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Mycogenic synthesis of different nanoparticles by endophytic fungi hosted higher plants

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ABSTRACT

Nanotechnology is a field that studies matter at the atomic and molecular scale, dealing with matter at the 1 billionth (i.e., 10^{-9} m = 1 nm) of a meter scale. A nanoparticle is the fundamental component in nanostructure fabrication, smaller than everyday objects but larger than atoms or simple molecules. Nanoparticles, typically ranging from 1 to 100 nm in size, possess unique physical and chemical properties compared to bulk metals. These properties, such as lower melting points, higher surface areas, optical properties, mechanical strengths, and magnetizations, make them attractive for various industrial applications. However, the definition of nanoparticles and nanomaterials varies depending on the specific application. This review addresses the use of endophytic fungi from higher plants in the mycogenic synthesis of various nanoparticles, highlighting their unique biological capabilities as eco-friendly and sustainable nanofactories. This review emphasizes how these fungi use their diverse metabolic pathways to produce nanoparticles with unique physicochemical properties that can be used in medicine, agriculture, and environmental remediation. Endophytic fungi can produce stable and biocompatible nanoparticles under mild conditions, but the review also discusses the challenges and future of this emerging field. This review seeks to explain how endophytic fungi can revolutionize nanoparticle synthesis and global problem-solving.

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Introduction

Nanoscale metals are crucial in fields like environment, medicine, and engineering. Currently, chemical methods are used, leading to environmental pollution, energy consumption, and health issues. Green synthesis, which uses plant extracts instead of industrial chemicals, has been developed to address these challenges. This approach is more cost-effective, reduces pollution, and enhances environmental and human health safety compared to traditional chemical synthesis (Ying et al. 2022). Nanometallic materials are metals and alloys with nanocrystalline grains ranging from 5 to 100 nm in size. These materials have larger surface areas and unique physical and chemical properties due to small size, surface, interface, and quantum effects that not found in non-nano metals. They have wide applications in biology, medicine, and engineering due to their wide range

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(Hussain et al. 2016; Zhao et al. 2016). Physical and chemical methods are gradually being replaced by green synthesis methods, Green synthesis is a clean, safe, cost-effective, and environmentally friendly process for creating nanomaterials using microorganisms like bacteria, yeast, fungi, algal species, and certain plants (Huston et al. 2021). Green synthesis offers numerous benefits over chemical and physical methods, including being non-toxic, pollution-free, environmentally friendly, economical, and more sustainable. However, it faces challenges in extracting raw materials, reaction time, and product quality. Issues include limited availability of raw materials, long synthesis times, and homogeneous particle size (Ying et al. 2022). One of the most used microorganisms in the process of the green synthesis of nanoparticles are endophytic fungi which found in plant tissues without causing damage, they are abundant bioactive compound producers with potential applications in pharmaceuticals, agriculture, and industry (Kamunkar & Nischitha, 2025). Endophytic fungi secrete numerous bioactive metabolites and antimicrobial compounds, enabling the synthesis of nanoparticles. They are preferred over bacteria due to their high protein yield, large biomass handling capacity, and optimal growth of mycelium with a large surface area. These fungi tailor their elemental compositions to suit the elemental compositions of their environment. Fungal biomass and supernatant are used as a reduction medium for nanoparticle formation from the precursor solution (Vijayanandan & Balakrishnan, 2018).

Nanoparticles and nanotechnology

Nanotechnology is a domain of research and technological advancement at the molecular, macromolecular and atomic levels, concentrating on the examination of structures and precise manipulation and devices measuring between 1 and 100 nanometers. Nanoparticles, with their small size, surface tailorability, improved solubility, and multifunctionality, offer new research avenues for biologists. These nanomaterials can engage with intricate biological processes, functioning at the biomolecular level. This swiftly advancing domain enables interdisciplinary researchers to engineer and create multifunctional nanoparticles capable of diagnosing, targeting and treating diseases such as cancer. (McNeil, 2005). Green synthesis is a bottom-up approach to producing nanoparticles (NPs) by replacing expensive chemical reducing agents with natural products like leaves or fruits. This method is eco-friendly, sustainable, and less expensive, making it suitable for mass production (Gopinath et al. 2014; Iravani, 2011; Jayaseelan et al. 2012). It also allows the recycling of expensive metal salts like gold and silver found in waste streams, which have limited resources and fluctuating prices (Wang et al. 2009). Nanoparticles (NPs) can be synthesized using either a "Top Down" or "Bottom Up" approach. The "Top Down" method involves size reduction through physical and chemical methods. The "Bottom Up" approach produces NPs from small entities like atoms and molecules, resulting in minimal defects and homogenous chemical composition (Hussain et al. 2016). There is also The Intracellular and Extracellular synthesis of nanoparticles which we use a microorganism like fungi as a method for the synthesis process. Fungi use intracellular synthesis to form nanoparticles by transporting ions into microbial cells, forming them with enzymes. These nanoparticles are smaller than extracellularly reduced nanoparticles, likely due to the nucleation process within the organism while on the other side Extracellular synthesis of nanoparticles by fungi offers more applications than intracellular synthesis due to the absence of unnecessary cellular components. Fungi's secretory components are responsible for the reduction and capping of nanoparticles, making it a more efficient method (Hasan, 2015).

Synthesis of silver nanoparticles

Silver nanoparticles attracted the attention of various researchers worldwide viz: Abu El-Saoud et al. (2015), Abu El-Saoud & Abdel-Azeem (2020), Abdel-Azeem et al. (2020) and Mossa et al (2024). In a study carried out by Balakumaran et al. (2015), they were synthesized silver nanoparticles in vitro using endophytic fungi isolated from medicinal plants to investigate their biological activities. Thirteen fungi were analyzed, but *Guignardia mangiferae* generated highly stable and well-dispersed silver nanoparticles under adjusted reaction conditions extracellularly in 12 hours. Nine leaf specimens were obtained from nine medicinal plant species. The medicinal plants included *Ocimum sanctum*, *A. indica*, *Catharanthus roseus*, etc. The research entailed cleansing leaf samples, segmenting them into small pieces, and isolating endophytic fungi through a modified methodology (Kamalraj & Muthumary 2012; Schulz et al. n.d.). The segments were sterilized and positioned on Petri dishes with PDA medium. The plates were incubated at 23 above or under 2 °C for a duration from four to seven days, during which fungal growth was observed. Fungal colonies were isolated, purified, and preserved through sub-culturing. Thirteen distinct endophytic fungi were isolated from the leaf samples. The study assessed 13 endophytic fungi for the production of silver nanoparticles. The fungi had been cultivated in potato dextrose broth under aerobic conditions and incubated for seven days at 27°C. Subsequent to incubation, the fungal mat had been washed with sterile distilled water "douple distilled" and kept for 48 hours under agitation. The mycelial-free filtrate subsequently reacted with silver nitrate, resulting in a silver ion concentration of 10−3 Molarity. The color alteration had been noted for up to 48 hours following the reaction (Fayaz et al. 2010). The color change in the appropriate medium for silver nanoparticle synthesis indicates the mycosynthesis of these nanoparticles, while the substrate solution and fungal biomass maintain their original color (Song et al. 2009). The silver nanoparticles derived from fungal endophytes had been characterized through multiple techniques, including HR-TEM, SAED analyses, UV-vis spectrophotometry, EDX and XRD, exhibiting a distinctive peak of absorption at 417 nm (Figure 1, Figure 2) (Balakumaran et al. 2015).

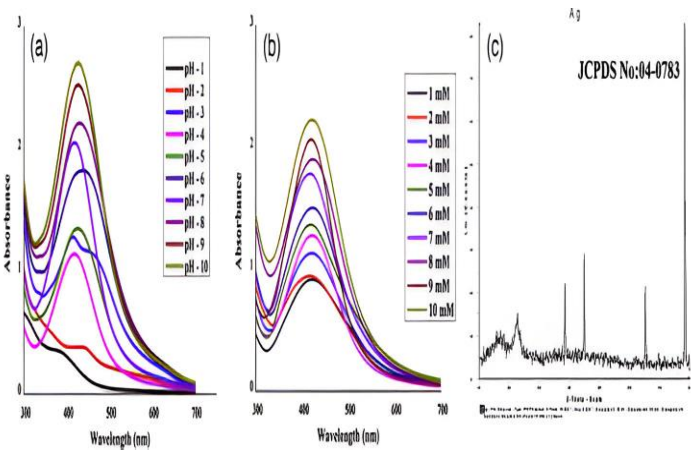

Fig 1. Characterization and Optimization of silver nanoparticles that had been synthesized by *G. mangiferae*., A refer to Influence of pH on the mycogenic production of silver nanoparticles. UV-visible absorption spectrum obtained under varying pH conditions (post color alteration), B refer to Impact of silver nitrate on the mycogenic production of silver nanoparticles. UV–visible absorption spectrum obtained at varying concentrations of silver nitrate from 1 to 10 mM (following color alteration), B refer to the X-Ray Diffraction pattern of *G. mangiferae*-derived silver nanoparticles is indexed at (1 1 1), (2 0 0), and (2 2 0), demonstrating the facets of crystalline silver. (Balakumaran et al. 2015).

In a separate study, a team of scientists initiated research on endophytic fungi associated with the Loranthaceae family and their capacity to synthesize silver nanoparticles. Furthermore, they utilized their findings to develop an agent with antioxidant, anti-diabetic, and anti-cholinesterase properties. This research seeks to synthesize silver nanoparticles (AgNPs) utilizing the aqueous extract of *Cladosporium* species derived from the healthy leaves of *Loranthus micranthus* and to examine their antioxidant, anti-diabetic, anti-acetylcholinesterase (AChE), and anti-butyrylcholinesterase (BChE) activities.

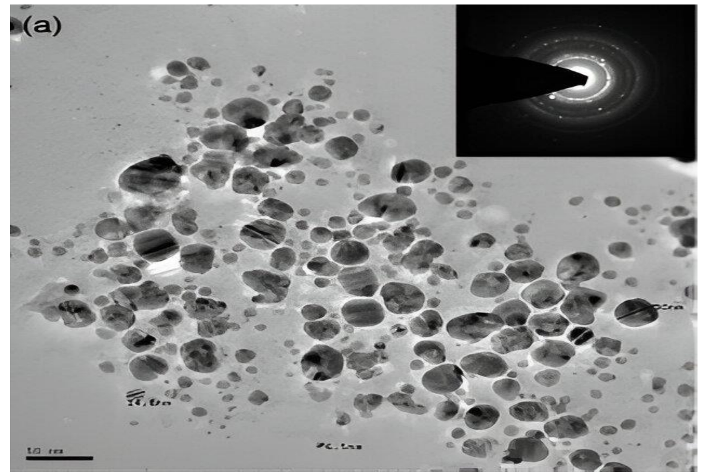

Fig 2. Transmission electron microscopy image of spherical silver nanoparticles in high resolution, measuring 8:20 nm in size "scale bar equal to 50 nm" with an image displaying the selected area electron diffraction (SAED) pattern (Balakumaran et al. 2015).

The nanoparticles were characterized by utilizing UV-Vis spectrophotometry, XRD, FTIR, and EDX techniques. (Popli et al. 2018). The *Cladosporium* species endophytic fungi was obtained from a stock culture at Dayananda Sagar College of Engineering in India. The fungi were mass-cultured in Potato Dextrose Broth for 15 days, subsequently dried and milled. The resultant AgNPs were examined for the presence of phytochemicals, such as carbohydrates, alkaloids, saponins, amino acids, proteins, diterpenes, phenols, phytosterols and tannins. The extract was heated to 100°C and combined with $AgNO_3$ solution, noting the color transition from light green to dark brown. (Farnsworth, 1966). The UV-2450 spectrophotometer measured the reduction of pure silver ions in the reaction medium after 5 hours, subsequently diluted with distilled water. The absorption spectrum of the incubated CsAgNPs solution at 438 nm after 30 minutes confirmed the formation of AgNPs, aligning with prior studies by (Li et al. 2012; Rajakumar et al. 2017), The intensity of the absorption peak escalates with prolonged incubation time. The research employed a Hitachi S-4500 scanning electron microscope to examine the morphological attributes of a sample. Thin film samples were fabricated on a carbon-coated copper grid, and the excess solution was eliminated. The films were subjected to drying under a mercury lamp for a duration of 5 minutes. on the other side X-ray diffraction (XRD) was employed to assess the formation and quality of CsAgNPs. The pattern obtained from drop coating film on a glass substrate was documented using a Philips PW 1830 instrument at a voltage of 40 kV and a current of 30 mA, utilizing Cu-Kα radiation. The XRD analysis of CsAgNPs demonstrated a crystalline structure, with diffraction peaks at 2θ signifying the face-centered cubic (fcc) arrangement of the AgNPs. The peaks result from the incorporation of AgNO3, utilized in the synthesis of silver nanoparticles. The widening of Bragg's peaks signifies the emergence of nanoparticles, averaging 24 nm in size. The XRD patterns indicate that the bioorganic phase crystallizes on the surface of the AgNPs (figure 3) (Popli et al. 2018).

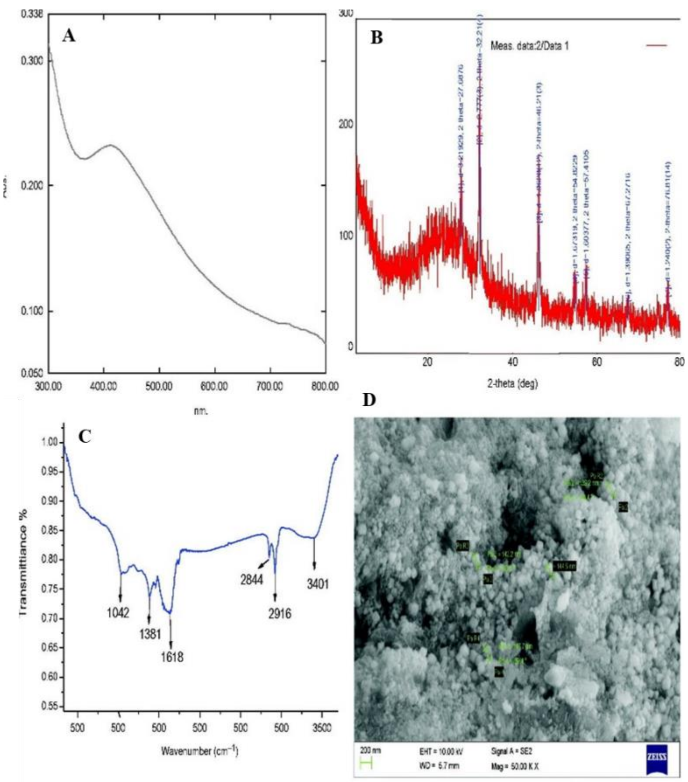

Fig 3. Characterization of silver nanoparticles synthesized from *Cladosporium* species. A refer to UV-Vis spectra, B refer to XRD pattern, C refer to IR-spectra, D refer to SEM images at 50KX magnification (Popli et al. 2018).

Synthesis of gold nanoparticles

A group of scientists conducted research on the antimicrobial and antifungal properties of gold nanoparticles (AuNps). The study aimed to evaluate the effectiveness of AuNPs as a new antimicrobial agent for controlling the activity of pathogens. The extracellular synthesis of AuNPs was performed using *Phoma* sp. as an endophytic fungus obtained from the vascular tissue of *Prunus persica* in Kerman province, Iran. Multiple samples were obtained from various regions of peach trees, from which *Phoma* sp. was isolated as an endophytic fungus utilized in this study for the extracellular synthesis of Gold Nanoparticles. The fungal biomass was grown in a medium of potato dextrose broth "PDB", and for 48 hours and temperature of 28°C, the mycelia had been incubated with HAuCl4 solution. The HAuCl4 ions were reduced to AuNPs via overnight exposure to fungal biomass. The hue of the fungal biomass transformed to red following 48 hours of immersion in the solution (Soltani Nejad et al. 2022).

The research employed 1 mL of supernatants for UV-Vis analysis within the 450-700 nm wavelength range following centrifugation at 4000 rpm for 10 minutes. The analyzed sample exhibited a significant peak at 530 nm (Figure 4). (González-Ballesteros et al. 2021), indicating AuNPs. The XRD pattern exhibited four distinct peaks at specific 2θ diffraction angles. The analysis verified that AuNPs were synthesized by *Phoma* sp. mycelial biomass, exhibiting a crystalline structure indicated by the distinctness of the peaks (Figure 5) (Khatami et al. 2016).

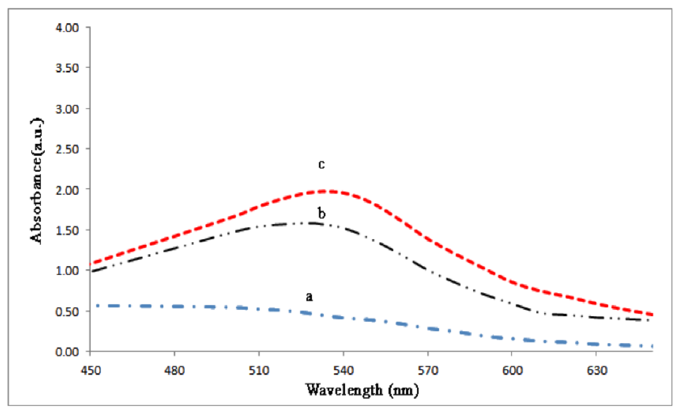

Fig 4. UV–Vis spectra of extracellular synthesized AuNPs by *Phoma* sp. mycelial biomass at different times: a, b and c are control, 24 h and 48 h after biosynthesis reaction, respectively (Soltani Nejad et al. 2022).

Another study deals with the synthesis of gold and zinc nanoparticles from endophytic fungi isolated from *Eucalyptus sideroxylon*, The research investigates the eco-friendly synthesis of gold nanoparticles (Au NPs) utilizing the cell filtrate of *Fusarium chlamydosporum* derived from *Eucalyptus sideroxylon* leaves. The environmentally sustainable nanoparticles were evaluated against cancerous cell lines and multidrug-resistant bacteria. The synthesized gold nanoparticles were characterized through UV-Vis spectroscopy, dynamic light scattering, X-ray diffraction, energy-dispersive X-ray spectroscopy and transmission electron microscopy (Hammad et al. 2025).

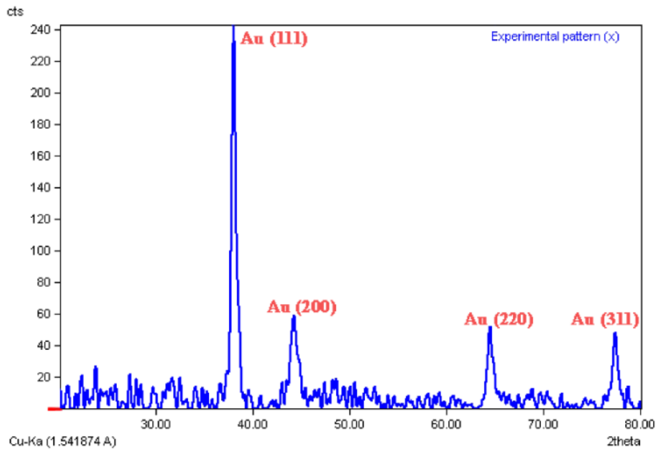

Fig 5. X-ray diffraction (XRD) spectrum of AuNPs synthesized by the biomass of *Phoma* sp (Soltani Nejad et al. 2022).

The research validates the presence of metallic gold nanoparticles exhibiting a significant absorption peak at 530 nm (Figure 6), consistent with prior studies (Iranmanesh et al. 2020; Doghish et al. 2022).

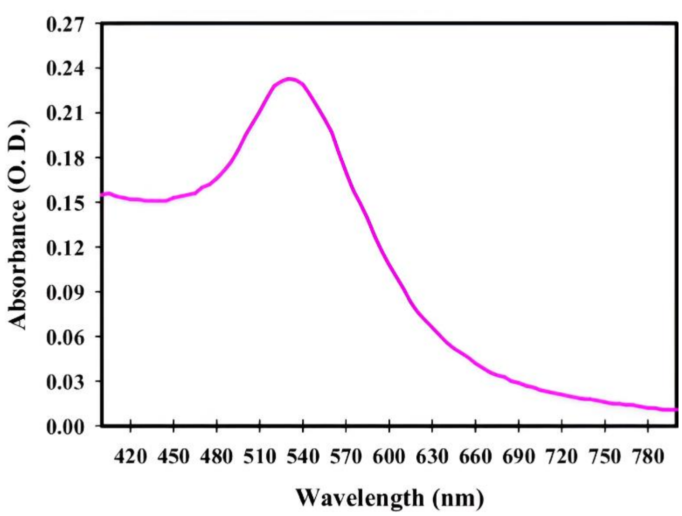

Fig 6. UV-Vis. spectrum of Au NPs.

The XRD pattern of Au nanoparticles exhibits the face-centered cubic (fcc) structure of gold, with diffraction peaks analogous to those of standard gold metal (Figure 7). The nanoparticles are predominantly crystalline, as verified by (Soni & Prakash, 2012).

Biosynthesized gold nanoparticles are promising antimicrobial and anticancer biomaterials, addressing the limitations of other bactericidal agents and resistant pharmaceuticals. Their distinctive combinations and methodologies will facilitate a promising future in infection and cancer treatments. Further investigation is required to comprehensively elucidate the mechanisms responsible for the antibacterial and anti-cancer properties of these biomaterials, along with their potential genotoxic effects (Hammad et al. 2025).

Synthesis of copper nanoparticles

Copper oxide nanoparticles (CuO-NPs) are garnering attention due to their cost-effectiveness, thermal stability, low toxicity and chemical stability, biocompatibility, high catalytic efficiency and compatibility with various materials for polymer synthesis. (Ssekatawa et al. 2022).

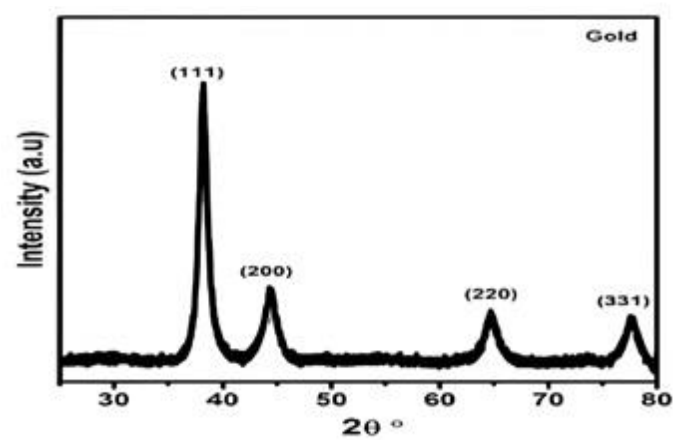

Fig 7. XRD analysis for AuNPs.

These nanoparticles possess diverse applications across multiple domains, including antibacterial, antifungal, anticancer, and anti-helminthic properties, in addition to their use in medical textiles, agriculture, and photocatalytic processes in addition to that CuO-NPs are more economically viable and exhibit lower toxicity than Ag-NPs in biomedical and biotechnological applications. (Dobrucka 2018; Peddi et al. 2021). A fungal strain designated BR.1 had been isolated from the root of a healthy *Allium sativum*. The samples of the root had been procured from land of agricultural in Governorate of El-Menofa, Egypt. The isolation process involved washing the samples with tap water and performing surface sterilization. The strain of endophytic fungal "BR.1" had been introduced into 100 millileter of potato dextrose broth media and it had been incubated for a duration of 5 days. The resultant fungal biomass was harvested via centrifugation at 10 K rpm for 5 minutes and subsequently rinsed with deionized water. Ten grams of biomass were subsequently combined with dissolving water for a duration of 24 hours under dark conditions while agitated. The supernatant was harvested and utilized as a biocatalyst for the biosynthesis of CuO nanoparticles (Nassar et al. 2023a). The biosynthesis of CuO nanoparticles utilizing fungal biomass filtrate was accomplished by dissolving 100 Microgram of $Cu(CH_3COO)_2 \cdot H_2O$ in specific amount of distilled water "5 milliliter" and combining it with amount equal to 95 milliliter of fungal biomass filtrate. The mixture was agitated for one hour at 100 rpm and adjusted to a pH of 8. The color change from colorless to greenish signifies the formation of copper oxide nanoparticles. The nanoparticles had been gathered, rinsed, and subjected to calcination at 200 °C for a duration of 2 hours (Consolo et al. 2020). The greenish color resulting from the amalgamation of fungal biomass and metal precursor was quantified using UV-Vis spectroscopy across a wavelength range from 200 nm to 800 nm. 2 milliliter of synthesized solution had been filled with a cuvette made of quartz, and at regular intervals the absorbance had been measured to investigate the peak resonance of the surface plasmon (Figure 8) (Peddi et al. 2021).

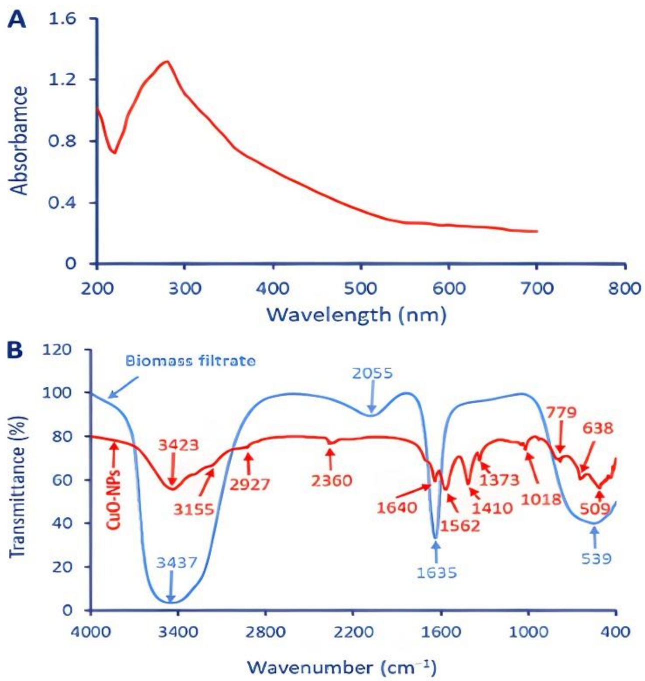

Fig 8. A depicts the UV-vis spectroscopy of copper oxide nanoparticles that had been synthesized by the strain *A. terreus* "BR.1" of endophytic fungal demonstrates maximum surface plasmon resonance at 280 nm wavelength; B presents the FT-IR spectrum of filtrate of fungal biomass in comparison to biosynthesized copper oxide nanoparticles, indicating various functional groups (Nassar et al. 2023a).

Transmission Electron Microscopy (TEM) and Energy Dispersive X-ray Spectroscopy (EDX) are frequently employed to identify the morphological properties of synthesized nanomaterials (Nassar et al. 2023b). The synthesis of CuO nanoparticles mediated by fungi demonstrates a spherical morphology and organized structure, with dimensions between 15 and 55 nm, averaging 39.6±11.1 nm (Figure 9).

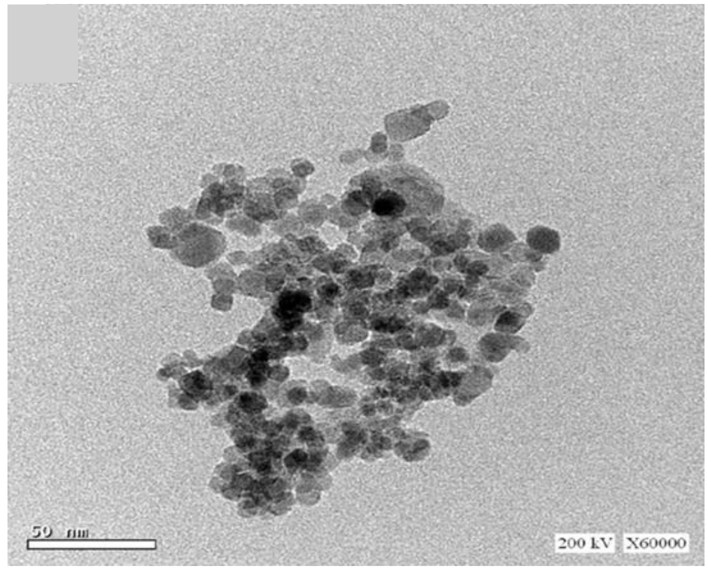

Fig 9. Transmission Electron Microscopy (TEM) micrograph showing spherical shape of copper nanoparticles (Nassar et al. 2023).

Green synthesized CuO nanoparticles exhibit significant efficacy against pathogenic bacteria, including both Gram-negative and Gram-positive strains, like *Candida* species. They demonstrate significant biocompatibility and a low IC50 value against cancer cells (Nassar et al. 2023a).

Synthesis of Zinc Nanoparticles using Endophytic fungi hosting higher plants

Another study that's seeks for the green synthesis of Zinc Nanoparticles in order to use its ability as an antimicrobial agent, The research sought to synthesize zinc oxide nanoparticles (ZnO NPs) utilizing the endophytic fungal extract of *Aspergillus niger*, which was isolated from the healthy leaves of *Acalypha hispida* Burm, collected from the Faculty of Pharmacy farm at Tanta University, Egypt. The synthesized ZnO nanoparticles were characterized, and their antibacterial efficacy was examined both in vitro and in vivo (Abdelkader et al. 2022). The optical characteristics of A. niger synthesized ZnO nanoparticles were examined utilizing UV-visible spectrophotometry within the wavelength range of 200-600 nm. The research validated the synthesis of zinc oxide nanocrystals exhibiting a peak wavelength of 330-390 nm (Figure 10), aligning with prior investigations (Alotaibi et al. 2022; Sumanth et al. 2020).

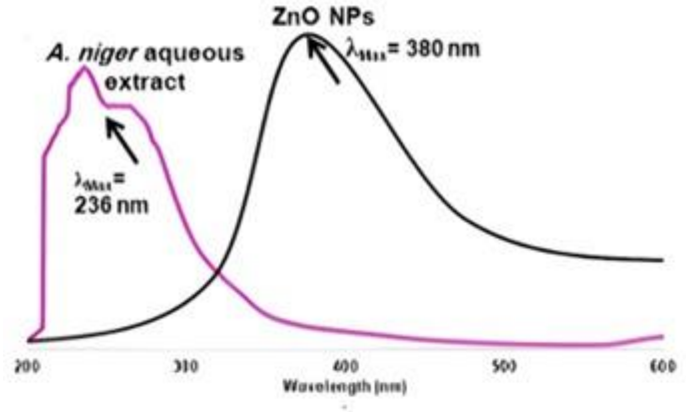

Fig 10. UV spectrum analytical spectra of ZnO NPs synthesized from *A. niger* (Abdelkader et al. 2022).

The crystalline phase of ZnO nanoparticles was determined utilizing an APD 2000 PRO X-Ray Diffractometer with Cu Kα radiation at a wavelength of 1.5406 nm. The XRD pattern was obtained at 2 Theta, and the external surface morphology was examined using a scanning electron microscope. The gold coating was applied utilizing SPIMODULE TM. The elemental composition of ZnO nanoparticles was examined using EDX. Following appropriate dilution, droplets of ZnO nanoparticle suspension were affixed to a carbon grid and subsequently dried prior to analysis using a transmission electron microscope (Alotaibi et al. 2022). The XRD spectrum exhibited no discernible peaks for additional impurities, thereby affirming the purity of hexagonal nanocrystals predominantly consisting of Zinc oxide (Figure 11) according to Sumanth et al. (2020).

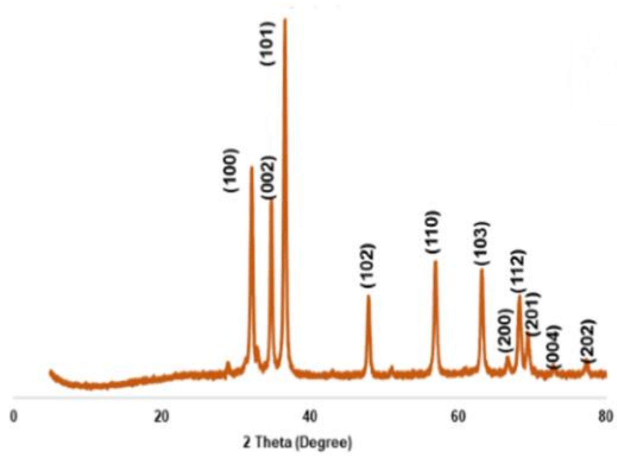

Fig 11. XRD pattern analytical spectra of ZnO NPs synthesized from *A. niger* (Abdelkader et al. 2022).

ZnO nanoparticles demonstrated antibacterial efficacy against S. aureus isolates, exhibiting MIC values between 8 and 128 μg/mL. Their diminutive size enables interaction with microbial surfaces, facilitating entry into microbial cells and exhibiting antimicrobial activity. Biofilms, which are communities of bacterial cells, are present on surfaces such as implanted medical devices and exhibit a resistance to antibiotics that is 1000 times greater than that of planktonic cells. These biofilms can result in chronic and persistent infections, rendering conventional antibiotic therapy ineffective. The study demonstrates that ZnO nanoparticles (NPs) exhibit antibiofilm properties, particularly against *S. aureus*, by reducing the prevalence of strong and moderate biofilm-forming cells from 50% to 20.83% and downregulating biofilm-associated genes in 29.17% of *S. aureus* isolates, underscoring their significance in the management of bacterial infections (Sharma et al. 2019; Zhang et al. 2020). The research illustrates the biogenic production of ZnO nanoparticles utilizing *A. niger* fungal filtrate as an economical and non-toxic approach. The nanoparticles were analyzed through multiple techniques and demonstrated antibacterial efficacy against clinical isolates of *S. aureus*. They suppressed biofilm formation and downregulated biofilm-associated genes. In vivo, they diminished congestion and fibrosis in hepatic and splenic tissues. These nanoparticles may serve multiple pharmaceutical and biological applications (Abdelkader et al. 2022).

In another study, Zinc oxide nanoparticles have been successfully produced using the filtrate of the culture of endophytic fungus *Alternaria tenuissima* via a eco-friendly, economical and rapid method. The manufacturing of zinc oxide nanoparticles had been rapidly accomplished, exhibiting a single-phase crystalline structure and considerable stability. The samples were examined for their antimicrobial, anticancer, antioxidant, and photocatalytic properties. ZnONPs demonstrated extensive antimicrobial efficacy against both plant and human pathogens, suppressing cell proliferation in human melanocytes, as well as breast and liver cancer cells. They demonstrated significant antioxidant potential with a 50% inhibitory concentration ranging from 102 to 13 μg ml−1. Furthermore, ZnONPs effectively degraded methylene blue dye. The synthesis was validated by UV-Vis spectroscopy after a duration of 20 minutes (Abdelhakim et al. 2020).

Nanoparticles as an antimicrobial agent

In a study that work on the green synthesis of zinc nanoparticles, a group of scientists start to study its ability as an antibacterial agent whether on human or an animal, This research included 30 Swiss albino mice, each weighing between 22 and 25 grams, sourced from the Faculty of Veterinary Medicine at Cairo University. The mice had been housed in cages with filtered water and standard pellets, maintained at 25◦C "above or under 2 degress" with a 12-hour light/dark cycle (H. Zhang et al. 2020), and acclimatized for seven days. The study encompassed infected mice administered with *S. aureus* suspension for two days, subsequently categorized into three groups, group I was untreated, group II was gentamicin-treated, and group III was ZnO NPs-treated. Treatments were delivered intraperitoneally over a duration of 20 days. All mice were anesthetized and subsequently euthanized through cervical dislocation. Blood samples and tissue samples from the liver and spleen were collected. CFU per g measurements were also obtained from the tissues (Alotaibi et al. 2022; Elmongy et al. 2022). ZnO nanoparticles markedly reduced the colony-forming units (CFU/g) in liver and spleen tissues, while enhancing the survival rate of infected mice. In group I, two mice succumbed after five days, three after eight days, and two after fifteen days, whereas in groups II and III, only two died after one week and one after nine days (Abdelkader et al. 2022).

In another study that focus on the synthesis of silver nanoparticles of endophytic fungi *Curvularia lunata* isolated from the leaves of *Catharanthu sroeus*, they start to test its antimicrobial activity, The research assessed the antimicrobial efficacy of synthesized AgNPs against gram-negative and gram-positive human pathogens utilizing the disc diffusion method. Cultures had been preserved at -80°C and subcultured for 24 hours in Nutrient Broth. AgNPs had been deposited onto sterile paper discs and incorporated into the Muller-Hinton agar medium. Dosages had been determined based on laboratory data, and the zones of inhibition had been assessed after incubation at 37°C for 24 hours. The synthesized AgNPs exhibited antimicrobial efficacy against several human pathogens, including Bacillus subtilis, *E. coli*, *Salmonella paratyphi*, *Pseudomonas aeruginosa*, *Bacillus cereus* and *Staphylococcus aureus*. There was a significant increase in the inhibition zone of various antibiotics against test strains in the absence and presence of AgNPs at a concentration of 10 µg/disk for both gram-positive and gram-negative bacteria (Ramalingmam et al. 2015).

Another study that focusing on the green synthesis of gold nanoparticles from endophytic fungi *phoma* sp isolated from *Prunus persica*, a group of scientists start to study its antimicrobial activity. A pathogenic isolate of *Rhizoctonia solani* and *Xanthomonas oryzae* AG1-IA had been obtained from the Biological Control Laboratory. The antibacterial efficacy of AuNPs was assessed through the disk diffusion method against *Xanthomonas oryzae*. The bacterium had been cultivated on NA plates and inhibited by 6 mm disks saturated with 30 µL of AuNPs solution. After 24 hours, the inhibition zone was quantified. An in vitro assay was conducted on PDA media treated with 30 µL of synthesized AuNPs solution to examine the antifungal activity and inhibitory effect on the mycelial growth of *R. solani* (Akhlaghi et al. 2020; Muniyappan et al. 2021). The bioassay tests were conducted using well-diffusion method, and the radial growth of fungal mycelium was evaluated. Every set of tests was conducted thrice. The diffusion of AuNPs resulted in inhibition zones surrounding the disks, clearly demonstrating the antifungal and antibacterial properties of AuNPs. The biosynthesis of AuNPs via fungi presents prospects for creating sustainable and eco-friendly nanoparticles with potent antibacterial characteristics. This study examines the advancement of nanotechnology-based agents for enhancing crop performance, resistance, and disease management (Soltani Nejad et al. 2022).

Nanoparticles as an anticancer and antitumor agent

A study that focus on the synthesis of gold nanoparticles of endophytic fungi *fusarium solani* isolated from *Chonemorpha fragrans*, a group of scientists start to study its anticancer and antitumor abilities, The research examined the anticancer effects of nanoparticles on HEK, HeLa, and MCF-7 cells, mitigating bacterial and fungal contamination through the addition of penicillin and streptomycin and cultured at 37°C with 5% CO2 in a CO2 incubator (Ramar et al. 2011; Vivek et al. 2011). The MTT assay had been employed to determine the inhibitory concentration (IC50) of the synthesized nanoparticles. To achieve 75% confluence, three cancer cell lines had been cultured in a microtitre plate for 48 hours. Subsequently, MTT was introduced, followed by incubation at 37°C for 4 hours, after which approximately 50 µL of DMSO was added and for 10 minutes it had been incubated. at 620 nm, The absorbance had been quantified utilizing a microtitre plate reader, and the percentage of viability was determined. Conversely, MCF-7 cells were subjected to NP treatment at varying concentrations, subsequently fixed with acetic acid and ethanol, and then for morphological examination it had been mounted on a glass slide. The research investigated the anticancer efficacy of synthesized nanoparticles on Human Embryonic Kidney cell lines, HeLa and MCF-7 utilizing the MTT assay (Clarance et al. 2020). The findings indicated that nanoparticles exhibited anticancer efficacy on HeLa cell lines, with an IC50 value of 1.3 above or under 1.0 µg per ml, and 0.8 above or under 0.5 µg per milliliter against the MCF-7 cell line. Nevertheless, minimal activity had been observed in the Human Embryonic Kidney (HEK) cell line. The morphology of MCF-7 cells had been examined using inverted phase contrast microscopy. The results indicated that cells treated with synthesized nanoparticles exhibited low density, increased condensation, irregularity, and fragmentation attributable to apoptotic cell death. The research indicated that gold NPs produced cytotoxic reactive oxygen species (ROS) upon light exposure (Valsalam et al. 2019; V.t et al. 2019), resulting in cellular damage and microbial cell death (Figure 12) (Paramanantham et al. 2018; Parasuraman et al. 2019).

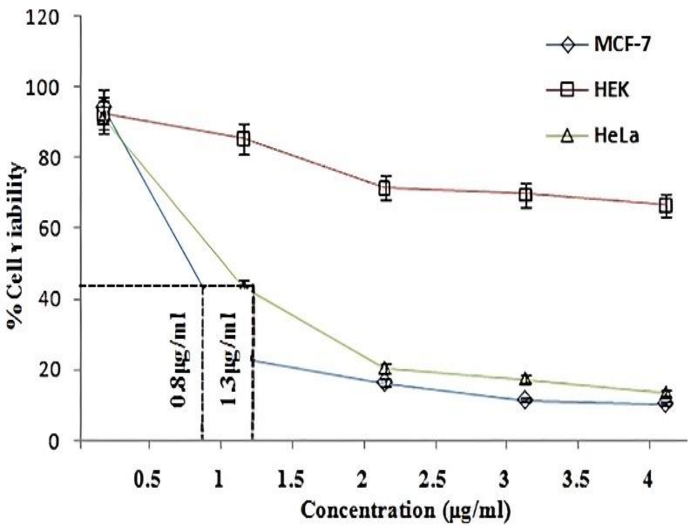

Fig 12. The cytotoxicity of the MTT reduction assay indicates the IC50 values for HeLa cells and MCF-7, while demonstrating an insignificant cytotoxic effect on HEK cells exposed to camptothecin-loaded gold nanoparticles (Clarance et al. 2020).

A study that focus on the synthesis of silver nanoparticles of endophytic fungi *Botryodiplodia theobromae* isolated from *Psidium guajava*, a group of scientists start to study its anticancer and antitumor abilities, MCF-7 and A549 breast and lung cancer cell lines had been cultured in DMEM supplemented with fetal calf serum with the percentage of 10 percent and 1 percent of antibiotic solution containing streptomycin and penicillin at 37 °C in 5% CO_2 until the monolayer reached sub-confluence. The cells were individually plated in 96-well plates at a concentration of 1×10^5 cells per well, washed twice with 100 µL of serum-free medium after 24 hours, and deprived of nutrients at 37°C for one hour. Following starvation, cells were administered a distinct test compound for a duration of 24 hours. Subsequent to treatment, serum-free medium with 0.5 mg per mL of MTT had been introduced and incubated at 37°C for 4 hours and the medium had been aspirated. The containing medium of MTT had been removed, in addition to that the cells had been rinsed with 200 Microliter of PBS. The crystals were solubilized in DMSO and thoroughly mixed. cytotoxicity was assessed with GraphPad Prism 5 Software, and the absorbance of the purple-blue formazan dye was quantified using a spectrophotometer at 570 nm in a microplate reader (Janakiraman et al. 2019).

Conclusion

There is no doubt that nanotechnology these days are one of the most important branch of biotechnology specifically the green synthesis of nanoparticles, we discuss in this review The eco-friendly synthesis of nanoparticles via microorganisms, especially fungi, and how it is rapid, economical, and effective, eliminating the need for toxic chemicals. We discuss also that Nanoparticles are highly effective for drug delivery because their diminutive size enables facile penetration of cell membranes. This property is utilized to address ailments such as cancer, which are associated with various adverse effects from chemotherapy. They also exhibit significant cytotoxic effects on MCF-7 and HeLa cells, indicating a potential alternative to traditional chemical methods in medical biotechnology. Nanoparticles also inhibited biofilm formation, downregulated biofilm-associated genes, and diminished liver and spleen tissue congestion and fibrosis, positioning them as promising nano-candidates for future pharmaceutical and biological applications, thereby highlighting their potential for effective utilization.

Conflict of interest

The authors declare that they have no conflict of interest.

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