 2009)
		<i>P. herbarum</i> ,	-	
		<i>F. semitectum</i>	Fungal blast in wheat	
		<i>Trichoderma</i> sp.	-	
	<i>Brassica rapa</i> L.	<i>Gloeophyllum abietinum</i> ,	brown-rot	(Narayanan and Park, 2014)
		<i>G. trabeum</i> ,	brown-rot	
		<i>Chaetomium globosum</i>	-	
		<i>Phanerochaete sordida</i>	white rot decay	
	<i>Bacillus</i> sp. strain GP-23	<i>F. oxysporum</i>	-	(Gopinath and Velusamy 2013)
	<i>Solanum nigrum</i>	<i>R. solani</i>	Root rot, collar rot and wire stem	(Akther and Hemalatha, 2019)
<i>Fusarium udum</i>		Wilt disease in pigeon pea plants		
<i>F. graminearum</i>		Fusarium head		

			blight or scab of wheat and barley	
CuNPs	<i>Citrus medica</i> Linn.	<i>F. culmorum</i> ,	Ear blight or scab	(Shende et al., 2015)
		<i>F. oxysporum</i>	-	
		<i>F. graminearum</i>	Fusarium head blight or scab of wheat and barley	
	<i>Streptomyces capillispiralis</i>	<i>Alternaria</i> spp.,	-	(Hassan et al., 2018)
		<i>Aspergillus niger</i> ,	Black mold of onions and garlic	
		<i>Fusarium</i> spp.	-	
		<i>Pythium</i> spp.	crown and root rot disease in several plants	
<i>S. griseus</i>	<i>Poria hypolateritia</i> Berk. ex Cooke	Red root rot disease of tea	(Ponmurugan et al., 2016)	
CuONPs	<i>Platanus orientalis</i>	<i>A. niger</i>	-	(Devi et al., 2019)
		<i>Mucor piriformis</i>	-	
SNPs	CTAB- and SDBS-mediated	<i>F. solani</i>	Early blight and Fusarium wilt disease of Tomato	(Rao and Paria 2013)
		<i>Venturia inaequalis</i>	Apple scab disease.	
ZnONPs	<i>Parthenium hysterophorus</i> L	<i>F. culmorum</i>	-	(Gunalan et al., 2013; Rajiv et al., 2013)
		<i>A. niger</i>	-	
		<i>A. flavus</i> ,	-	
	<i>P. hysterophorus</i> L	<i>Sclerotinia homoeocarpa</i>	Dollar spot of turfgrasses	(Li et al., 2017)
	<i>Nyctanthes arbor-tristis</i>	<i>A. alternate</i> ,	-	(Ingle et al., 2020)
		<i>A. niger</i> ,	-	
		<i>B. cinerea</i> ,	Grey mould disease	
		<i>F. oxysporum</i>	-	
		<i>Penicillium expansum</i> ,	Blue mould	

Towards Eco-Friendly Nanofungicides

Nanobiotechnology also belongs to the ecological and conservative methods of managing diseases caused by fungi in crops using biocontrol agents without damaging augments on crop safety (Álvarez et al., 2016). Nanohybrid antifungals are gaining popularity as they act through dual modes involving mixtures of NMs and biopolymers (Hashim et al, 2024). Numerous metal-based NMs have emerged, such as titanium, palladium, gold, zinc, copper, iron, aluminum and silver. However, silver SNPs have shown effective because to their strong antifungal efficaciousnes (Guo et al., 2003). CuNPs were synthesized by chemically reducing Cu 2+ in the presence of

isopropyl alcohol and cetyl trimethyl ammonium bromide showed strong antifungal action against plant pathogenic fungi, *Curvularia lunata*, *A. alternata*, *F. oxysporum* and *Phoma destructiva*, (Kanhed et al., 2014). Applying SNPs to soil or seed/seedlings not only controls the phytopathogen but also promotes plant growth through different pathways (Patel et al., 2014). Biosynthesize SNPs with white radish exhibit wide-spectrum antifungal action against *F. graminearum*, *F. oxysporum*, *F. solani*, and *Penicillium expansum* (Safaa et al., 2015). Gopinath and Velusamy (2013) found that AgNPs synthesized with the *Bacillus* sp. can effective against *Fusarium* sp. Similarly, Krishnaraj et al., (2012) found that AgNPs synthesized with *Acalypha indica* leaf extract as a reducer, were effective against several fungal pathogens affecting plants. These included *A. alternata*, *B. cinerea*, *C. lunata*, *R. solani* *M. phaseolina*, and *S. sclerotiorum*,

Polymer-Based and Nanohybrid Antifungals:

Polymer-based and nanohybrid antifungals have emerged as promising solutions for the management of fungal diseases. Nanohybrid antifungals composed of nano-materials in combination with biopolymers have been synthesized to curb fungal pathogens (Alghuthaymi et al., 2021). The incorporation of bio-derived or biodegradable ingredients like maize oil, beeswax and lecithin or cashew gum, has effectively examined (Nguyen et al., 2012; Abreu et al., 2013). An fungicidal diffusion solution equipped by synthesizing O-chitosan nanoparticles were exhibited effective potential against various pathogenic fungi infecting plants. *Nigrospora sphaerica*, *N. oryzae*, *Botryosphaeria dothidea*, and *A. tenuissima* were shown to be chitosan-sensitive in a mycelium growth (Xing et al., 2016). Chitosan as sources for polymers extracted from the plant pathogenic *F. oxysporum* to combat against wilt disease of tomato were effective and also increases the productivity (Sathiyabama and Einstein 2015).

Mycogenic Nanoparticles

Mycogenic nanoparticles, produced by fungal biomass, have been identified as playing an important role in combating fungal plants diseases (Khan et al., 2019; Yadav et al., 2023). These NPs can be used in combination with other nano-technological approaches for improved pathogen diagnosis and management. The toxicity CuONPs effect cell defense by the provoking of defense mechanism, culminating into cells death or permanent destruction (Naz et al., 2020). In addition, myco-nanoparticles that are derived from nanoparticle by fungal strains have demonstrated great potential to be used in disease management of phytopathogens including phyto-pathogenic fungi (Alghuthaymi et al., 2015). For instance, myco-nanoparticles synthesized from *F. oxysporum* were found to possess fungicidal activity against different phyto-pathogenic fungi (Alghuthaymi et al., 2021).). In addition, mycogenic CuONPs synthesized from an aqueous cell-free extract of *P. glomerata* promise to be effective in controlling fungal diseases in plants (Ingle et al., 2023).

One particular challenge is the development of techniques for the controlled production of metal NPs with precise dimensions, shapes, and compositions. Regarding NPs synthesis, fungi are superior to other microorganisms due to produce an abundance of enzymes and easy for researchers to manipulate in *in-vitro* (Mandal et al., 2006; Mohanpuria et al., 2007). Furthermore; fungal mycelial fabric is more robust than plant stuff or bacteria when flow pressure and available in bioreactors (Narayanan et al., 2010). The majority of fungi are highly capable of wall-binding, intracellular metal absorption, and metal tolerance. A list of fungi that can synthesize NPs with their synthesis used and their mode of action against fungal pathogens is provided in Table 3.

Table 3. List of Mycogenic nanoparticles against fungal diseases

Fungus Spp.	Type	Average Size (nm)	Shape	Synthesis Used	Mode of action	Source
<i>Alternaria alternate</i>	Ag	20–60	-	Extracellularly	Inhibited spore germination	(Gajbhiye et al., 2009)
<i>Aspergillus japonicas</i>	Fe ₃ O ₄	82	-	Extracellular production of cell-free filtrate in fungus	Antifungal activity	(Abdeen et al., 2016)
<i>A. niger</i>	Au	13	spherical, elliptical	cell-free filtrate	Antifungal activity	(Fateixa et al., 2009)
<i>Beauveria spp</i>	Ag	25	-	extracellular fungal culture	Inhibited spore germination	(Bhadani et al., 2022)

<i>Candida utilis</i>	Ag	20-80	spherical	Synthesized extracellularly	Antifungal activity	(Šebesta et al., 2022)
<i>Macrophomina phaseolina</i>	Ag	20–60	-	Extracellularly	Inhibited spore germination	(Gajbhiye et al., 2009)
<i>Neurospora crassa</i>	Co	64	-	Extracellularly	-	(Rashmi et al., 2004)
<i>Pezizales</i>	Ag	19	irregular	Mycogenic fabrication	Antifungal activity	(Owaid et al., 2022)
<i>Picoa</i> ,	Ag	-	irregular	Mycosynthesis	Antifungal activity	(Dheyab et al., 2020)
<i>Rhizoctonia solani</i>	Ag	20–60	-	Extracellularly	Inhibited spore germination	(Gajbhiye et al., 2009)
<i>Trichoderma viridae</i>	Ag	5 - 40	rod, spherical	Cell-free filtrate	Antimicrobial activities	(Fayaz et al., 2010)
<i>Verticillium sp.</i>	Au	20	spherical, triangular, hexagonal, quasi-hexagonal	Synthesis intracellular	Antimicrobial activities	(Mukherjee et al., 2001)
<i>Volvariella volvacea</i>	Ag	20–150	-	Extracellular	Antifungal activity	(Philip, 2009)

Extracellular extracts of *R. stolonifer* were used to biologically synthesize AgNPs. This study demonstrated the cost-effectiveness of *R. stolonifer*-mediated synthesis and offered evidence that the particles of liquid mycelial extract of the plant assist the production of AgNPs. Additionally, both concentration and temperature can manage through AgNPs (Abdel-Rahim et al., 2017). Vala (2015) employed the marine derived fungus *Aspergillus sydowii* to show the bio synthesis of AuNPs. Honary et al., (2012) employed *Penicillium aurantiogriseum*, *P. citrinum* and *P. waksmanii* to illustrate the biosynthesis of CuONPs. Several physical parameters and metal salts concentration used during the synthesis process led to changes in the synthesis yield, size, and shape of the produced nanoparticles.

Furthermore, sulfur and copper nanoparticles suppress the post-harvest diseases of cucumber by affecting the growth of both *B. cinerea* and *Sclerotinia sclerotiorum* (Sadek et al., 2022). In addition, mycogenic nanoparticles produced from fungal biomass protect plant from pests and pathogens (Murali et al., 2021; Ram et al., 2023). For instance, cucurbit powdery mildew successfully managed was established if the nano-silica-silver particles were treated in the field at any time reaching up to 25 days. There has been success in managing the disease of rhizome rot caused by, the soil-borne oomycete *P. aphanidermatum* through GNPs namely β glucan that have been investigated as a resistance inducer in turmeric (Anusuya and Sathiyabama 2015). Anusuya and Sathiyabama (2015) demonstrated the efficacy of β -D glucan nanoparticles (GNPs), finding that they significantly reduced the incidence of rot and provided 77% protection against *Curcuma longa* (syn *C. domestica*). New biohybrid nanofungicide materials can be utilized as an environmentally friendly antibacterial against various fungal phytopathogenic organisms.

Nanobiosensors for Fungal Plant Pathogen Detection

Nanobiosensors have emerged as the potential detectors for identification of fungal plant pathogens. Nanobiosensors are the devices that use nanotechnology to determine and count the level of biological molecules including those produced from the fungal pathogens. The precise quantum dot-based nanosensor has been established for revealing *Candidatus Phytoplasma aurantifolia* in lime with low concentrations (Khiyami et al., 2014). Additionally, the causing agent of tomato and pepper spot diseases *Vesicatoria*, can potentially be quickly identified using fluorescent silica nanoparticles linked with antibody molecules (Dyussebayev et al., 2021). Application of nanomaterials in enzyme immobilization is one example of the use of nanosensors (Kim et al., 2006). However, DNA-based electrochemical nanobiosensors have been reported for the detection of *Phytophthora palmivora*, which causes black pod rot in cacao pods (Dyussebayev et al., 2021).

There are diverse approaches to plant diseases diagnosis, however, each method has its own limitations. Conventional methods that are based on cultivating bacteria are time-consuming; techniques relying on antibodies such as ELISA are highly specific but they are very expensive; while polymerase chain reaction is the most expensive but it enhances accuracy through analysis of genetic material (Gao et al., 2004; Doorn et al., 2007). A novel approach employs NPs to identify microorganisms, which is revolutionizing agriculture. This method is based on using silica nanoparticles i.e. small particles sized less than 500 nm which contains fluorescence dye and is conjugated with antibodies that bind to specific pathogens thereby making it suitable for identifying plant pathogens. This indicates that the use of nanobiosensors can contribute to the early detection and quantification of fungal plant pathogens, promoting sustainability and innovation in modern agronomic approaches

CONCLUSION

In this review, a critical and comprehensive analysis of the existing advancement on the successful application of NPs for controlling fungi diseases in plants was presented. The emergence of nanotechnology have shown promising tool for advance pathogen diagnostics with high accuracy and reliability. NPs also has exhibited several novel characteristics in their mechanisms of action such as cell wall damage, lipid peroxidation, protein denaturation, DNA damage and destruction of mitochondria to combat fungal plant diseases. These studies found that alternation in synthesis method of NPs have shown exceptional antifungal activities. The studies also discussed future prospects along with several success that have been achieved so far. Nevertheless, further studies are required to establish the long-term consequences and potential for scaling up of these nanotechnologies in agriculture.

In general, nanotechnology has revolutionized crop production through enhancement of plant growth, disease resistance, and nutrient utilization by controlled delivery of agrochemicals. These results provide evidence of the possible benefits of nanotechnology in combating fungal disease in crops. The future of plant disease management by nanotechnology is bright as nanoparticles with smaller sizes have shown higher effectiveness in managing diseases caused by bacteria, fungi, virus, and nematodes. Developing novel nanomaterials and innovative control methods for plant diseases in agriculture will lead to sustainability of agriculture and food security.

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AUTHOR CONTRIBUTIONS

All authors contributed equally to this research.

COMPETING OF INTEREST

The authors declare no competing interest.

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