

Review Article

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Nanotechnology Unleashed: Revolutionizing Legume Farming for Enhanced Biotic Stress Management



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ABSTRACT

Biotic stresses pose a significant challenge to global agriculture, severely affecting crop productivity, quality and food security. In the present day, nanotechnology has emerged as a promising and innovative approach to mitigating the detrimental effects of biotic stresses on crops, offering novel strategies for enhancing plant resilience and protection. The utilization of nanomaterials, including nanoparticles and nanocomposites, has shown substantial potential in enhancing plant resistance against various biotic stressors, for instance, pathogens, pests, and weeds. Key advancements in this field include antimicrobial nanoparticles, precision target delivery systems for bio-pesticides, and nanoscale sensors that enable early detection of plant diseases. Moreover, nanotechnology grants exclusive opportunities to improve the efficiency of conventional agricultural practices while reducing environmental impact and fostering sustainable farming. However, challenges in this context include limited field level validation, variability in nanomaterial behavior under diverse agro-climatic conditions and a lack of long term impact assessment. Despite these limitations, this work contributes by consolidating recent innovations, identifying key areas of application and emphasizing the integration of nanotechnology into precision agriculture. Nevertheless, contempt its auspicious aptitude, an array of encounters and ethical apprehensions must be addressed for its accountable application in agriculture. Main issues encompass nanoparticle toxicity, ecological significance, regulatory agendas, and public insight, all of which necessitate thorough appraisal to warrant the nonviolent and sustainable amalgamation of nanotechnological progressions. In the era of precision agriculture, nanotechnology arises as a transformative invention with enormous capability to redefine biotic stress management and pointedly advance the goal of sustainable global food production.

Keywords: Biotic stress; Crop protection; Early disease detection; Nanocomposites; Nanoparticles; Nanotechnology; Nanoscale; Precision agriculture; Sensors

Introduction

Since the Nobel laureate Richard P. Feynman presented the concept of nanotechnology in his seminal 1959 lecture, *There's plenty of room at the bottom* (Feynman, 1960), the field has witnessed amazing progress and revolutionary advancements [1]. Norio Taniguchi was the proponent of the term "Nanotechnology" [2]. Nanotechnology, derived from the Greek word *Nano*, meaning dwarf, refers to a scale reduction by a factor of 10^{-9} , making it a thousand times smaller than a micron. This nanoscale measurement, undistinguishable to the naked human eye [3], comprehends structures and materials at the atomic and molecular levels, creating the foundation of nanotechnology. In recent years, nanomaterials have expanded substantially in status across multiple fields, including medicine, cosmetics, communication, electronics, energy production, agriculture, food processing, and textiles [4].

These materials are demarcated as assemblies, aggregates, or agglomerates with at least one external dimension measuring less than 100 nanometers [5] or as substances possessing a volume precise surface area (VSSA) greater than $60 \text{ m}^2 \text{ cm}^{-3}$ [6]. A key distinguishing feature of nanomaterials is their exceptionally high surface area to volume ratio, which plays a crucial role in determining their bio-stimulant potential [7]. This asset also contributes to substantial modifications in their physicochemical actions equated to bulk materials [1,8].

Nanotechnology involves the study and manipulation of nanomaterials, which are defined by their dimensions ranging from 1 to 100 nanometers [2,4]. This definition encompasses a broad spectrum of both naturally occurring and engineered materials classified as nanoparticles. Naturally occurring particles are found in different environmental forms, including volcanic ash and oceanic salt aerosols [9,10] (Fig.1). The nanoscale sizes of nanoparticles, united with their high surface area to volume ratio, enhance their reactivity, enabling efficient binding, absorption and transport of different compounds, including small molecule drugs, probes, DNA, proteins and RNA [11]. Beyond their enhanced surface area, nanoparticles exhibit unique physicochemical properties that distinguish them from their bulk counterparts (Fig. 2).

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DOI: <https://doi.org/10.21276/AATCCReview.2025.13.03.153>

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For example, gold (Au), which is traditionally inert and exhibits a golden color in bulk form, becomes reactive and appears red at the nanoscale. Similarly, titanium dioxide (TiO_2) and zinc oxide (ZnO), typically white in bulk form, become transparent when reduced to the nanoscale. Moreover, nanoparticles often display lower melting points and enhanced reactivity compared to their larger counterparts. These distinct properties have been extensively leveraged in nanotechnology, leading to advancements across numerous industries and the development of a vast array of innovative products and applications [12]. Additionally, nanomaterials can be generally categorized according to their shape and composition. They are classified as either inorganic or organic nanoparticles based on their composition (Fig.3). These are also categorized structurally as anisotropic as displaying direction-dependent properties, and isotropic, having uniform properties in all directions [13].

Nanotechnology remains in its nascent stages, offering significant potential to transform agricultural practices (Fig. 4). Its advancements have attracted microbiologists and researchers, encouraging their contributions to food safety through innovative strategies based on green chemistry principles [14]. In 2021, food insecurity impacted approximately 1.2 billion people worldwide. Nevertheless, its effects are not uniformly distributed across populations, and climate-related shock poses a significant risk of further aggravating food insecurity and its associated health implications [15]. Agricultural productivity endures to be embarrassed by an array of biotic and abiotic factors, including weeds, diseases and insect pests significantly reducing potential crop yields (Fig. 5). Empirical evidence indicates that pest infestations account for yield losses of approximately 25% of rice, 5-10% in wheat, 30% in pulses, 35% in oilseeds, 20% in sugarcane and 50% in cotton [16, 17].



Figure 1: Different examples of nanostructures present in nature

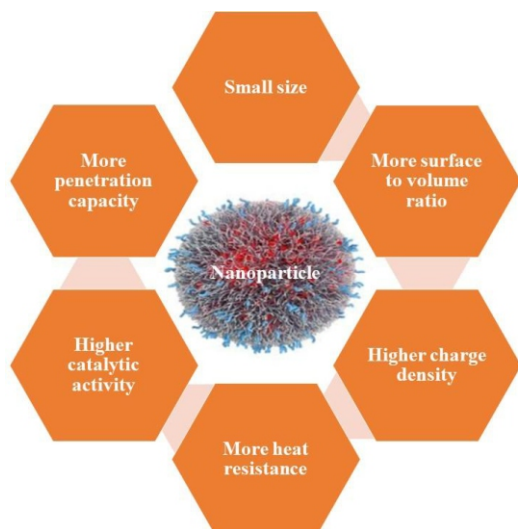


Figure 2: Properties of nanoparticles

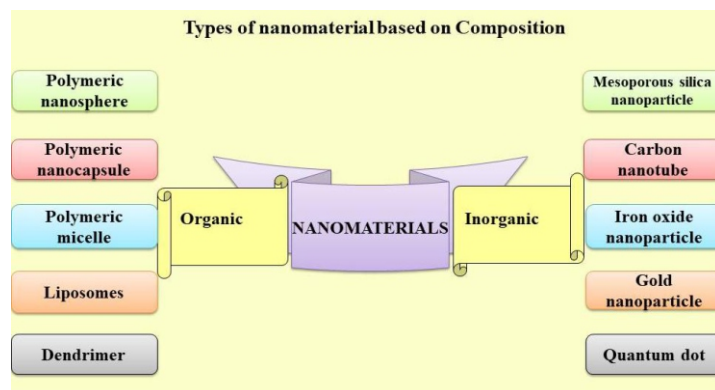


Figure 3: Types of nanomaterial based on composition



Figure 4: Application of nanotechnology in agriculture

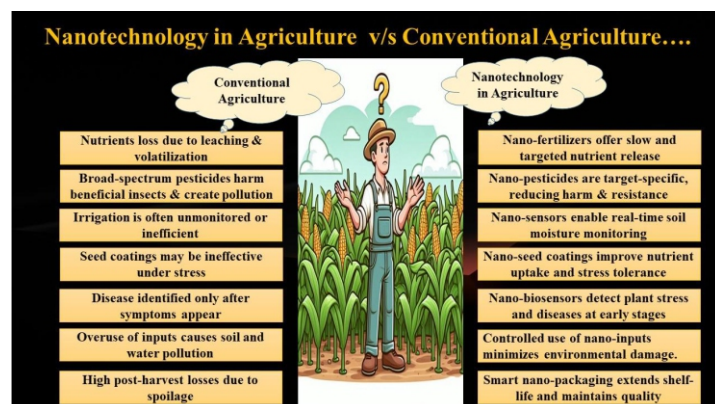


Figure 5: Comparison of nanotechnology in agriculture versus conventional agriculture

Legumes serve as crucial components of both human and animal diets, while also playing a fundamental role in cropping systems. Their contributions significantly enhance soil health, promote sustainable agriculture, and support global environmental sustainability [18,19,20,21]. Legumes, belonging to the expansive plant family Fabaceae, which ranks as the third largest, encompass a vast array of over 20,000 species cultivated across diverse agro-climatic zones and soil types [22]. These leguminaceous crops play pivotal roles in enhancing soil fertility through nitrogen fixation, serving as a beneficial disease deterrent for cereal and oilseed crops, and serving as staple foods [23,24,25]. They contribute significantly to dietary nutrition by providing substantial amounts of dietary fibers and minerals, essential amino acids, carbohydrates, and proteins. Furthermore, certain legumes hold considerable economic value as high-quality commercial export value food crops, collectively constituting 27% of the global crop

production [26]. Grain legumes face vulnerability to a multitude of pathogens encompassing bacteria, fungi, viruses, nematodes, and parasitic plants, leading to substantial economic repercussions on a global scale [27]. Addressing the pressing need to meet the demand for grain legumes underlines the importance of emphasizing the adoption of innovative breeding and agronomic technological interventions for their genetic improvement. Additionally, the implementation of suitable policies and action plans is crucial to promote the cultivation of these crops [18].

2. Nanotechnology in biotic stress management- overview

Biotic stress, caused by pathogens, pests and weeds, significantly reduces global agricultural productivity. Traditional approaches of pest and disease management, together with chemical pesticides and biological control agents, often face limitations such as environmental toxicity, resistance development, and reduced efficacy under changing climatic conditions. Nanotechnology presents a promising alternative,

offering novel solutions for efficient, targeted and sustainable biotic stress management by its different formulations and nanoparticles (Table 1, Fig. 6).

Nanomaterials, including metal nanoparticles, nano-formulated pesticides and nanocarriers for biocontrol agents, enhance plant defense mechanisms while minimizing ecological impact. Silver, copper, zinc oxide and other nanoparticles have demonstrated antimicrobial properties, effectively suppressing bacterial, fungal growth and bio-availability of biopesticides, ensuring controlled and prolonged activity against pathogens and pests.

Beyond direct antimicrobial action, nanotechnology plays a crucial role in plant immunity enhancement. Engineered nanomaterials can induce systemic resistance in plants by modulating stress-responsive pathways, activating defense enzymes, and influencing hormonal signaling. Furthermore, nano-structured coatings and smart nanosensors enable early detection and precise management of biotic stress, ensuring timely interventions and reducing yield losses.

Table 1: Different types of nanoparticles used in agriculture

Category	Examples	Uses in agriculture	References
Nanoparticles as fertilizers (Nanofertilizers)	Nano-urea, nano phosphorus, nano potassium, nano zinc	Enhances nutrient uptake, reduces fertilizer loss, improves soil health	[20,28]
Nanoparticles for pest control (Nanopesticides)	Silver, copper and titanium dioxide nanoparticles	Effective contrary to pest and pathogens, reduces chemical pesticide use, minimizes environmental impact	[29]
Nanoparticles for disease management (Nano fungicides & Nano bactericides)	Chitosan nanoparticles, zinc oxide nanoparticles, silver nanoparticles	Controls fungal and bacterial diseases, promotes plant defense mechanisms	[30]
Nanocarriers for slow release of agrochemicals	Silica nanoparticles, polymeric nanoparticles, liposomes	Provide controlled release of pesticides, fertilizers and herbicides, reduces dosage and toxicity	[31]
Nanosensors for precision agriculture	Carbon nanotubes, quantum dots, Gold nanoparticles	Detects soil moisture, nutrient levels, pathogens and environmental conditions in real time	[32]
Nano enabled soil remediation agents	Iron oxide nanoparticles, Biochar nanoparticles, Graphene oxide	Improves soil fertility, removes heavy metals and contaminants, enhances microbial activity	[33,34]
Nano based growth enhancers	Carbon based nanomaterials, silica nanoparticles	Stimulates seed germination, enhances root and shoot development, improves stress tolerance	[35,36]
Nanomaterials for post-harvest management	Silver nanoparticles, zinc oxide nanoparticles, nano emulsion	Extends shelf life prevents microbial spoilage, maintains quality during storage	[37]

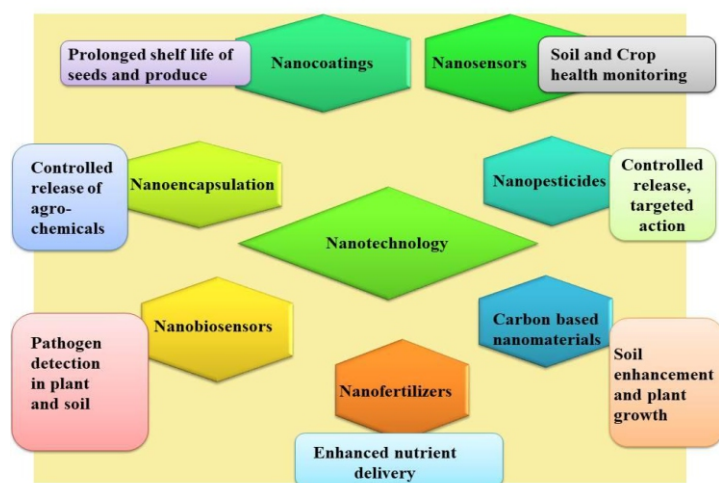


Figure 6: Different formulations of nanomaterials in nanotechnology used in agriculture

2.1 Nanotechnology for disease management

The relevance of nanotechnology in the realms of plant disease management, diagnostic approaches, and genetic transformations is nascent, representing an area that has only recently commenced exploration within the plant pathology literature [12]. Phytopathogens are projected to incur a substantial annual crop loss, estimated to range between 20 to 30% [38]. This phenomenon is recognized as a formidable challenge to global food security, particularly in light of the escalating human population, as acknowledged by the United Nations in 2015 [39].

In addressing the formidable challenge posed by plant diseases, both traditional breeding and chemical interventions have proven insufficient [40,41]. The indiscriminate application of synthetic pesticides to manage plant biotic stresses has deleterious ramifications on human and animal health, the environment, and exacerbates the emergence of resistance in numerous pathogens and pests [42]. Consequently, the search for more effective and environmentally safe agents to combat this danger to worldwide food safety has become imperative.

Nano-agrochemicals have recently emerged as a compelling avenue for enhancing crop yield and global food security, offering distinct recompenses over traditional products and methods. These advantages are intricately linked to heightened efficiency, abridged effort requirements, and lesser ecological toxicity [43,44,45]. Diverse nanomaterials have demonstrated multifaceted utility, including acting as antimicrobial agents capable of directly impeding pathogenic virulence. Nanoparticles of metals, for instance silver (Ag NPs), copper (Cu/Cu₂O NPs), and zinc (ZnO/ nanocopper composite) have exhibited potent antibacterial or antifungal capabilities against pathogens such as *Fusarium oxysporum*, *Phytophthora infestans* and *Xanthomonas perforans* [46,47,48,49]. Moreover, nanomaterials job as elicitors, stimulating plant innate immunity to bolster resistance against biotic stresses. For instance, Ag NPs, Ag-silica hybrid complexes, and other metal NPs have been recognized as persuaders of plant immunity, leading to enhanced production of phenolic compounds, oxidative enzymes and up-regulation of systemic acquired resistance (SAR) marker genes [50,51,52].

Furthermore, nanomaterials can act as carriers for active ingredients without directly contributing to the action. They operate as delivery systems for pesticides, micronutrients, and elicitors [39,53,54]. Investigations have verified the potential of nanomaterials, including Fe₂O₃ NPs, TiO₂ NPs, and carbon-based NPs at suitable concentrations to suppress pathogen infections and enhance plant growth, as evidenced in the model system of tobacco (*Nicotiana benthamiana*) infected with Turnip mosaic virus (TuMV) [55]. Therefore, the detection of operative nanomaterials and the exploration of specific application concentrations are crucial. Given the substantial potential of nano-agrochemicals in plant disease management, it is of utmost importance to rigorously assess their disease suppressive effects and establish safe and operative application approaches for different nanomaterials [56].

2.2. Nanotechnology for insect pest management

Insects, constituting over two-thirds of the known animal species worldwide, inhabit diverse environments. They exhibit a broad spectrum of feeding habits, targeting various plants, as well as crop plants, forest trees, medicinal plants and weeds. Additionally, insects pose threats to stored products such as food, leading to substantial losses and quality deterioration in storage facilities like godowns, bins and packages. In their pursuit of sustenance, insects can directly or indirectly harm plants and stored goods [17]. Traditional pest control methods involve the widespread application of commercially available pesticides in significant quantities, resulting in an additional financial burden on crop production. The excessive use of pesticides not only contaminates water sources but also adversely impacts the surrounding environment. There is a critical necessity to minimize the use of pesticides to safeguard the environment and reduce costs in crop production [57]. A viable approach to achieving this objective is to explore non-toxic substances that can effectively combat pests [58].

2.3 Nanotechnology for weed management

Weeds pose significant threats to agriculture, competing for essential nutrients meant for food crops and thereby depriving the food crop of its rightful share. The conventional approach to address this issue involves their eradication. Nanotechnology presents a promising solution for weed control through the usage of nano-herbicides in an environmentally friendly way, leaving negligible toxic residues in the soil or the surrounding environment [59]. The Nano-herbicides, with their nano-sized particles, offer several advantages over conventional herbicides. Their small size facilitates thorough mixing with soil particles and effective elimination of all above-ground weeds. By incorporating specific herbicides with nanoparticles, the nano-herbicides target specific receptors in weed roots. Upon penetration into weed roots, they inhibit the glycolysis pathway, leading to a shortage of energy-rich ATP molecules and ultimately causing the demise of weeds in a single application [58].

In recent years, nanotechnology has appeared as a transformative strength in agriculture, exhibiting advanced solutions to improve crop protection and promote sustainable farming practices. Nanoparticle-based antimicrobial agents, encapsulated pesticides and nano biosensors have become key players in the quest for efficient, eco-friendly and targeted approaches to manage plant diseases, pests and weeds. This revolutionary shift towards nanotechnology in agriculture is driven by the unique properties exhibited by nanomaterials,

which allow for precise control, increased efficacy and reduced environmental impact.

1. Nanopesticides based on antimicrobial agents

Nanomaterials (NMs) find various applications in plant protection, with a primary focus on their role as antimicrobial agents to enhance disease management and promote plant health. Amongst the widely investigated NMs with antimicrobial properties are metal nanoparticles (NPs), including Zinc (Zn), Copper (Cu), and Silver (Ag). Furthermore, carbon and polymer-based nanomaterials have also been studied for their potential as antimicrobial agents in the context of plant protection (Table 2). In the realm of nanoparticle synthesis, various fungi endowed with robust biocontrol capabilities are harnessed and notably, *Trichoderma harzianum* stands out as a widely employed species with documented applications in biopesticides and biofertilizers [60]. The facile growth requirements of *T. harzianum* have significantly expanded its utility within the realms of biotechnology and nanotechnology [61]. The *Trichoderma* genus, to which *T. harzianum* belongs, has been recognized for its possession of NADH-dependent enzymes and NADH co-enzymes, particularly nitrate reductase, playing a pivotal role in the synthesis and capping of nanoparticles [62]. Zinc oxide nanoparticles (ZnO NPs) emerge as noteworthy entities in this context, having been documented for their utility as antimicrobial agents, particularly in impeding the growth of diverse fungi and bacteria [63]. *Trichoderma harzianum* was employed in nanotechnology to synthesize zinc oxide nanoparticles (ZnO NPs) for controlling Fusarium wilt in chickpea. The characterized ZnO NPs, produced with *T. harzianum*, displayed substantial inhibition of *Fusarium oxysporum* *in vitro*. In green house experiments, priming chickpea seeds with these nanoparticles occasioned a 90% reduction in disease incidence. The ZnO NP primed seeds enhanced antioxidant activity, leading to increased resistance against wilt. The study suggests that *Trichoderma harzianum* assisted ZnO NPs could serve as an environment-friendly and effective nano-fungicide for managing Fusarium wilt in chickpea, with the potential for practical field application [64]. Similarly, Kumari et al. (2017) [48] documented the elevated bactericidal activity of silver nanoparticles (Ag NPs) derived from *Trichoderma viridae* in comparison to chemically synthesized Ag NPs. This heightened activity was ascribed to the surface coating provided by secondary antimicrobial metabolites originating from *T. viridae* [39]. Hashem et al. (2021) [65] conducted a study wherein they bio-synthesized selenium nanoparticles (SeNPs) utilizing the culture supernatant of *Bacillus megaterium* ATCC 55000. The resultant green Se NPs exhibited notable antifungal efficacy against *Rhizoctonia solani* both *in vitro* and *in vivo*. This compelling finding suggests the potential utilization of these Se NPs as an effective agent for the management of *R. solani* diseases in faba bean cultivation. Elkhodary et al. (2023) [66] employed *Streptomyces gancidicus* for the biofabrication of biometallic ZnO B₂O₃ nanoparticles (NPs), alongside monometallic ZnO NPs and B₂O₃ NPs. The synthesized bimetallic ZnO-B₂O₃ NPs showed remarkable antifungal effectiveness against *Phytophthora irregularis*, a causative agent of damping off in plants. Treatment with ZnO-B₂O₃ NPs resulted in a statistically significant reduction in the percent disease incidence (PDI) by 7.5%, accompanied by a remarkable increase in protection by 91.1%. Moreover, ZnO-B₂O₃ NPs induced a substantial augmentation in total soluble protein levels in *P. irregularis*-infected pea plants,

underlining their potential as efficacious biological alternatives for the management of *P. irregulare* in agricultural settings and reducing yield losses in pea. Abdelkhalek et al. (2023) [67] utilized the *Rhizobium leguminosarum* bv. Viciae strain 33504-Borg 1, recognized for its nitrogen-fixing capabilities, was used for the biofabrication of silver nanoparticles (AgNPs) aimed at mitigating bean yellow mosaic virus (BYMV) disease in faba bean. Results from green house trials demonstrated that foliar application of AgNPs on faba bean leaves, performed 24 hours prior to BYMV inoculation, elicited plant resistance, leading to reduced disease severity and diminished virus accumulation. Furthermore, this treatment exhibited positive effects on plant health, as evidenced by enhanced growth parameters, elevated levels of peroxidase (POX) and polyphenol oxidase (PPO) enzymes and decreased concentrations of oxidative stress markers, for instance, hydrogen peroxide (H₂O₂) and malondialdehyde (MDA).

Table 2: Nanoparticle based antimicrobial agents for different pathogens

Nanoparticle	Antimicrobial agent	Pathogen	Crop	References
Zinc oxide nanoparticles (ZnO NPs)	<i>Trichoderma harzianum</i>	<i>Fusarium oxysporum</i>	Chickpea (<i>Cicer arietinum</i>)	[64]
Silver nanoparticles (Ag NPs)	<i>Trichoderma viride</i>	bactericidal activity	-	[48]
Selenium nanoparticles (Se-NPs)	<i>Bacillus megaterium</i>	<i>Rhizoctonia solani</i>	Faba bean (<i>Vicia faba</i>)	[65]
ZnO-B ₂ O ₃	<i>Streptomyces gancidicus</i>	<i>Phytophthora irregulare</i>	Pea (<i>Pisum sativum</i>)	[66]
ZnO NPs	<i>Bacillus thuringiensis</i>	<i>Callosobruchus maculatus</i>	Black gram (<i>Vigna mungo</i>), Cowpea (<i>Vigna unguiculata</i>)	[68,69]
Pumilacidin (Lipopeptide)	<i>Bacillus pumilus</i>	<i>Pythium aphanidermatum</i> , <i>Rhizoctonia solani</i> and <i>Sclerotium rolfsii</i>	Soybean (<i>Glycine max</i>)	[69]
Silver nanoparticles (AgNPs)	<i>Rhizobium leguminosarum</i> bv. viciae	Bean yellow mosaic virus (BYMV)	Faba bean (<i>Vicia faba</i>)	[67]

2. Nanoparticle-encapsulated pesticides

Biological biocides exhibit considerable promise for mitigating plant biotic stresses and promoting developmental vigor, however, their intrinsic instability and rapid degradation pose challenges. Biological control agents (BCAs) represent an environmentally friendly strategy that effectively inhibits plant diseases and enhances crop productivity. In response to the escalating demand for BCAs, research on encapsulation has witnessed a notable upswing in recent decades. Encapsulation, delineated as the process by which an active core is physically incorporated into a matrix structure and subsequently stabilized through chemical means, stands as a pivotal method in overcoming the inherent limitations of free-form formulations. These encapsulation formulations offer a viable avenue for ameliorating the challenges associated with biological biocide applications. Notably, they enhance the efficacy of BCAs by extending shelf-life and facilitating the controlled release of bio-active components. Adopting an innovative approach, encapsulation emerges as a promising platform for the regulation of biotic stressors, particularly plant pathogens. Among the key polymers integral to this encapsulation paradigm, chitosan and alginate stand out, exhibiting significant potential in confining and preserving BCAs. This nuanced strategy not only addresses the logistical constraints of biological biocide utilization but also emphasizes its capacity as a dynamic device for sustainable plant pathogen management [42]. The encapsulation process is characterized as a technique that involves enclosing substances within an inert material in miniature dimensions at the nanoscale [70].

In an investigation, chitosan thiamine nanoparticles (CNPs) were employed to induce defensive responses against *Fusarium oxysporum* f. sp. ciceris (Foc) in chickpeas [8]. Foc, a prominent soil-borne pathogen, is known to cause severe yield losses, reaching up to 100% [71]. The utilization of CNPs resulted in notable enhancements in both non-enzymatic and enzymatic antioxidants, accompanied by increased lignin deposition within the vascular bundles of chickpea stem tissues, compared to the control [8,72].

Chitosan nanoparticles (CNPs) have emerged as pivotal contributors to abating diverse stress circumstances in plants, orchestrating a spectrum of defense retorts that encompass the

induction of pathogen-related proteins, phytoalexins, proteinase inhibitors, antioxidants, callose deposition, lignin deposition and other relevant mechanisms. This multifaceted role positions CNPs as an environmentally friendly alternative to chemical agents, offering a sustainable strategy for preventing seed infestations by many pests. The findings emphasize the potential of chitosan thiamine nanoparticles in bolstering the resilience of chickpeas against the detrimental impact of *Fusarium oxysporum* f. sp. ciceris, providing insights into an ecologically sound approach for agricultural pest management [71]. Moreover, [47] observed that the utilization of biosynthesized silver nanoparticles (Ag NPs) at a concentration of 100 µg ml⁻¹ resulted in a substantial reduction of chickpea *Fusarium* wilt incidence by 73.33%. This efficacy surpassed that of the commercial fungicide control (CuOCl), which exhibited a reduction of 26.67%, indicating an enhanced effectiveness of 46% [39].

Abdallah et al. (2022) [73] conducted an investigation aimed at elucidating the influence of varying concentrations of MgO nanoparticles (MgONPs) on mung bean plants and evaluating the commonness of *Fusarium solani* (*F. solani*) and *Fusarium oxysporum* (*F. oxysporum*) under *in vivo* conditions. Moreover, the study sought to delineate the consequential effects on soil health. *In vitro* investigations revealed that MgONPs exhibited inhibitory properties against fungal growth. Transitioning to green house conditions, the strictness of disease induced by *F. solani* experienced a notable reduction, decreasing from approximately 44% to 25%. Similarly, the impact of *F. oxysporum* diminished, with disease severity decreasing from 39% to 11.4%.

In current centuries, there has been a growing utilization of various nanomaterials for the progress of revised release systems of herbicides. This trend accentuates the necessity to evaluate the competence of these nanocarriers in weed control and their potential residual effects on susceptible crops. Newly, the investigation conducted by [74] highlighted the significance of nanotechnology in herbicide application, specifically focusing on atrazine-loaded polycaprolactone (PCL) nanocapsules for weed control in soybean cultivation. Nanoencapsulation of atrazine progresses pre-emergence herbicidal activity against *Bidens pilosa* (*B. pilosa*), a common weed, without exacerbating

long-term residual effects on soybean plants. The findings suggested that, when respecting recommended intervals, this environmentally friendly nanoatrazine formulation has the potential to efficiently control weeds, reduce environmental contamination, and minimize herbicide content in the soil. The use of PCL, a safe and biodegradable polymer, further supports the eco-friendly nature of this nanotechnology application in soybean cultivation [74].

In the current market, chitosan combined with either tripolyphosphate or glutaraldehyde nanoparticles is readily available for the encapsulation of the biopesticide PONNEEM. The encapsulation process has been observed to diminish the antifeedant efficacy of PONNEEM against *Helicoverpa armigera* across all tested concentrations (0.1%, 0.2%, and 0.3%). Remarkably, the larvicidal effectiveness of the nanoparticles, with the exception of CS/GLA 0.2%, surpasses that of the nonencapsulated PONNEEM [75].

Callosobruchus maculatus (F.) stands as a prominent pest affecting leguminous grains such as green gram, cowpeas, lentils, and black gram [76,77,78,79,80,81]. Zinc oxide nanoparticles were biogenically synthesized utilizing the leaf extract of *Pongamia pinnata*. The assessment of their pesticidal efficacy against *Callosobruchus maculatus*, a cowpea weevil [82] focusing on mortality rates and alterations in midgut digestive enzyme activities. The application of zinc nanoparticles resulted in a dose-dependent reduction in both the quantity of eggs laid by *C. maculatus* and the subsequent hatchability of its adults. Furthermore, the treatment with nanoparticles exhibited a discernible impact on the growth and developmental stages of *C. maculatus*, spanning larval, pupal, and total developmental phases [75].

Among the highly effective nanomaterials, aluminosilicate nanotubes stand out. These nanotubes can serve as efficient carriers for loading garlic essential oil onto nanoparticles. The incorporation of garlic essential oil onto nanoparticles has demonstrated efficacy against *Tribolium castaneum* Herbst [83]. Moreover, NiO NPs nanoparticles created with an aqueous extract derived from *Rauvolfia serpentina* leaves were also investigated for their efficacy against *Callosobruchus maculatus* infestation in *Vigna mungo*. The findings indicated a dose-dependent impact on the treated insects, manifesting in decreased fecundity and extended developmental periods [84].

3. Nanobiosensors for early detection

Nanosensors represent potent instruments for discerning nutrient insufficiency, toxicity and diseases in both plant and animal organisms. Additionally, they play a crucial part in monitoring the health status of plants, ensuring the quality and safety of food products. The integration of nanosensors contributes to the enhancement of agricultural production by optimizing input efficiency, thereby minimizing losses in essential resources such as irrigation, fertilizers and pesticides [14]. Nanosensors encompass chemical or mechanical sensing devices designed to detect specific substances or quantify physical attributes at a nanoscale, wherein at least one sensing dimension does not exceed 100 nm. These sensors can be categorized broadly as ocular nanosensors, electromagnetic nanosensors, and mechanical or vibrational nanosensors [85,86]. Timely and dependable identification of plant pathogens is vital for monitoring crop health, curtailing disease dissemination and implementing efficient management strategies.

Various methods, such as visual symptom inspection, serological assays, and DNA-based pathogen detection, have been utilized for diagnosing crop diseases. However, these techniques exhibit limitations, particularly in detecting asymptomatic stages, involving time-intensive processes, expensive equipment, susceptibility to false negatives owing to cross-contamination and dependence on professional expertise. Moreover, their application in farmers' arenas is restricted.

To surmount these challenges, recent progress in nanotechnology have facilitated the expansion of small processes, leading to the creation of biosensors for identifying pathogenic incidence in plants. These biosensors utilize antibodies, DNA and volatile complexes as sensing receptors. Consequently, nanobiosensor-dependent technology introduces a novel dimension to plant disease diagnostic systems, offering non-destructive, minimally hostile, profitable and user-responsive solutions with enhanced capabilities for detection limits, sensitivity, uniquely and on-site identification of pathogens of plants [87].

Types of Nanobiosensors

1. Fluorescence Resonance Energy Transfer (FRET) Nanosensor:

A FRET nanosensor operates on fluorescence resonance energy transfer, utilizing a donor and acceptor dye for energy transfer within 1-10 nm [88]. A substrate-specific binding domain alters fluorophore distance or orientation, causing measurable energy transfer changes [89]. It has been applied in biosensing, such as diagnosing *Colletotrichum lindemuthianum* using fluorescent bioreceptors [27].

2. Surface Enhanced Raman Scattering (SERS) Nanosensor:

SERS is a powerful spectroscopy tool that analyzes at the molecular level employing metal nanoparticle-coated surfaces to enhance Raman signals [90]. It operates through electromagnetic and chemical effects, amplifying signals up to 10^{14} times [91]. Gold and silver nanoparticles allow SERS-based sensors for pathogen identification in plants [92,87].

3. Electrochemical Nanosensors (ECN): ECN offer a highly sensitive, cost-effective method for early crop pathogen detection by converting biological events into electrical signals. Utilizing nanomaterial-based electrodes, ECN enhances the detection of redox and ion species in plants. They have been applied for field diagnostics, including microfluid microarrays for identifying fungal pathogens like *Botrytis* spp. in crops like faba bean and lentil [93,27].

4. Piezoelectric Nanosensor (PZN): PZNs convert mechanical and biofluid energy into electrical signals, enabling real-time pathogen monitoring in plants. They detect mass changes from biomolecular interaction, such as antigen-antibody binding, utilizing Quartz Crystal Microbalance (QCM). This technology provides precise pathogen detection through frequency shifts in quartz crystals [87].

Additionally, [14] demonstrated that a gold nano electrode combined with copper nanoparticles provides exceptional accuracy in quantifying salicylic acid levels in oilseeds infected by *Sclerotinia sclerotiorum*. Lau et al. (2017) [94] developed a robust point of care diagnostic method integrating surface boosted Raman scattering (SERS) with recombinase polymerase amplification (RPA) for the precise detection of multiple plant pathogens, including *Pseudomonas syringae*, *Botrytis cinerea*, and *Fusarium oxysporum* [95].

4. Nanofertilizers for nutrient management

The soil serves as the primary reservoir from which plants acquire both macronutrients and micronutrients [96]. Despite the increased crop yields resulting from the green revolution and modern farming practices, there has been a decline in vital micronutrients available for plant growth and development in the soil [97]. Consequently, there is a pressing need to enhance study tests for micronutrients, aiming for methods that are cost-effective, sensitive and capable of providing three-dimensional and consecutive insights into the bioavailable nutrient pool in both soil and plants [96].

To address this challenge, nanoformulations containing micronutrients offer a potential solution. These formulations can be employed through foliar or soil application, facilitating the availability of micronutrients for uptake by plant roots and thus facilitating the enhancement of soil health and vigor [98,99]. This approach holds promise for ensuring an adequate supply of essential nutrients to plants, mitigating the adverse effects of micronutrient deficiencies caused by changes in agricultural practices (Table 3).

Nanoformulations of iron demonstrate positive impacts on different plants, for instance, an elevation in chlorophyll levels and mitigation of chlorotic signs related to iron deficiency in soybeans [100]. Furthermore, these formulations contribute to the enhancement of growth parameters, yield metrics, and spike eminence, including 1000-grain weight, biological yield, grain yield, and grain protein content in wheat [101]. In the case of black-eyed peas, the application of iron nanoformulations results in a substantial increase of 47% in the number of pods per plant, 7% in the weight of 1000 seeds, 34% in the iron content in leaves, and 10% in chlorophyll content compared to control plants [102].

Likewise, manganese nanoparticles exhibit an encouraging influence on the yield and growth of mung beans (*Vigna radiata*) and enhance photosynthetic processes [103]. Additionally, zinc oxide nanoparticles, when applied at low concentrations, have

been shown to promote the growth of mung beans and *Cicer arietinum* seedlings [104]. These findings underline the potential of nanomaterials in positively influencing the physiological and biochemical aspects of different crops, thereby enhancing their overall productivity and nutritional content [99]. Recently, alginate/chitosan nanoparticles have been employed to augment the growth and developmental features of soybean. The findings indicate that the utilization of alginate/chitosan-based nanoparticles leads to a substantial improvement in both root and shoot length, showcasing their noteworthy potential in alleviating the impact of drought stress [105,106].

Hashem et al. (2021) [65] investigated the influence of selenium nanoparticles (Se-NPs) on the growth and development of *Vicia faba* plants. The study revealed that Se-NPs served as potent growth promoters for *Vicia faba*, influencing different morphological, metabolic, and genetic parameters. Notably, the application of Se-NPs led to a substantial enhancement in the total chlorophyll content and carotenoids when compared to control plants. The most favorable outcomes were observed when Se-NPs were administered through soaking and foliar spraying.

Krutyakov et al. (2022) [107] investigated the impact of foliar treatments employing aqueous dispersions of silver nanoparticles stabilized by polyhexamethylene biguanide hydrochloride on legume-Rhizobium symbiosis, a crucial factor influencing soil nitrogen assimilation and soybean yield. The investigation revealed that application of low doses of silver nanoparticles significantly enhanced the number of root nodules and subsequently increased soybean yield. The experimentally determined biological efficacy of silver nanoparticle dispersals was attributed to heightened enzymatic activity of peroxidases and polyphenol oxidases in the aerial portions of plants, elucidating the mechanisms behind the positive effects on legume-Rhizobium symbiosis and soybean productivity.

Table 3: Effect of different nanoparticles and nanofertilizers on crop growth

Nanoparticles/ nanofertilizers	Crop	Effect	Reference
Carbon nanotubes (CNTs)	<i>Cicer arietinum</i>	Increase in water absorption	[108]
(ZnO)	<i>Arachis hypogaea</i>	Increase in yield	[109]
ZnO, FeO and ZnFeCu-oxide	<i>Vigna radiata</i>	Increase in shoot and root biomass	[110]
Multi-wall carbon nanotubes (MWCNTs)	<i>Glycine max</i> L. Merrill	Enhanced germination and seedling growth	[111]
FeS ₂	<i>Cicer arietinum</i>	Improved seed germination, crop stand and seed yield	[112,113]
Fe/SiO ₂	<i>Arachis hypogaea</i>	Increased plant growth and biomass production	[114]
Ag NPs	<i>Glycine max</i> L. Merrill	Increase in number of root nodules, increased yield, enhanced enzymatic activity (peroxidase, polyphenol oxidase), improved rhizobium symbiosis	[107]
Fe ₂ O ₃ NP	<i>Glycine max</i> L. Merrill; <i>Cicer arietinum</i>	Increase in number of nodules and nitrogenase activity, Increased nitrogen fixation	[115]

Future Prospect

The integration of nanotechnology in legume farming holds immense potential for transforming biotic stress management in the coming decades. Future research should focus on developing crop-specific nanomaterials that offer targeted protection against key pathogens, pests and parasitic weeds affecting legumes. Advancements in nano-enabled smart delivery systems for biopesticides and RNA interface (RNAi) based technologies may significantly enhance plant defense mechanisms while minimizing environmental toxicity. There is also a need to explore the synergistic effects of combining nanotechnology with other modern approaches, such as gene editing (e.g., CRISPR), microbial consortia and integrated pest management (IPM), to create holistic and sustainable solutions.

Long term field trials across diverse agro-ecological zones will be critical to validate lab-drawn results and ensure scalability. Moreover, the development of eco-friendly, biodegradable nanomaterials can address current concerns related to nanoparticle toxicity and ecological impact. In addition, the incorporation of nanosensors and digital agriculture tools in legume farming can revolutionize real-time stress monitoring, early disease detection and precision intervention. Policy support, farmer awareness programs and robust regulatory frameworks will also play pivotal roles in ensuring the responsible and widespread adoption of nanotechnology in agriculture. With continued innovation and interdisciplinary collaboration, nanotechnology is poised to play a transformative role in securing legume productivity and food security under increasing biotic stress pressures.

Conclusion

In conclusion, the utilization of nanotechnology for biotic stress management in leguminous crops represents a groundbreaking avenue with considerable potential for revolutionizing agricultural practices, offering innovative solutions that address the complex challenges related to pests, diseases, weeds, and nutrient deficiencies. The ability to precisely target and modulate plant responses at the nano-scale offers a level of precision and efficiency that traditional methods struggle to achieve. Moreover, the deployment of nanotechnological interventions not only contributes to bolstering crop resistance but also holds promise in mitigating environmental impacts associated with conventional pest and pathogen control measures. By promoting sustainable farming practices, nanotechnology aligns with the imperative to address global food security challenges while minimizing adverse ecological consequences. However, the journey towards incorporating nanotechnology into leguminous crop management is not without challenges. Concerns surrounding nanoparticle toxicity, environmental persistence, and regulatory frameworks necessitate careful consideration to safeguard the harmless and accountable submission of these innovations. In essence, the multifaceted application of nanotechnology in biotic stress management for leguminous crops holds great promise for sustainable agriculture. By leveraging the unique capabilities of nanoparticles, we can advance precision farming, reduce environmental impact, and contribute to global food security. Continued research, coupled with thoughtful regulation and widespread adoption, will be instrumental in realizing the full potential of nanotechnology to transform leguminous crop cultivation into a resilient, efficient, and sustainable endeavor for the future.

Acknowledgement: Nil

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this manuscript.

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