__Review Article__

MANAGEMENTOFPLANTDISEASESWITHGREENSYNTHESIZEDNANOPARTICLESUSINGPLANTEXTRACTS

Abstract

Nanoparticles (NPs) can typically be synthesized using two different techniques. The first method, the top-down approach, involves physical processes like sonication, laser ablation, radiation, and thermal decomposition.The distinctive features of nanoparticles, such as large surface area to volume ratio, surface plasmon resonance (SPR), and their unique biological, optical, and electrical properties, have significantly increased their demand. Furthermore, the production of nanoparticles has garnered substantial popularity among scientists and researchers worldwide in recent years.The green synthesis of nanoparticles is often referred to as biosynthesis, involving a range of biological sources. In green nanoparticle synthesis, microorganisms or their derivatives, as well as plant extracts, are used instead of synthetic chemicals, with minimal impact on human health and the environment.There is a lack of comprehensive knowledge regarding the extended or prolonged effects of GSNPs on plants and the environment. This knowledge gap hinders our understanding of how GSNPs may impact crops and ecosystems over extended periods. Research efforts should address these questions to ensure the safe and effective utilization of GSNPs in agriculture and environmental applications.

Keywords: Nanoparticles, sonication, synthetic chemicals, environmental applications

INTRODUCTION

Agriculture faces numerous hurdles, including inefficient use of resources, expensive capitalequipment, threats from pests and environmental factors, and a declining interest among theyounger generation. In this context, the adoption of cutting-edge technologies plays a crucialrole in overcoming these agricultural challenges and boosting crop yields. Nanotechnology isa rapidly advancing field that has effectively tackled various issues across multiple industries,includingagriculture(BegumandJayawardana,2023).Nanotechnologyinvolvesworkingwithnanomaterials, the particles typically having sizes ranging from 1 to 100 nanometers (Hossain*et al.*, 2019). Prof. Norio Taniguchi coined the term nanotechnology, and Richard Feynman isknownasthefatherofnanotechnology.Thedistinctivefeaturesofnanoparticles,suchaslargesurface area to volume ratio, surface plasmon resonance (SPR), and their unique biological,optical, and electrical properties, have significantly increased their demand. Furthermore, theproductionofnanoparticleshasgarneredsubstantialpopularityamongscientistsandresearchers worldwideinrecentyears(Jadoun*etal.*,2021).Nanotechnologyhasfoundwidespread applications

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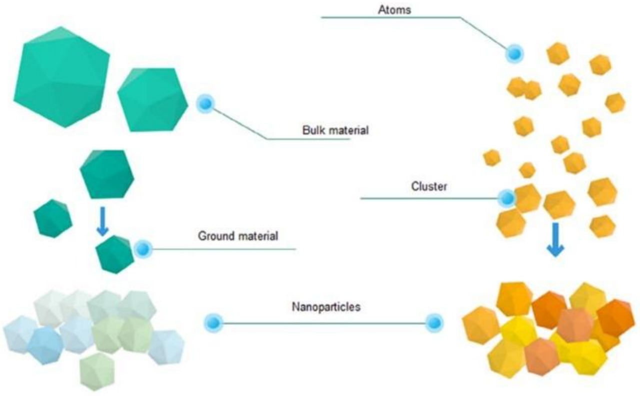
in everyday life and is progressively becoming more significant insociety. It is being utilized in various fields, including medicine, pharmaceuticals, electronics,energy, environmental sciences, chemistry, food industries, and, more recently, agriculture(HernandezDiaz*etal*.,2021).Inagriculture,nanotechnologyplaysaroleintheproductionofnanoscale fertilizers, pesticides, and herbicides (Elemike*et al*., 2019). Nanoparticles are usedinplant diseasemanagement aswell.

SYNTHESISOFNANOPARTICLES



Top-downapproach

Bottom-upapproach

Fig.1-Techniquesofnanoparticlesynthesis

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Nanoparticles (NPs) can typically be synthesized using two different techniques. The firstmethod, the top-down approach, involves physical processes like sonication, laser ablation,radiation,andthermaldecomposition.Inthisapproach,NPsaregeneratedbyreducingthesizeof a larger material, resulting in agglomerates with nano sized particles. Drawbacks of thismethod are, producing a variety of particle sizes (polydispersity), introducing imperfections(includingcontaminationfromtheinitialmaterial),requiringsignificantenergyandspecializedlaboratory equipment, and being costly (Pirtarighat*et al.*, 2019)**.** The second approach, thebottom-upmethod,involvesbuildingnanoscalestructuresfromatomicandmolecularcomponents. NPs are formed through chemical and biological synthesis. Chemical synthesistechniquesincludeelectrochemistry,vaporfluxcondensation,thesol-gelmethod,andchemicalreduction.Amongthese,chemicalreduction,whichutilizeschemicalslikesodiumborohydrideand sodium citrate (Wuithschick*et al*., 2015), is one of the most commonly used methods forNP generation. Nonetheless, chemical methods often involve multiple chemical species ormolecules, which can increase particle reactivity and toxicity. They may also have adverseeffects on human health (Fu *et al*., 2014) and the environment due to the decomposition ofchemicalgroups,thegenerationofby-products,andhighenergydemand(RahimiandDoostmohammadi, 2020).

Differentmethodsofnanoparticlesynthesis

Nanoparticles can be synthesized using various methods, generally categorized into physical,chemical,andbiologicalmethods.Thesemethodsemploydifferentsourcesasreducingagentsorelectrondonorsforthesynthesisprocess.Someofthemethodsarelisted below.

Methodsofnanoparticlesynthesis(Table1)

Methodsofnanoparticlesynthesis		
Physicalmethods	**Chemicalmethods**	**Biologicalmethods**
• Gasphasedeposition • Electronbeam lithography • Powderballmilling • Pulsedlaserablation • Aerosol	• Coprecipitation • Sonochemical • Thermaldecomposition • Microemulsion • Hydrothermal • Electrochemical deposition	• Plants • Fungi • Algae • Bacteria • Biopolymers

(Jadoun*etal*.,2021)

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Even though all these methods have been used for nanoparticle synthesis, the widespreadapplicationofphysicalandchemicalapproachesinagricultureandhealthcarehasbeenrelatively limited (Li *et al*., 1999). The challenges linked to the synthesis of nanoparticles byphysicalandchemicalmethodsstemfromseveral factors.Theseincludetheuseofpotentiallyhazardouschemicals,theneedforcostlyequipmentandmachinery,therequirementforlargerlaboratory spaces, demanding processing conditions like high temperature and pressure, andsignificant energy consumption (Ahmad and Jaffri, 2019; Hossain *et al*., 2019; Some *et al.*,2019). Moreover, the time required for synthesis, the generation of harmful by-products, theoverall high costs involved, and the adverse environmental effects further compound thedifficultiesassociated withnanoparticleproduction(Kausar*etal*., 2022).

GREENSYNTHESISOFNANOPARTICLES

The green synthesis of nanoparticles is often referred to as biosynthesis, involving a range ofbiologicalsources.Ingreennanoparticlesynthesis,microorganismsortheirderivatives,aswellasplantextracts,areusedinsteadofsyntheticchemicals,withminimalimpactonhumanhealthandtheenvironment.Utilizinggreennanotechnologiescanbeapotentapproachtoaddresstheintricate scientific and technological hurdles in enhancing the safety of the entire agriculturalandfoodproductionchain(Choudhary*etal*.,2021).Dr.KatteshV.Kattiisknownasthefatherof green nanotechnology. Green nanomaterials offer significant potential in developing nano-based pesticide formulations due to their small size, large surface area, and properties that canbe tailored for specific targets. This has the potential to enhance the effectiveness, safety, andeconomic impact of conventional pesticides by prolonging their duration of action, reducingthe necessary dosage, enabling the controlled release of active ingredients, ensuring stability,andminimizing runoffand environmentalresidues(Choudhary*et al*.,2021).

Thegreensynthesisofnanoparticlesinvolvesthreekeycomponents:reducingagents,solvents,andcappingagents.Biomolecules,whichcanbenaturallyoccurringorproducedbyplantsandmicrobes, such as polyphenols, flavonoids, terpenoids, tannins, alkaloids, polysaccharides,proteins, amino acids, and vitamins, serve as reducing agents during nanoparticle synthesis(Hossain *et al*., 2019). These biomolecules reduce metal ions to a zero-valence state, and thefunctionalgroupswithintheseprimarybiopolymersandphytochemicalsplayaroleinstabilizing the resulting nanoparticles. Among various biological methods, utilizing plants fornanoparticlesynthesisispreferredduetotheirwidespreadavailability,safety,lackoftoxicity,

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andthepresenceofawiderangeofphytoconstituentsthatcanactasreducingagents(Uddin *etal*.,2021).

Thegreensynthesisofnanoparticlesisregardedasasafeapproachfornanoparticleproductionand has numerous advantages compared to physical and chemical methods. This method isenvironmentally friendly, cost-efficient, provides high yields, steady, involves a simple andeasy synthesis process, relies on renewable and biocompatible materials, avoids the use ofdangerous toxic chemicals, allows for easy accessibility and easy handling and is an energy-saving process, among other benefits (Uddin *et al*., 2021; El-Nagger *et al*., 2017). However, afewdrawbacksareassociatedwithit,suchasthepotentialecologicalimbalancecausedbytheoverexploitation of natural biological sources and the seasonal variation in the availability ofphytochemicals(Altaf*etal*., 2021).

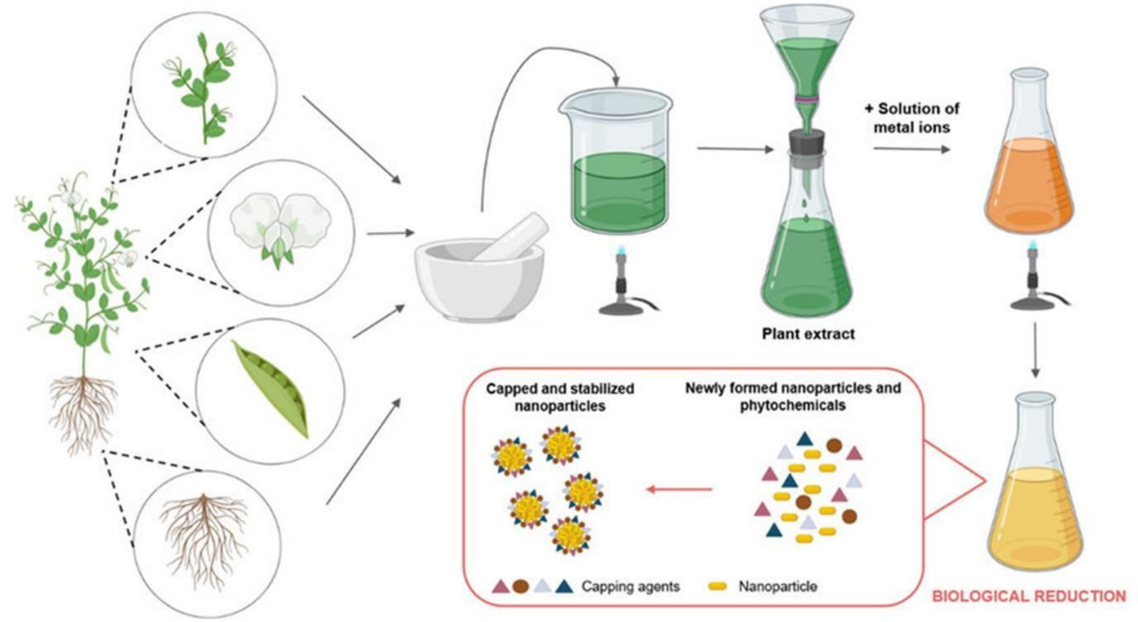

Fig.2-Generalstepsofgreensynthesisofnanoparticles

Differentpartsofplants,includingfruits,leaves,stems,androots,havebeenwidelyemployedfor the green synthesis of nanoparticles because of the valuable phytochemicals they contain(Iravani,2011).Innanoparticlesynthesisusingplantmaterials, thespecificpartoftheplanttobeusediscleanedandboiledwithdistilledwater.Afterward,theresulting mixtureissqueezed,filtered, and combined with the desired solutions for nanoparticle synthesis. As the solutioncolourchanges,indicatingtheformationofnanoparticles,theycanbeseparatedandcollected.Thisnaturalplantextract-basedsynthesisisenvironmentallyfriendlyandcost-

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effective, eliminating the need for intermediate chemical reagents. Nanoparticle synthesis occurs in a

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three-step sequence: (i) the reduction of metal ions, often signalled by observable colourchanges, (ii) the clustering of nanoparticles; and (iii) the stabilization of these nanoparticles(Some*et al*., 2019).

FACTORSAFFECTINGGREENSYNTHESISOFNANOPARTICLES

SolutionpH

ThepHlevelofasolutionplaysacriticalroleinsynthesizingnanoparticlesfromplantsources(GerickeandPinches,2006).IthasbeenemphasizedthatthepHofthesolutioncansignificantlyaffectthetimeittakesforsynthesis,aswellasthesizeandshapeoftheresultingnanoparticles (Vijayaraghavan and Ashokkumar, 2017). The formation of nucleation centersduring nanoparticle synthesis is highly dependent on the pH level. An increase in pH can leadtotheformationofmorenucleationcenters,whichinturnacceleratesthereductionofmetallicions into metal nanoparticles. The time it takes to reduce metal salts is closely associated withthepHofthereactionmediumbecausepHaffectstheinteractionbetweenthefunctionalgroupspresent in the plant extract and the metal ions (Bali and Harris, 2010). Scientific research hasdemonstrated that the production of smaller nanoparticles is more likely to occur in a basicsolutioncompared to anacidicone (Dubey *et al.*,2010).

Reactiontime

Theincubationandreactionperiodlengthplaysacrucialroleindeterminingthecharacteristics,morphology, and yield of nanoparticles (Kuchibatla*et al*., 2012). Changes in the incubationtimeandstorageconditionsalsoimpactthepropertiesofthenanoparticlesproduced(Mudunkotuwa*et al.*, 2012). Extending the incubation period can result in aggregation anddecrease the potential for reducing nanoparticles during the synthesis process (Baer, 2011).Certain research findings have indicated that effective nanoparticle synthesis tends to occurwithlonger reaction times (Darroudi*et al.*,2011).

Temperature

Temperature is another influential factor in the synthesis of nanoparticles, and it has a similarimpact on their morphological properties as pH does. Temperature also affects the formationofnucleationcenters,withlowertemperaturesleadingtoareducedformationofthesecenters,subsequently slowing down the synthesis rate (Pham *et al*., 2019). Because of the specificsecondarymetabolitespresentinplantextracts,ithasbeenrecommendedtocarryoutnanoparticlesynthesisatroom temperature topreventthe degradationandalterationof

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functional groups(Jemilugba*et al.*, 2019). Nevertheless, research has demonstrated thattriangular-shaped nanoparticles are formed at lower temperatures, whereas spherical-shapednanoparticles are generated at higher temperatures (Raju *et al.*, 2011). Studies have reportedthat smaller volumes of plant extract are needed for stable nanoparticle synthesis at highertemperatures,andlarger-sizednanoparticlestendtobeproducedathighertemperatures(Iravaniand Zolfaghari,2013).

Effectofplantextractconcentration

The concentration of plant extract plays a crucial role in synthesizing metal nanoparticles as itprovides the electrons needed for reducing metal ions. A reduction in the amount of plantextractresultsinareducedformationofnanoparticles(Kiruba*etal.*,2013).Ontheotherhand,employing a larger volume of plant extract in metal nanoparticle synthesis leads to a higherquantity of phytochemicals, accelerating the reduction of metal salt. Nevertheless, the fasterthisreductionoccurs,thesmallerthesizeoftheresultingmetalnanoparticles(DinandRehan,2017).

Concentrationandnatureofmetalsalt

The choice of metal salt employed in the synthesis significantly impacts the characteristics,structure, and size of the nanoparticles produced. For instance, copper salts such as copperchloride,coppersulphate,copperacetate,andcoppernitratearecommonlyusedfornanoparticle synthesis. Research findings have indicated that when copper chloride salt isutilized,triangularandtetrahedral-shapedcoppernanoparticles(Cu-NPs)areformed,whereasrod-shaped Cu-NPs are obtained with copper acetate salt (Shankar and Rhim, 2014). Whencopper sulphate salt is used, spherical Cu-NPs are synthesized (Shah *et al.*, 2015). It has alsobeenreportedthatanincreaseintheconcentration ofmetalsaltleadstoanincreaseinthesizeofthenanoparticles (Din*et al.*, 2017).

Pressure

Numerous research investigations have demonstrated that pressure plays a role in influencingthe morphological characteristics of nanoparticles synthesized from plant sources (Akintelu*etal.*, 2021).

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CHARACTERIZATIONOFNANOPARTICLES

Metalnanoparticlesarecharacterizedforseveralpurposes,includingtrackingthecompletenessofreduction,identifyingthefunctionalgroupsinvolvedinthebio-reductionprocess,assessingpuritylevels,andanalyzingtheirmorphologicalattributes.Thecommonlyemployedtechniquesfor thesepurposesarehighlighted below;

UV–visiblespectrophotometry

Theinteractionbetweenplantextractandmetalsaltresultsinobservablecolourchangesinthesolution due to the excitation of surface plasmon vibrations in the NPs. The absorption bandcorrespondingtoeachmetalcanbeconfirmedbyanalyzingthesolutionwithaUVspectrophotometer.UV-visiblespectrophotometrytracksthecharacteristicpeaksgeneratedbymetal salt-derived nanoparticles (NPs) at various absorption wavelengths during the synthesisprocess. For instance, in the case of Cu-NPs, a distinctive absorption occurs in the range of520-600 nm within the visible region due to surface plasmon resonance (SPR). UV-visiblespectroscopy is also employed to estimate the aggregation state, size, and size distribution ofNPs(Lotha*et al.*, 2019).

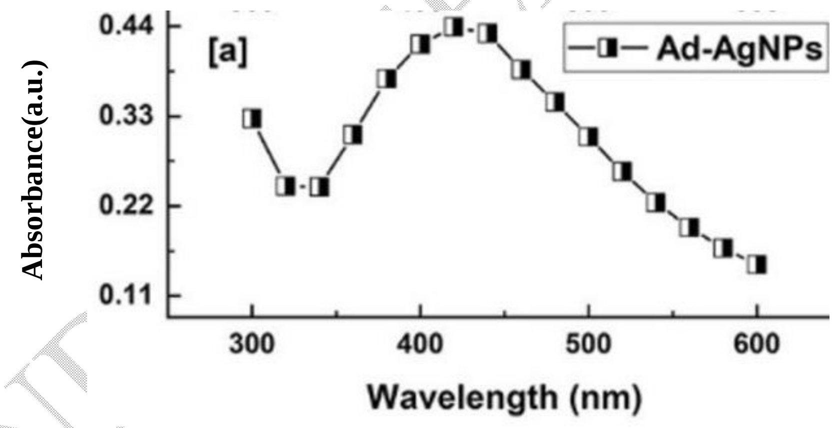

Fig.3-UVvisibleabsorptionspectraofsilvernanoparticlesgreensynthesisedfrom *Amaranthusdubius*

Fouriertransforminfrared(FTIR)spectroscopy

FTIR spectroscopy can be used to detect functional groups capable of donating electrons forthe reduction of metal salts (Sebeia*et al.*, 2019). FTIR spectrophotometer measures thewavelengthoflightagainstinfraredintensity.ResearcherstypicallycomparetheFTIRspectra

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oftheplantextractandthesynthesizednanoparticlestodeterminewhichfunctionalgroupsareresponsiblefor reducingthemetal ions(Akintelu*etal*., 2020).

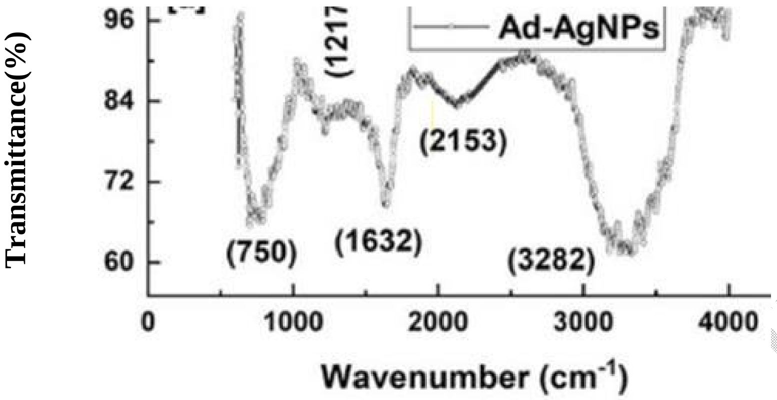

Fig.4-FTIRspectraofsilvernanoparticlesgreensynthesisedfrom*Amaranthusdubius*

X-raydiffraction(XRD)

The X-ray diffraction (XRD) technique is utilized to gather structural data regarding thecrystallinenatureofnanoparticles(Sharma*etal*.,2019).DuringXRDanalysis,thehigh-energyX-ray rays emitted by the machine penetrate deeply into the nanoparticles, yielding valuableinsights into their structure (Jamdade*et al*., 2019). The formation of nanoparticles in thenanoscalerangeistypicallycharacterizedby broadening thepeaksobservedinXRDanalysis.

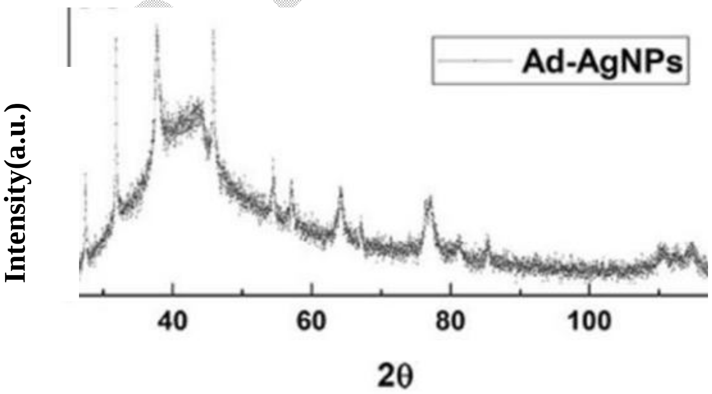

Fig.5-X-raydiffractionanalyticalpatternofsilvernanoparticlesgreensynthesisedfrom *Amaranthusdubius*

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Scanningelectronmicroscopy(SEM)

The morphological characteristics of nanoparticles are assessed through the use of scanningelectron microscopy (SEM). Additionally, SEM analysis can be employed to estimate theaverage size of nanoparticles with the assistance of certain statistical software tools (Akintelu*etal*., 2021).

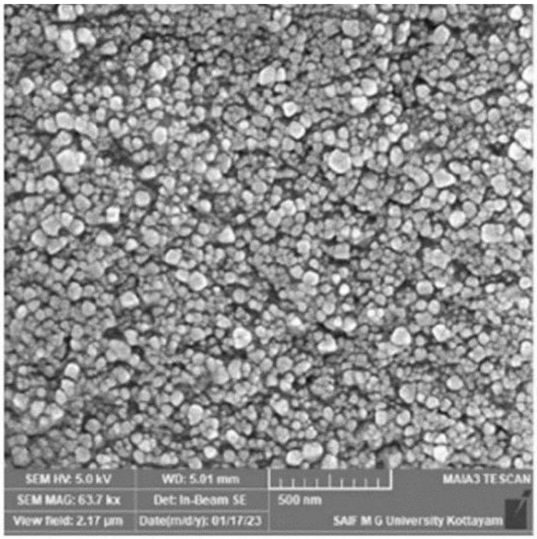

Fig.6-FESEMimageofsilvernanoparticlesgreensynthesisedfrom*Amaranthusdubius*

Transmissionelectronmicroscopy(TEM)

TEMprovideshighermagnification,superiorresolution,andmorepreciseinformationregarding shape, crystallinity, and size compared to SEM (Akintelu*et al*., 2021). Additionally,TEM is particularly advantageous due to its ability to distinguish between crystalline andamorphous structures using selected area electron diffraction techniques, which makes it evenmorebeneficial for nanoparticle characterization(Caroling*et al*., 2015).

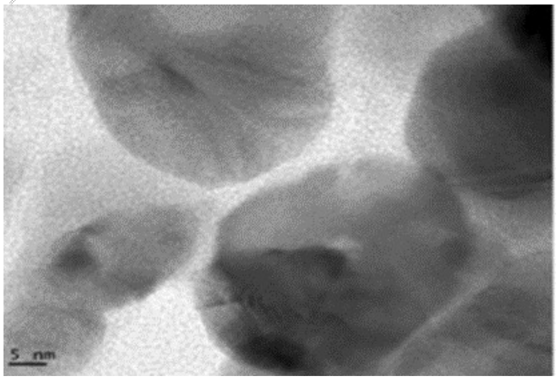

Fig.7-TEMimageofsilvernanoparticlesgreensynthesisedfrom*Amaranthusdubius*

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Atomicforcemicroscopy(AFM)

Atomicforcemicroscopy(AFM)isusedformorphologicalassessmentofnanoparticles(Chidanandappa and Nargund, 2020).

EnergydispersiveX-rayspectroscopy(EDX)

EDX spectroscopy is widely accepted as a suitable method for determining the elementalcompositionofnanoparticles.Thisisachievedbyexaminingthedistinctgroupsofpeaksint heX-ray spectrum produced by the unique atomic structure of each element, facilitating theidentificationoftheseelements (Noruzi, 2015).

MECHANISMOFACTIONOFNANOPARTICLESAGAINSTPHYTOPATHOGENS

Green-synthesized nanoparticles (GSNPs) have primarily been studied for their effectivenessincombattingphytopathogens.However,ourunderstandingofhowthesenanoparticle sinhibitor kill microorganisms remains limited. The mechanism of action of nanoparticles can bebroadly categorized as disruption of the peptidoglycan layer in bacterial cell walls, toxicityresulting from the release of toxic metal ions into the cytoplasm leading to imbalances innutrient uptake, impairment of membrane function including membrane damage and the lossof membrane potential, generation of reactive oxygen species (ROS) and the production ofantioxidants, damage to genetic material such as double-helix strand breaks, dysfunction ofproteinsetc.

Apart from the impact of metal ions, different metabolites found in plant extracts have beenobserved to induce cell death in pathogens and stimulate systemic resistance in plants (Mishra*et al.*, 2017). Alkaloids, phenolics, and natural compounds present in plant extracts have beenshowntopossessbactericidalandfungicidalpropertiesagainstplantpathogens,therebyenhanc ingthe efficacyof green-synthesizednanoparticles (Choudhary *et al.*,2021).

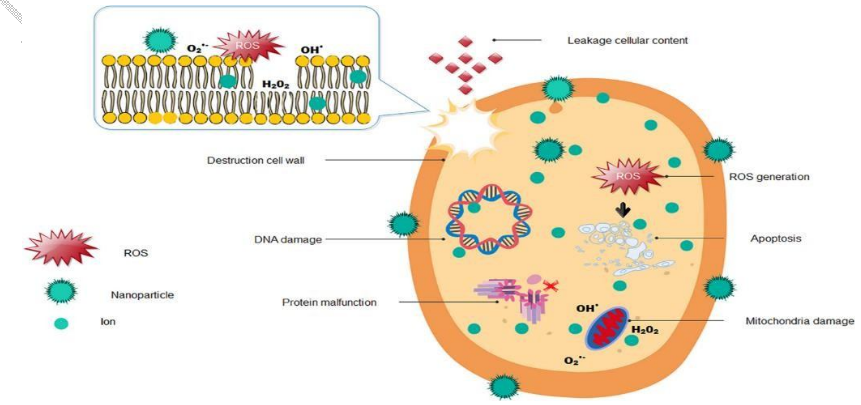

Fig.8-Mechanismofactionofnanoparticlesagainstphytopathogens

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MECHANISM OFACTIONOFDIFFERENTMETALNANOPARTICLESAGAINST PHYTOPATHOGENS(Table 2)

Nanoparticle type	**Ionic form**	**Mechanismofaction**	**Phytopathogen type**
AgNPs	Ag^+	• Releaseofionsthataretoxictowardspathogens • Increase the permeability of thebacterialmembrane • Disruptionofthebacterialmembrane • Damage to the cell components (lipids,DNAand proteins) • Inhibitionofenzymeactivity • InhibitionofDNAreplication	Bacteria and fungi
Al_2O_3NPs	Al^{3+}	• Releaseofionsthataretoxictowardspathogens • IncreaseROSproduction • Depolarizationofcellmembranes	Bacteria
AuNPs	Au^{3+}	• Disruption of bacterial membrane andalterationofmetabolism • Damagetocellorganelles(cellwallandmitochondria) • InhibitionofDNAuncoilingandtranscriptionmediatedbybindingofAuNPsto bacterial DNA	Bacteria
CeO_2NPs	Ce^{3+}, Ce^{4+}	• Inhibitionofiontransportthroughpumps • Induction of oxidative degradation oflipidsand/orproteinsofthepathogen's plasmamembrane • Impairmentofelectronfluxandbacterialrespiration • Inhibitionoffungalenzymeactivity	Gram-positivebacteria and fungi

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CdONPs	Cd^{2+}	• Releaseofionsthataretoxicagainstpathogens • Induction of oxidative stress onbacterialcells • IncreaseROSproduction • Interrupted transmembrane electrontransportand mitochondriadamage	Bacteria
CuNPs/CuONPs	Cu^{2+}	• Inhibitionofenzymeactivityessentialtothemicroorganisms • IncreaseROSproduction • DamagetoessentialmoleculessuchasDNA	Bacteria and fungi
SeNPs	Se^{6+}, Se^{4+}	• IntracellularATPdepletion • InductionofoxidativestressthroughROS production • Alteration of bacterial membranepotential • Disruptionofbacterialmembrane • Inhibitionoffungalsporegermination	Bacteria and fungi
SiNPs/SiO_2NPs	Si^{4+}	• Inductionofmechanicaldamagetobacterialmembrane • IncreasedROSproduction • Inductionofoxidativestress	Bacteria and fungi
TiO_2NPs	Ti^{4+}	• IncreaseinROSproduction • Inductionofphotocatalyticdamage	Bacteria and fungi
ZnONPs	Zn^{2+}	• Release of ions that are toxic towardspathogens • IncreasedROSproduction • Disruptionofmitochondrialfunction • Inductionofchangesincellmorphologyandreleaseofcellcomponents	Bacteria

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Fe_2O_3NPs	Fe^{3+}, Fe^{2+}	• Induction of oxidativestress throughROSproduction • Destructionofcellmembranes,inducing changes in cell morphologyandreleaseof cell components • Damage to essential molecules such asproteinsand DNA • Releaseofironionsleadingtooxidativedamageby Fentonreaction	Bacteria and fungi

(Hernandez Diaz*etal.*, 2021)

ANTIMICROBIAL AND DISEASE-MANAGING PROPERTIES OFNANOPARTICLESGREENSYNTHESIZEDFROMPLANTEXTRACTS

Greensynthesizednanoparticlesarereportedtobeeffectiveagainstseveralplantpathogenicfungi,bacteriaand viruses.

GREENSYNTHESIZEDNANOPARTICLESAGAINST PHYTOPATHOGENICFUNGI (Table3)

Plant	Plant part used	NP	Pathogen	Effective concn.	Experimental approach	Reference
Oryzasativa	Leaf	Ag	*Rhizoctonia solani*	-	-	Kora*etal.*, 2020
Solanum tuberosum	Leaf	Ag	*Alternaria alternata, R. solani,Botrytis cinerea,Fusariumoxysporum*	22.8 µg/mL	*Invitro*	Almadiy andNenaah,2018
Abelmoschus esculentus	Seed	Au	*Pucciniagraministritici, Aspergillus flavus,Aspergillusniger,Candidaalbicans*	-	-	Jayaseelan *etal.*, 2013

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Citrussinensis	Fruit	Cu	*Colletotrichum capsici*	-	*Invitro*	Divte*et al.*,2019
Aloevera	Leaf	Se	*Colletotrichum coccodes, Penicillium digitatum*	-	*Invitro*	Fardsadegh andJafarizadeh - Malmiri, 2019
Curcuma longa	Roots	TiO_2	*Fusariumgraminearum*	0.2–20 mg/mL	*Invitro*	Abdul *etal.*,2016
Ecliptaalba	Leaf	ZnO	*Sclerospora graminicola*	-	Field	Nandhini*et al.*, 2019
Punicagranatum	Peels	ZnO	*Aspergillus niger*	50 µg/mL	*Invitro*	Mishraand Sharma,2015

Silver nanoparticles green synthesized with leaf extract of disease-resistant amaranthusgenotypes effectively suppress leaf blight (*Rhizoctonia solani*) disease in a susceptible redamaranthuscultivar(Divya *et al*., 2023)

Silver nanoparticles (AgNPs) were produced through green synthesis using leaf extracts fromdifferenttypesofamaranthusplants:theleafblightdisease-susceptibleredamaranthus(*Amaranthus tricolor* L.), the disease-resistant green amaranthus (*A. dubius*), and the wildamaranthus (*A. viridis*) genotypes. These nanoparticles were then characterized using varioustechniquesandevaluatedfortheirantifungaleffectsagainst thepathogen *Rhizoctoniasolani*.

The green-synthesized nanostructures exhibited absorption characteristics typical of silvernanoparticles in spectroscopy and demonstrated a face-centered cubic structure in X-raydiffractionanalysis.FieldEmissionScanningElectronMicroscopyandHigh-ResolutionTransmission Electron Microscopy revealed that the nanoparticles had a size range of 35–45nmacross all samples.

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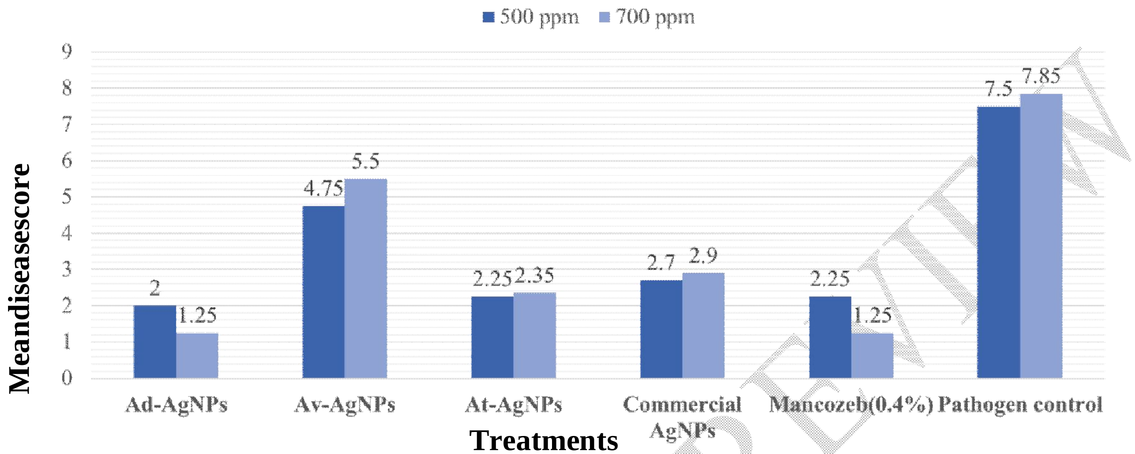
In vitro experiments using a poisoned-food assay showed that the nanoparticles inhibited themycelialgrowthofthepathogenwhenappliedatconcentrationsof500and750ppm.Moreover,detachedleavesfromtheredamaranthusvarietywerespraedwiththenanoparticles and then exposed to the pathogen. It was observed that leaves treated with Ad-AgNPs and Av-AgNPs had significantly fewer lesions compared to those treated with At-AgNPs.



Graph1:LeafblightincidenceinA.*tricolorvar.*Aruninthedetachedleafassay

Mycelial growth inhibition of the amaranthus leaf blight pathogen *Rhizoctonia solani*inpoisoned-foodassay (Table4)

Treatments	**Percentagediseaseindex**			
	500 ppm		**750 ppm**	
	3DAI	**5DAI**	**3DAI**	**5DAI**
Ad-AgNPs	1.64±1.42^{d}	16.04±1.23^{e}	1.64±0.71^{c}	7.40±2.13^{e}
Av-AgNPs	4.11±0.71^{d}	24.28±1.88^{d}	2.46±0.00^{c}	17.69±1.88^{d}
At-AgNPs	16.45±3.10^{b}	51.44±1.42^{b}	8.77±0.23^{b}	49.38±2.13^{b}
CommercialAgNPs	10.29±0.72^{c}	30.04±1.42^{c}	8.23±0.71^{b}	27.57±1.88^{c}
Mancozeb(0.4%)	4.93±2.47^{d}	10.69±1.42^{f}	1.23±1.23^{c}	6.99±2.56^{e}
PathogenControl	39.91± 377^{a}	84.36±5.70^{a}	11.93±1.42^{a}	86.83±1.42^{a}
AbsoluteControl	0.00	0.00	0.00	0.00

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Inan*invivo*assay,wheresusceptibleredamaranthusplantswerechallengedwiththepathogenafter foliage spraying with AgNPs at a concentration of 750 ppm, the lowest disease index(7.40)wasrecordedforAd-AgNPs,followedbyAv-AgNPs(17.69)fivedaysafterinoculation.Incontrast,At-AgNPs-treated plants had adiseaseindexof49.38.

These findings suggest that the application of AgNPs, green-synthesized using leaf extractsfrom disease-resistant amaranthus genotypes, effectively reduced the incidence of leaf blightdiseasein a susceptibleamaranthus variety.

GREENSYNTHESIZEDNANOPARTICLESAGAINST PHYTOPATHOGENICBACTERIA

(Table5)

Plant	Plant part used	NP	Pathogen	Effective concn.	Experimental approach	Reference
Pipernigrum	Stem	Ag	*Citrobacter freundii* *Erwiniac acticida*	-	*Invitro*	Paulkumar*et al.*, 2014
Azadirachta indica	Leaf	Ag	*Xanthomonas oryzae* pv.*oryzae*	-	*Invitro*	Mankad*et al.*, 2020
Phyllanthus emblica	Fruit	Ag	*Acidovorax oryzae*	30 µg/mL	*Invitro*	Masum*etal.*, 2019
Cymbopoga citratus	Leaf	Al_2O_3	*Pseudomonas aeruginosa*	2000 µg/mL	*Invitro*	Ansari*etal.*, 2015
Gloriosa superba	Leaf	CeO_2	*Pseudomonas aeruginosa*	100 µg/mL	*Invitro*	Arumugum*et al.*, 2015
Carica papaya	Leaf	CuO	*Ralstoniasola nacearum*	250 µg/mL	*In vitro* &greenhouse	Chen*etal.*, 2019
Rosmarinus officinalis	Flower	MgO	*X.oryzae*pv.*oryzae*	-	*Invitro*	Abdallah*et al.*, 2019
Withania somnifera	Leaf	Se	*Bacillus subtilis*	25 µg/mL	*Invitro*	AlagesanandVenugopal,2019
Cynodon dactylon	Leaf	Si	*Pseudomonas aeruginosa*	60 µg/mL	*Invitro*	Babu*etal.*, 2018

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Trigonella foenum-graecum	Leaf	TiO_2	*Bacillus subtilis*	10 mg/mL	*Invitro*	SubhapriyaandGomathipriya , 2018
Solanum lycopersicum	Fruit	ZnO	*X.oryzae*pv. *oryzae*	-	*Invitro*	Ogunyemi*et al.*, 2019

Phytofabricationofsilvernanoparticlesusingthreeflowerextractsandtheirantibacterialactivitiesagainstpathogen*Ralstoniasolanacearum*strainYY06ofbacterialwilt(Cheng*et al.*, 2020)

Bacterial wilt, caused by the phytopathogen *Ralstonia solanacearum*, is a significant globalproblem. The conventional use of bactericides and antibiotics to control bacterial wilt hasshownlimitedeffectivenessandposedenvironmentalrisks.Thisstudyaimedtoproducesilvernanoparticles (AgNPs) through a natural process using extracts from the flowers of canna lily(*Cannaindica*L.),Cosmos(*Cosmosbipinnata*Cav.),andLantana(*Lantanacamara*L.)plants.

The synthesized AgNPs were validated and characterized using various techniques, includingUV-visible spectroscopy, Fourier transform infrared spectroscopy (FTIR), X-ray diffraction(XRD), transmission electron microscopy (TEM), and scanning electron microscopy (SEM).UV-visiblespectrarevealeddistinctiveabsorptionpeakbandsat448nm,440nm,and428nmfor AgNPs synthesized using *C. indica* L., *C. bipinnata*Cav., and *L. camara* L. flowers,respectively.FTIRspectraconfirmedtheinvolvementofbiofunctionalgroupsfromtheflowerextractsintheAgNPsynthesisprocess,acting ascappingandstabilizing agents.

ThesphericalAgNPssynthesizedfromthethreedifferentflowersourceshadaveragediametersof 43.1 nm, 36.1 nm, and 24.5 nm for *C. indica* L., *C. bipinnata*Cav., and *L. camara* L.,respectively. The AgNPs produced by *L. camara* L. flowers at a concentration of 10.0 µg/mlexhibited the highest suppression zone of 18 mm against the *R. solanacearum* strain YY06,surpassingtheefficacyofAgNPssynthesizedby *C.indica*L.and*C.bipinnata*Cav.flowers.

Furthermore, the AgNPs negatively impacted various aspects of *R. solanacearum*, includingbacterialgrowth,biofilmformation,swimmingmotility,nucleicacidefflux,celldeath,cell

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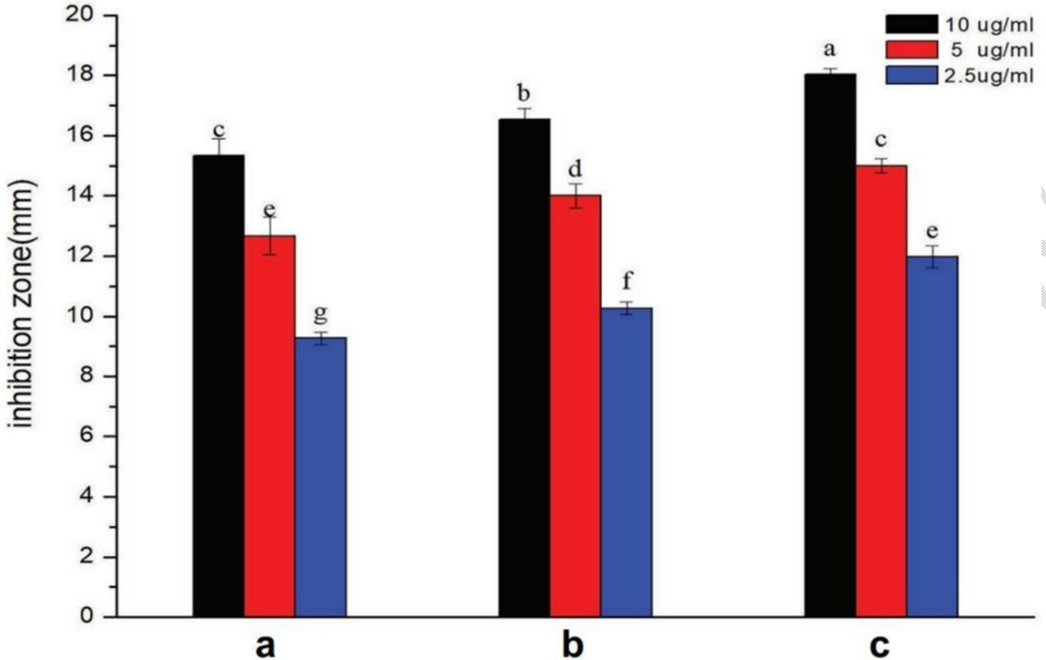
membrane damage, and the generation of reactive oxygen species (ROS), mainly when theywerepresent inhigh concentrations andhadsmallparticlesizes.



Graph 2: Diameter of bacterial growth inhibition (mm) caused by AgNPs which weresynthesized by(a) *C. indica* L. flower; (b) *C. bipinnata*Cav. flower; and (c) *L. camara* L.flower

Insummary,thebiosynthesized AgNPshavethepotentialtoserveasaneffectiveandenvironmentallyfriendlyantibacterialagentfor reasonablyinhibiting*R.solanacearum*.

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GREEN SYNTHESIZED NANOPARTICLES HAVING BOTH ANTIFUNGAL ANDANTIBACTERIALPROPERTIES(Table6)

Plant	Plant partused	NP	Pathogen	Effective concn.	Experimental approach	Reference
Azadirachta indica	Leaf	Ag	*R.solanacearum, Aspergillus*sp. *Fusarium*sp.	-	*Invitro*	Haroon*et al.*, 2019
Leucaena leucocephala	Leaf	CdO	*P.aeruginosa, Aspergillusniger*	500 µg/mL	*Invitro*	Savale*etal.*, 2017
Ocimum sanctum	Leaf	Cu	*Alternaria carthami,A.niger, Colletotrichumgloeosporioides, Colletotrichum lindemuthianum, Drechslerasorghicola, F. oxysporum,Macrophominaphaseolina, Rhizoctonia bataticola, R. solani,Xanthomonas axonopodis*pv.*citri, X.axonopodis*pv. *punicae*	10–60 µg/mL	*Invitro*	Shende*et al.*, 2016

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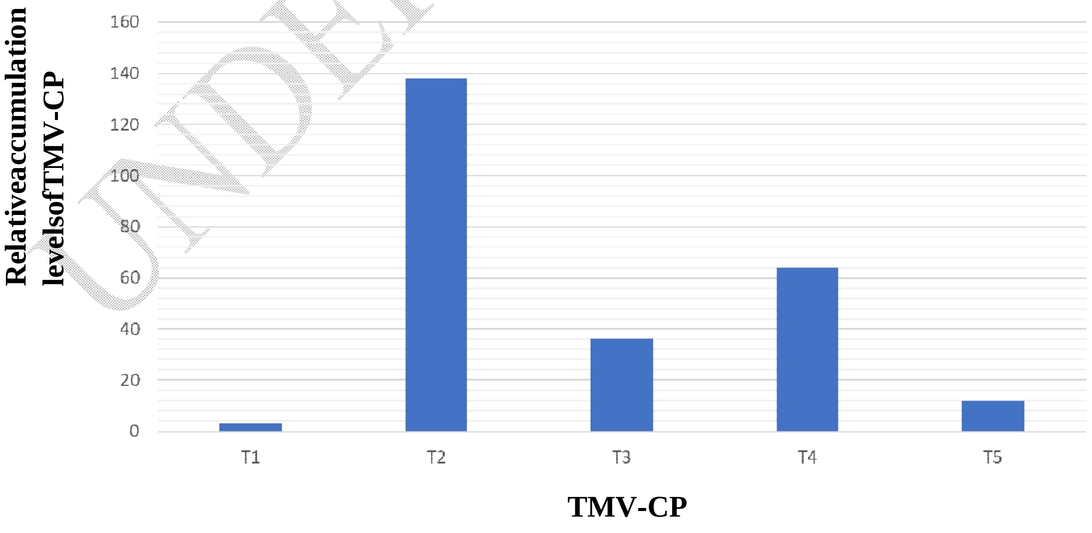
GREENSYNTHESIZEDNANOPARTICLESAGAINST PHYTOPATHOGENICVIRUSES

Green synthesized ZnO nanoparticles mediated by *Mentha spicata* extract induce plantsystemicresistanceagainst*TobaccoMosaicVirus*(AbdelkhalekandAl-Askar,2020)

Globally, plant viral infections pose a significant threat to food security, leading to substantialcropproductionlosses.Nanoparticleshaveproventobeeffectiveagentsforcontrollingvariousplantpathogens.However,ourunderstandingoftheireffectsagainstviralinfectionsislimited.In this study, they achieved the green synthesis of zinc oxide nanoparticles (ZnO NPs) usinganaqueous leaf extract of *Mentha spicata.*

X-ray diffraction patterns confirmed that the prepared ZnO NPs had a crystalline structure.Analyses using dynamic light scattering and scanning electron microscopy showed that theresulting ZnO NPs were spherical in shape, with particle sizes ranging from 11 to 88 nm.Fourier transmission infrared spectroscopy identified different functional groups, capping andstabilityagents and revealedthepresenceof Zn-Obonds at awavenumberof 487cm^{-1}.

Under greenhouse conditions, we evaluated the antiviral activity of biologically synthesizedZnO NPs (at a concentration of 100 µg/mL) against the *Tobacco mosaic virus* (TMV). Themost effective treatment involved double foliar application of the prepared ZnO NPs 24 hoursbeforeand24hoursafterTMVinoculation,resultingina90.21%reductioninviralaccumulation and disease severity. Additionally, the transcriptional levels of genes associatedwithplantdefensemechanismswereinduced andup-regulatedin allZnONPs-treatedplants.



Graph 3:Relative expression level of TMV-CP gene in TMV-infected tomato plants at21 days after inoculation (T1 = Mock-treated plants (Control), T2 = plants inoculated with TMVonly, T3 = plants treated with ZnO NPs (24 h before TMV inoculation), T4 = plants treated with ZnONPs

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(24 h after TMV inoculation) and T5= plant treated with ZnO NPs (24 h before TMV inoculationand24 hafterTMVinoculation)

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Significantly, results demonstrated that the aqueous extract of *Mentha spicata* served as aneffective reducing agent for the green synthesis of ZnO NPs, which exhibited substantialantiviral activity. Ultimately, the protective and curative effects of ZnO NPs against TMVsuggesttheir potentialapplication inmanagingplant viral diseases.

CHALLENGES

Theuseofbiologicalsourcesfornanoparticlesynthesismayimposeconstraintsonthepossibilityoflarge-scalecommercialproductionofnanoparticles.Thislimitationarisesbecause the availability and prevalence of the specific plants or biological materials used canfluctuate depending on the season and geographic location. Furthermore, it is crucial toconsidertheoptimalgrowthstageoftheseplantswhenutilizingthemfornanoparticlesynthesis,asthisfactorsignificantlyinfluencesthe qualityandyieldofthe nanoparticles.

Green-synthesized nanoparticles (GSNPs) can exhibit a wide range of sizes, which can posechallenges in achieving uniformity in their properties for various applications. Additionally,GSNPsareoftenlessstableandpronetooxidation,whichcanimpacttheirlong-termeffectivenessand shelf life(Begum and Jayawardana,2023).

One significant drawback is that many experiments investigating nanoparticles' effects haveprimarilyfocusedonshort-termcropsorimmediateimpacts.Thislimitedscopemeansthatwemay not have a comprehensive understanding of the cumulative and long-term effects ofnanoparticles on plants, the environment, and even human health. As nanoparticles becomemorewidelyusedinagricultureandotherfields,itbecomesincreasinglyimportanttoconduct researchthataddressestheirpotentiallong-termimpactsandcumulativetoxicity.Thiswillhelpto ensure the safe and sustainable application of nanoparticles in various sectors (Choudhary*etal*., 2021).

FUTUREPROSPECTS

It is essential to conduct field trials with various crops and diseases to evaluate the efficacy ofallsynthesizednanoparticlescomparedtocommercialpesticidesandbiocontrolagents.Research should explore the use of less phytotoxic metals like Cu, Zn, Mn, Fe, and Mg aspotential alternatives to costly silver nanoparticles (AgNPs). A thorough understanding of thestructural properties of green nanoparticles, including morphology, size, functional groups,loadingcapacity,andtheirimpactonplants,shouldbeestablished.Amoreefficient,rapid,and scalableprotocolforformulatinggreennanoparticlesisnecessaryforsuccessfullarge-scale

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production. The effects of nanoparticles on soil, wildlife, plant biodiversity, crop yields, andfarmer income should be investigated. The toxicity and effectiveness of green-synthesizednanoparticles should be compared to chemically synthesized nanoparticles to assess theirsuitabilityforvarious applications.

CONCLUSION

Green-synthesized nanoparticles (GSNPs) are known for their ease of production and cost-effectiveness.Theyofferseveraladvantages,includingtheirabilitytoinducesystemicresistancetodiseases,andexhibitfungicidalandbactericidalproperties.However,despitetheirpromising attributes, questions remain about their efficacy and long-term sustainability whenused in field conditions. There is a lack of comprehensive knowledge regarding the extendedorprolongedeffectsofGSNPsonplantsandtheenvironment.Thisknowledgegaphindersourunderstanding of how GSNPs may impact crops and ecosystems over extended periods.Research efforts should address these questions to ensure the safe and effective utilization ofGSNPsin agricultureand environmental applications.

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