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The Future of Nanotechnology in Plant Pathology

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Abstract

Engineered nanoparticles are materials between 1 and 100 nm and exist as metalloids, metallic oxides, nonmetals, and carbon nanomaterials and as functionalized dendrimers, liposomes, and quantum dots. Their small size, large surface area, and high reactivity have enabled their use as bactericides/fungicides and nanofertilizers. Nanoparticles can be designed as biosensors for plant disease diagnostics and as delivery vehicles for genetic material, probes, and agrichemicals. In the past decade, reports of nanotechnology in phytopathology have grown exponentially. Nanomaterials have been integrated into disease management strategies and diagnostics and as molecular tools. Most reports summarized herein are directed toward pathogen inhibition using metalloid/metallic oxide nanoparticles as bactericides/fungicides and as nanofertilizers to enhance health. The use of nanoparticles as biosensors in plant disease diagnostics is also reviewed. As global demand for food production escalates against a changing climate, nanotechnology could sustainably mitigate many challenges in disease management by reducing chemical inputs and promoting rapid detection of pathogens.

INTRODUCTION

Recent projections have stated that global demand for food production will need to double by 2050 (129). This alarming prediction becomes more dire as climate changes are likely to cause a disruption of food production cycles by extending drought events and raising average daily temperatures in many agriculturally sensitive regions. The challenges faced by plant pathologists and other agriculturalists are daunting. However, nanotechnology stands as a new weapon in our arsenal against these mounting challenges in disease management and plant health. The utilization of nanotechnology in plant disease management, diagnosis, and genetic transformations is still in its infancy and has only begun to be explored in the plant pathology literature. For example, from January 1, 2017, to December 1, 2017, the number of US patents issued for nanotechnology with applicability for human medicine surpassed 3,100, whereas US patents with applicability to agriculture (including animal husbandry) were fewer than 650 in the same 11-month period. In fact, of the 147 articles cited in this review, only 23% were taken from journals that traditionally report plant pathology research. This demonstrates a lack of awareness among plant pathologists of how nanotechnology can be applied to plant disease systems. In fact, the possibilities are numerous and extend from the direct application of nanoparticles and their composites to plants as fertilizers/fungicides to more indirect utilization of nanotechnology in diagnostics and precision agriculture.

In accepting this assignment for the *Annual Review of Phytopathology*, we now admit that we were somewhat unprepared for the daunting task of summarizing the cascade of information on nanoparticles that has appeared since our last summary (118). Therefore, we have restricted the review to brief definitions, syntheses, and a review of uses of nanotechnology in disease management and disease diagnostics, specifically as they relate to bactericides/fungicides and novel fertilizers to enhance disease resistance. Although outside the scope of this report, there is also an expanding array of new tools and techniques using nanoparticles as delivery vehicles for genetic material in plants. Our aim is to inform and excite researchers about the rapidly expanding opportunities associated with nanotechnology in phytopathology.

DEFINITION, PROPERTIES, AND SYNTHESIS

Nanoparticles are currently defined as any material that has one or more dimensions at the scale of 1 to 100 nm (117). This definition has allowed for a huge array of possible natural and engineered composites/materials to be labeled as nanoparticles. Natural nanoparticles also occur in many forms, such as volcanic dust and oceanic salt sprays (62, 140). In addition, many viral and viroid particles fall within the definition of a nanoparticle but are not discussed. Naturally occurring nonviral nanoparticles are quite irregular and vary in size, whereas engineered nanoparticles are generally more uniform and can have unique shapes, such as spherical balls, sheets, rods, and, in more intricate arrangements, multiwalled tubes (carbon nanotubes) or bifurcating tree-like structures (dendrimers). Engineered nanoparticles can be manufactured to specific dimensions and designed in countless composite arrays, making their function and utility applicable within many disciplines. This review discusses engineered nanoparticles manufactured both chemically and biosynthetically.

As a result of nanoparticles' small size and large surface-area-to-volume ratio, they can be reactive and bind, absorb, and carry compounds such as small-molecule drugs, DNA, RNA, proteins, and probes with high efficiency (5). Aside from surface area, nanoparticles differ in other properties when compared with their larger bulk equivalents. Gold (Au), which is inert and obviously golden in the larger form, can be reactive and red in color at the nanoscale size. TiO and ZnO are normally white but appear colorless at the nanoscale size. Nanoparticles can melt at lower temperatures and are frequently more reactive than their larger bulked equivalents. Nanotechnology has sought to exploit many of these behavioral changes, opening the door to many industries with a vast array of new products and uses.

Nanoparticles can be synthesized by numerous methods, and the reader is referred to reviews documenting these procedures (11, 33, 94). New processes and platforms are being developed so fast that any description is likely to be outdated soon. A considerable amount of literature has been devoted to the production of nanoparticles in vivo by plants and microorganisms. The biosynthetic methods are quite diverse, but organisms or their extracts are briefly exposed to a metallic salt that biologically reduces the metal to a nanoscale size. The nanoparticles are then harvested, characterized, and available for use (55, 86). This single-step green-synthesis process is rapid, can be conducted at ambient temperature and pressure, and is readily scalable. A 2015 review reported the production of metallic nanoparticles from more than 48 species of fungi (6). Several species in the plant-pathogenic genera of *Fusarium*, *Aspergillus*, *Verticillium*, and *Penicillium* have been employed to biosynthesize nanoparticles. The inherent irony of using the very fungal pathogens being suppressed to make the fungicidal nanoparticles is not lost on the authors. Green synthesis of nanoparticles could have great impact on their use in organic production disease management if the certifying governing boards of organic growers decide to approve the green synthesis of these materials as a treatment option.

TYPES OF NANOPARTICLES USED IN PLANT PATHOLOGY

Of the many nanoparticles that exist, we must mention that most currently do not have application in plant pathology. However, this will likely change as applications currently being explored in nanomedicine against human pathogens make their way into the plant disease management arena (**Table 1**). At present, nanoparticles of metalloids, metallic oxides, nonmetals (single and composites), carbon nanomaterials (single- and multiwalled carbon nanotubes, graphene oxides, and fullerenes), and functionalized forms of dendrimers, liposomes, and quantum dots have begun to infiltrate plant pathology (**Figure 1**).

Metalloids, Metallic Oxides, and Nonmetal Nanoparticles

Most nanoparticle information relating to plant disease has involved the use of metalloids, metallic oxides, or nonmetals as either bactericide/fungicides or nanofertilizers to affect disease resistance (27). The illustration highlights some of the keynote studies that demonstrated the value of nanoparticles in suppression of foliar, stem, fruit, and root rot pathogens (**Figure 2**). The following text summarizes the research as it pertains to the metalloids B and Si; the metallic oxides of Ag, Al, Au, Ce, Cu, Fe, Mg, Mn, Ni, Ti, and Zn; and the nonmetal S. Given that the vast majority of phytopathology literature focuses on the nanoparticles Ag, Cu, and Zn, they are discussed first and in greater detail.

Nano-Silver. Nanoparticles of Ag were the first to be investigated in plant disease management given their historically known antimicrobial activity (110). Park et al. (98) combined nano-Ag and nano-Si with a water-soluble polymer and applied it to the leaves of cucumbers at 0.3 μg/mL three days before the plants were exposed to *Podosphaera xanthii.* The pathogen was not detected on the leaves but was evident on untreated controls. Because Park et al. (100) did not test the Ag nanoparticle separately, one cannot assess the individual effect of Ag alone. However, Kim et al. (71) investigated the effectiveness of nanoparticles of colloidal Ag against rose powdery mildew. The authors prepared a double-capsulized nanoscale version of Ag by reacting Ag ions

Table 1 Types of nanoparticles and their definitions and potential uses in plant pathology

Type	Definition	Potential use in plant pathology
Metalloids, metallic oxides, nonmetals, and their composites	Engineered metals at nanoscale in cubes, spheres, bars, and sheets	Bactericides/Fungicides Nanofertilizers Delivery vehicle for antimicrobials and genetic material
Carbon nanomaterials	Allotropes of carbon designed at the nanoscale	Multiple uses
Single-walled or multiwalled nanotubes	Graphene sheets rolled into single or multiple tubes	Antimicrobial agents Delivery vehicle for antimicrobials and genetic material
Fullerenes (buckyballs)	60 carbon atoms in a specific soccer-ball arrangement	Antimicrobial agents Delivery vehicle for antimicrobials and genetic material
Graphene oxide sheet (reduced or oxide forms)	Graphene oxide sheet	Antimicrobial agents Delivery vehicle for antimicrobials and genetic material
Liposomes	A lipid enclosing a water core	Delivery vehicle for genetic or antimicrobial products
Dendrimers	Nanomaterial with tree-like appendages that radiate from a central core	Delivery vehicle for genetic or antimicrobial products
Nanobiosensor	A nanoparticle that combines a biological component for detection	Diagnostics, research tool
Nanoshell	Nanoparticles composed of a gold shell surrounding a semiconductor	Diagnostics, research tool
Quantum dots	Inorganic fluorescent, crystalline semiconductor nanoparticles used in biosensors	Diagnostics, research tool

with a reducing agent in the presence of a stabilizer. When it was sprayed at 10 μg/mL onto roses symptomatic with powdery mildew, the researchers observed a slow disappearance of the colonies of *Sphaerotheca pannosa* on the leaves and did not observe pathogen presence until seven days later. Although their experiment was not repeated and did not include a control, it suggested that nanoparticles of Ag may have curative effects. Lamsal et al. (75) explored this concept further and applied nanoparticles of Ag at 10, 30, 50, and 100 μg/mL to leaves of cucumbers and pumpkin either three or four weeks before powdery mildew appeared or after the plants became infected. At both times of application, plants sprayed with 100 μg/mL of nanoscale Ag had only a 20% incidence of disease as opposed to an 80% incidence in untreated plants. This study was noteworthy for two reasons. First, the nanoparticle treatment used in this trial performed as well as the commercial fungicide (20% versus 26% disease incidence, respectively), and, second, a curative response was documented, suggesting future rescue treatments could be realized with nano-Ag. Lamsal et al. (74) found nano-Ag applied at 100 μg/mL to peppers before anthracnose outbreaks effectively suppressed the disease. Jagana et al. (56) raised the prospect of postharvest treatments with nano-Ag. They biosynthesized nano-Ag from plant extracts and treated banana fruits at varying rates for postharvest control of *Colletotrichum musae*. Disease severity was lowest (6.7%) at the highest rate of 2,000 μg/mL nano-Ag, with the untreated control at 75.6%.

Other studies have confirmed the efficacy of Ag nanoparticles against foliar fungal pathogens, with the greatest efficacy observed when the nanoparticle was applied preventatively (60, 85). Moussa et al. (88) produced Ag nanoparticles in the culture supernatant of *Serratia* spp. and

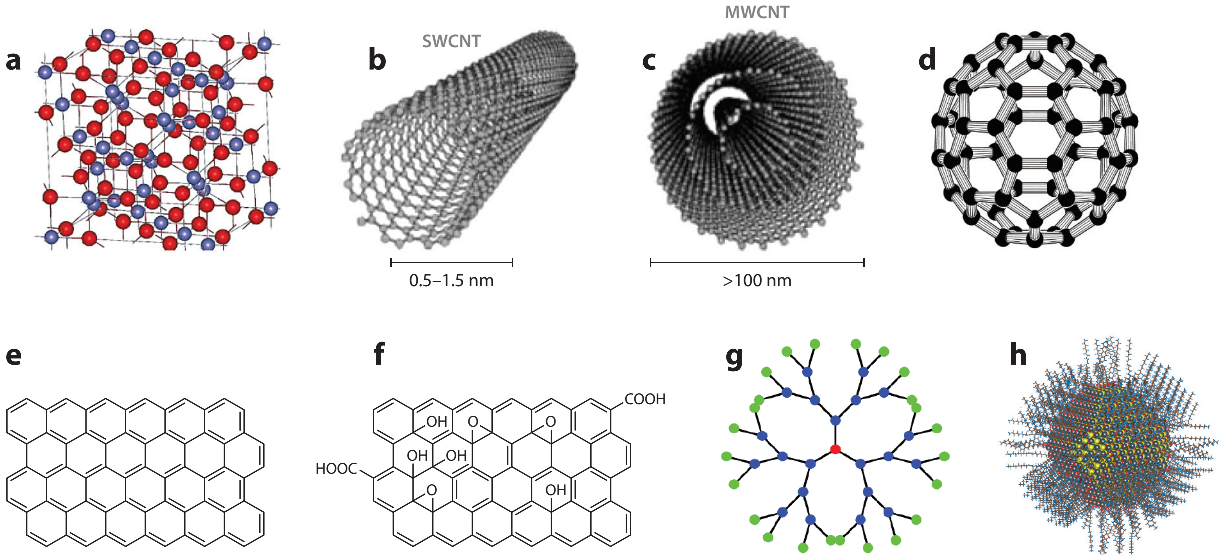

Figure 1

Schematic diagrams of the different types of nanoparticles used in phytopathological research. (*a*) Molecular arrangements of metal oxide nanoparticles. (*b*) Single-walled carbon nanotube (SWCNT). (*c*) Multiwalled carbon nanotube (MWCNT). (*d*) Fullerene. (*e*) Graphene nanoparticle sheet. (*f*) Graphene-oxidized nanoparticle sheet. (*g*) Dendrimer. (*h*) Quantum dot. Panel *a* used with permission from Creative Commons BY-SA 3.0. Panels *b* and *c* used with permission from Dr. Bruno Ribeiro, Universidade Federal de São Paulo Brazil. Panel *d* used with permission from Creative Commons BY-SA 3.0. Panels *e* and *f* used with permission from Dr. Kurt Sieberns, TCI Europe N.V. Panel *g* used with permission from Dr. Hak Fun Chow, Chinese University of Hong Kong, China. Panel *h* used with permission from Creative Commons BY-SA 3.0.

found that concentrations as low as 2 μg/mL were 100% effective in preventing both conidial germination of *Bipolaris sorokiniana*, the agent that causes spot blotch of wheat on detached leaf assays, and on-leaf infection of the pathogen in greenhouse trials. Histochemical staining revealed that nano-Ag was associated with the induction of lignin deposition in the vascular bundles. This type of green synthesis of nano-Ag offers a unique and potentially effective approach for disease management.

As of this writing, the only soilborne diseases that have been suppressed with Ag nanoparticles are those caused by *Phytophthora parasitica*, *Fusarium* spp., and *Meloidogyne* spp. Low doses of less than 100 μg/mL of nanoscale Ag are highly toxic. Ali et al. (7) found that single applications of 10 or 100 μg/mL of nano-Ag 24 h before inoculating *Artemisia absinthium* with *P. parasitica* resulted in 78% or 95% plant survival, respectively, compared to 5% survival in the untreated controls. Mallaiah (82) found that nanoparticles of Ag (800 μg/mL) suppressed Fusarium wilt of the commercial ornamental flower *Crossandra* spp. in pot culture. Wilt was reduced from 75% in the untreated control to 55%. Aguilar-Méndez et al. (3) and Jung et al. (61) tested various formulations of nanoscale Ag against *Sclerotium cepivorum* and *Colletotrichum gloeosporioides* and found that all were inhibitory. Plant biomass was increased following application to healthy green onions, but because inoculated plants were not similarly evaluated, particle efficacy as a management tool could not be determined (61).

The effect of nano-Ag on suppressing plant-parasitic nematodes has yielded very promising results. Four studies have documented the effect of nano-Ag on species of *Meloidogyne* (1, 10, 24, 91). Using engineered or biosynthesized nano-Ag on stage two juveniles of *Meloidogyne*

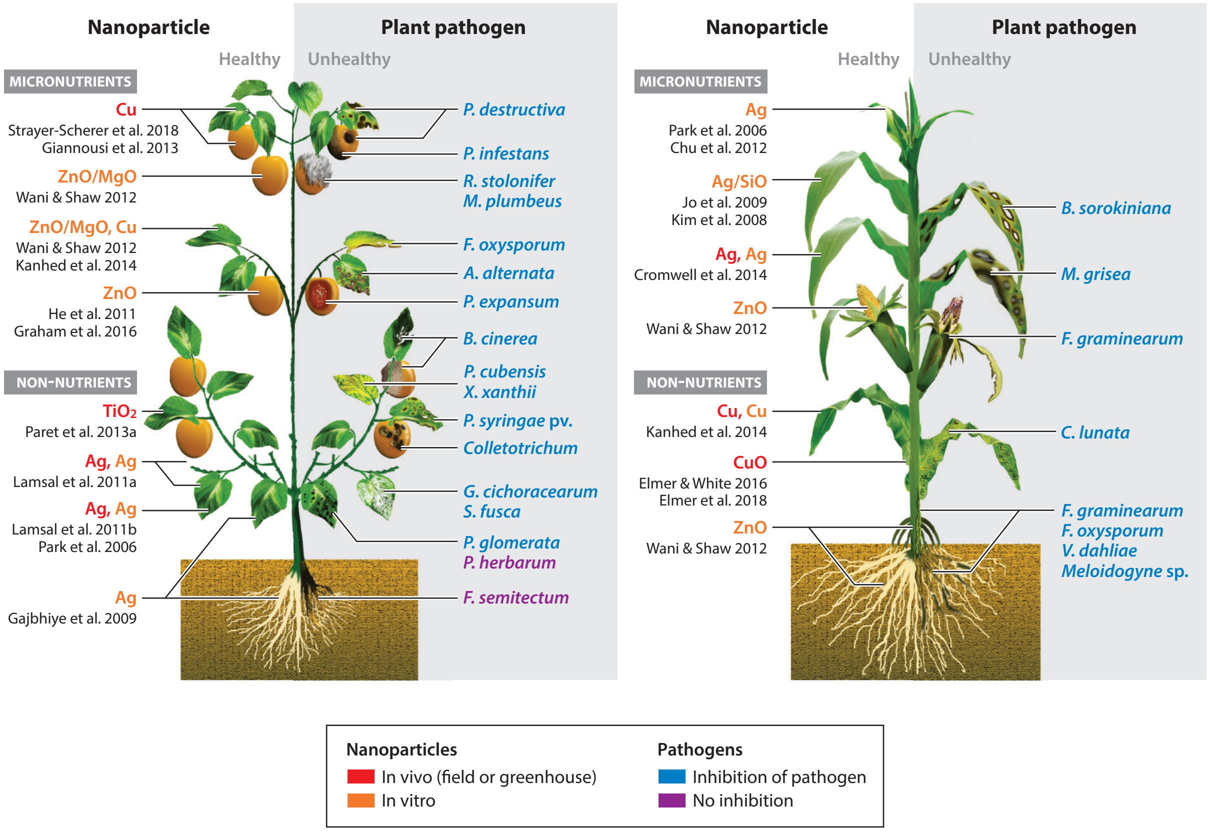

Figure 2

Illustrative examples of nanoparticle studies with micronutrients and non-nutrients on diseases of citrus and maize. Adapted with permission from Reference 118.

spp. completely inactivated all juveniles in these studies, typically within 6 h. A commercial putting green infested with *Meloidogyne graminis* received biweekly application of nano-Ag at 90.4 mg/m^2 and had enhanced turfgrass quality and reduced root gall formation (24). Suppression of *Meloidogyne incognita* and *Meloidogyne javanica* was achieved with low rates of nano-Ag and was equal to the level of control provided by conventional nematicides (1, 10). Nassar (91) added biosynthesized nano-Ag to the nematicide organophosphorodithioate and found that the LC_{50} to suppress *M. incognita* was lowered from 7.2 to 5.6. Because nano-Ag was not examined individually, it was not clear how much derived benefit might be obtained from this combination.

Ocsoy et al. (95) advanced a new nano-Ag product. They developed a dsDNA-directed Ag nanoparticle prepared on graphene oxide to suppress a bacterial disease caused by *Xanthomonas perforans* on tomatoes in the greenhouse. A 100 μg/mL application reduced the severity of bacterial spot disease when compared with untreated controls and was comparable to conventional bactericide treatments. These results have major importance given the high degree of Cu tolerance among the population of *X. perforans* and that the nano-Ag treatment was comparable to the conventional spray treatment. Solgi et al. (123) found that Ag nanoparticles applied at 5 μg/mL cut Gerbera flowers extended vase life, but the authors reported that the control was not superior to an equivalent solution with $AgNO_3$. The Solgi et al. (123) study raises important issues regarding the value of the nanoparticle form versus the standard formulations of $AgNO_3$ and highlights the need to run appropriate salt and larger bulk equivalent forms as controls when performing this type of nanoparticle research.

The potential for using nanoscale Ag with a biocontrol agent is encouraging. Mallaiah (82) observed further enhancement of Fusarium wilt suppression on *Crossandra* spp. when the Ag was combined with the biocontrol agents *Bacillus subtilis*, *Pseudomonas fluorescens*, and *Trichoderma viride*. Flower yield was increased 5% over the control with nano-Ag but increased 12%, 14%, and 15% when Ag was combined with *B. subtilis*, *P. fluorescens*, and *T. viride*, respectively. Combining nano-Ag with fungicides offers many new possibilities for reducing chemical inputs and delaying pathogen resistance. Fungicidal activity of fluconazole increased 0.35- to 0.37-fold against *Alternaria alternata*, *Cladosporium herbarum*, and *Fusarium oxysporum* (15). Gajbhiye et al. (41) biosynthesized extracellular Ag nanoparticles from *A. alternata* and found that when combined with the fungicide fluconazole, the antifungal activity was enhanced against the plant pathogen *Phoma glomerata* but not against *Phoma herbarum* or *Fusarium semitectum*.

Although of less value in assessing the applicability to disease management, there are a number of studies documenting the inhibitory effect of Ag nanoparticles against plant pathogens in vitro (3, 25, 30, 35, 60, 65, 72, 73, 77, 84, 90, 96, 141). *Citrobacter freundii*, *Erwinia cacticida*, and all xanthomonads tested in vitro have been inhibited by nano-Ag at rates as low as 2.5 μg/mL (77, 101, 107). Biosynthesis of nano-Ag by fungi is not difficult. Moussa et al. (88) reported that more than 48 species of fungi were capable of producing nanoparticles of Ag, including species of plant-pathogenic fungi such as *Aspergillus*, *Fusarium*, *Penicillium*, and *Verticillium*.

Other than direct antimicrobial action, it is not clear whether Ag nanoparticles also activate defense mechanisms. When nano-Ag was hybridized with silica and applied as a pretreatment to *Arabidopsis thaliana* at 1–10 μg/mL, an increase in PR1, PR2, and PR5 and induced disease resistance to the bacterial disease pathogen *Pseudomonas syringae* pv. *tomato* were noted when compared to the water control treatment (23). However, Si alone can induce these defense products (28), and the individual effects of nano-Ag need to be examined. In another disease system, soybean seedlings were misted with 2–4 mL of 500 μg/mL nano-Ag (roughly 1–2 mg AgO per seedling) and then were transplanted into new potting soil infested with *Fusarium virguliforme*. After several weeks, the disease ratings were reduced from 75% in the untreated control to 55% in the nano-Ag treatment (C.D. Perez & W.H. Elmer, unpublished data). Although further validation is required, an induction of disease resistance may be operating.

One criticism of many of the early studies with Ag nanoparticles is the lack of controls with standard salts such as $AgNO_3$ or other conventional products. The environmental impacts and costs of using nano-Ag in plant disease management are therefore ambiguous. The current value of Ag is $0.55 per g; thus, a rate of 100 μg/mL applied at 936 L/ha (100 gal/ac) would cost approximately $51.50 per ha. If the environmental aspects can be adequately addressed, nanoparticles of Ag might serve as candidates for disease management when other conventional strategies are ineffective.

Nano-Copper. The well-known antimicrobial properties of Cu and its long history in controlling plant diseases (36) make nano-Cu a logical choice for plant disease management. The first studies that examined nano-Cu as a bactericide/fungicide in the field were done by Giannousi et al. (44) who engineered nano-CuO, Cu_2O, and Cu/Cu_2O composites and compared foliar applications to registered commercial Cu-based fungicides, including Kocide 2000 35 WG, Kocide Opti 30 WG, Cuprofix Disperss®, and Ridomil Gold Plus, for their ability to suppress *Phytophthora infestans* in tomato. They monitored the disease for 10 days once symptoms appeared and reported that the most effective product for suppressing leaf lesions was the CuO nanoparticles at 150–340 μg/mL, followed by the nano-Cu/Cu_2O composite. Importantly, the nano-Cu products were equal or superior to the commercial Cu-based products, delivered less Cu/ha and did not cause overt phytotoxicity.

Strayer-Scherer et al. (125) advanced a new strategy using engineered nano-Cu nanoparticle composites that possess a core-shell Cu, multivalent Cu, and fixed quaternary ammonium copper and compared these materials as alternatives to standard Cu-based products to suppress bacterial spot caused by *X. perforans*. Although the bacterium had acquired resistance to Cu bactericides, all of the nano-Cu products significantly reduced bacterial spot disease severity in the greenhouse and field when compared to Cu fungicides and water controls ($P < 0.05$). The nano-Cu products were believed to more effectively penetrate bacterial membranes and release higher amounts of the active Cu ions. Although yields were unaffected by treatment, given that the nanoparticle formulations were more effective than the conventional Cu-mancozeb and delivered less metallic Cu, these findings highlight the value of nanoscale Cu formulations in managing Cu-tolerant bacterial diseases.

Elmer and colleagues (36, 37) explored a new direction for using Cu nanoparticles in disease management by examining their efficacy as nanofertilizers/supplements to enhance disease resistance (38). After screening six metallic oxides, the authors reported that Cu nanoparticles were the most effective at suppressing Fusarium wilt of tomato and Verticillium wilt of eggplant. When nanoparticles of CuO were applied foliarly to leaves of young seedlings at 500 μg/mL and then planted into potting mix infested with *F. oxysporum* f. sp. *lycopersici* or *Verticillium dahliae*, disease severity ratings were consistently lower than the controls and the bulked oxide equivalents (37). In field studies, treating young transplants with nano-CuO produced seasonal increases in tomato and eggplant yields. Eggplants treated with nanoparticles of CuO averaged 24% greater yield than controls. Fruits had no more Cu in the edible flesh than fruit from untreated plants and root digests from greenhouse assays revealed higher levels of copper, presumably due to nanoenabled basipetal translocation. A $50 investment in a novel (nanoscale) form of a plant micronutrient resulted in an estimated $10,000 per acre increase in yield for the grower.

Elmer et al. (36) extended the foliar application technique by spraying CuO nanoparticles on watermelon seedlings, followed by transplantation of the seedlings into potting mix infested with *F. oxysporum* f. sp. *niveum.* In five out of eight greenhouse experiments, nano-CuO suppressed Fusarium wilt. Acid digests of plant roots revealed greater Cu in the roots of nano-CuO-treated plants relative to other treatments or controls, again suggesting basipetal translocation was occurring as a result of the nano size (36, 138). In one field experiment, nano-CuO applied twice to seedlings yielded 39% more fruit than untreated controls; in another, a single application of nano-CuO to transplants produced an average of 45% more fruit when compared to untreated controls or to the commercial Cu fungicides Kocide® 2000 (Cu hydroxide) and Cu octanoate. Analysis of gene expression [by RT-qPCR (quantitative reverse transcription polymerase chain reaction)] in watermelon roots revealed strong upregulation of polyphenol oxidase and pathogen-related genes in plants treated with nano-CuO and inoculated with *F. oxysporum* f. sp. *niveum*. Enzymatic assays also found polyphenol oxidase activity was elevated in plants treated with CuO nanoparticles, although the results were more variable. The data suggest that nano-CuO may serve as a highly effective delivery agent for Cu that can subsequently enhance disease suppression. One unresolved question is whether the nanoparticle itself or the ions are being transported within the plant (136). Additional work is needed to optimize potential treatment platforms.

Ponmurugan et al. (105) conducted a very similar study in India using biosynthesized nano-Cu from *Streptomyces griseus* cultures to suppress the red root rot disease of tea caused by the fungus *Poria hypolateritia*. Increasing rates of nano-CuO were sprayed on plants infected with *P. hypolateritia* (1.5 L/bush) and compared to the bulked CuO equivalent or the fungicide carbendazim. The highest rate of nano-Cu (2.5 μg/mL) gave a 53% reduction in disease severity as compared to 57% reduction with carbendazim. Nano-Cu gave the highest leaf yield and improved the edaphic properties, presumably through enhancing root health. It would be interesting to determine whether similar physiology responses were occurring in tea as were observed in the watermelon above.

The only study to date to examine the effect of nano-Cu on nematodes was on *Xiphinema index* on grapes in Hungary (26). A nano-Cu form of Bordeaux mixture (Bordóilé Neo SC) was shown to provide control of *X. index* in grape orchids, but it was not clear whether the nano-Cu was superior to the standard formulation. Given the suppression observed with nano-Ag, it would seem reasonable to think nano-Cu may also have comparable efficacy against nematodes.

Although the antibacterial activity of CuO nanoparticles toward plant-pathogenic xanthomonads is well documented (95, 125), several have reported antifungal activity against *A. alternata*, *Botrytis cinerea*, *Curvularia lunata*, *F. oxysporum*, and *Phoma destructiva* (18, 63, 96, 133). Unpublished research (W.H. Elmer) found that increasing rates of CuO nanoparticles in shake culture with *F. virguliforme* in repeated trials and found mycelial biomass was reduced by 54% to 47% at a dose of 100 μg/mL. In contrast, when nanoparticles were mixed into the agar medium, Elmer & White (37) did not observe a restriction in radial growth of *F. oxysporum* f. sp. *lycopersici* at doses up to 1,000 μg/mL of CuO nanoparticles. Notably, the role of the agar medium in restricting the release and availability of Cu ions is not known.

Given the high amount of Cu currently applied in agriculture and the increasing number of Cu-tolerant bacterial strains that have emerged, it could be argued that any strategy using Cu would further erode the use of Cu as a management tool. Arguments against this claim are that the amount of Cu being supplied in the nano form is currently much less than the conventional Cu-based product. For example, Strayer-Scherer et al. (125) calculated that nanoscale Cu composites applied to suppress bacterial spot of tomato delivered 88% less metallic Cu than the conventional Cu-mancozeb. Furthermore, Elmer & White (37) found that foliar application of nanoparticles of CuO to young seedlings resulted in only 1 to 2 mg of CuO applied per seedling (roughly equivalent to 7.5–15.0 g/ha, assuming 7,500 plant/ha). Arguably, these studies show nano-Cu delivers a more active load of Cu at significantly smaller nominal rates and support the growing consensus that nano-Cu and its composites offer multiple benefits in plant disease management.

Nano-Zinc. Similar to Ag and Cu, the antimicrobial activity of Zn nanoparticles to plant pathogens has been examined by several laboratories (47, 48, 54, 108, 139). Most in vitro assays have found that nano-Zn inhibits bacteria (47, 54, 66), a range of fungal pathogens, including *A. alternata*, *B. cinerea*, *F. oxysporum*, *Mucor plumbeus*, *Penicillium expansum*, *Rhizoctonia solani*, *Rhizopus stolonifera*, and *Sclerotinia sclerotiorum* (50, 114, 139), as well as the nematode *M. incognita* (66). More importantly, field and greenhouse studies have demonstrated disease suppression with nano-Zn (31, 47, 98). More attention has been given to managing bacterial diseases with nano-Zn than diseases caused by other pathogens (47, 98). Paret et al. (98) employed photocatalyst technology with nanotechnology to develop a novel antimicrobial light-activated TiO_2/Zn nanoparticle composite to suppress bacterial leaf spot on rose caused by a *Xanthomonas* sp. Experimental rose plots were exposed to the nano-TiO_2/Zn product at 500 to 800 μg/mL and activity was compared to a conventional bactericide. TiO_2/Zn activity was comparable with the ornamental industry standard for management of rose diseases. In fact, one Zn-based nanoproduct (Zinkicide™) is in registration for use in controlling citrus canker (145). In 2014 and 2015, Graham et al. (47) compared two nanoparticle formulations of Zinkicide™, a plate-like Zinkicide™ SG4 and particulate Zinkicide™ SG6, on citrus canker caused by *Xanthomonas citri* subsp. *citri*. Both formulations were equivalent in suppressing canker lesions. When Zinkicide™ SG6 was foliarly applied to sweet orange in the greenhouse and inoculated using an injection filtration two days later, canker lesions were reduced by 38% and 42% in two repetitions of the experiment. In field trials, Zinkicide™ SG6 reduced disease incidence more than did traditional cuprous oxide and cuprous oxide/zinc oxide bactericides. Other noted benefits with Zinkicide™ treatments were suppressed citrus scab (*Elsinoe fawcettii*) and melanose (*Diaporthe citri*) on grapefruit. In Egypt, Derbalah et al. (31) compared foliar applications of ZnO nanoparticles at 500 μg/mL to the conventional fungicide tetraconazole along with additional treatments such as nano-silica, diatomaceous earth, and six bacterial biocontrol agents, to suppress Cercospora leaf blight of sugar beet. In a two-year study, the nano-Zn treatment was second only to tetraconazole at increasing leaf dry weight, root yield, and sugar content and significantly reducing disease severity. Dimkpa et al. (32) examined the interaction of nanoparticle of Zn with the biocontrol agent *Pseudomonas chlororaphis* O6 and *Fusarium graminearum* in vitro. These findings confirm many studies that associate Zn nutrition with increased biocontrol efficacy (34). ZnO nanoparticles did not affect the biocontrol bacterium and had no impact on the effectiveness of metabolites from the bacterium at inhibiting *F. graminearum* in vitro. These findings demonstrate the potential for combining nano-Zn formulations with other existing strategies for improving crop health. Future studies with Zn-based nanoparticles look very promising. In addition, the recent advances in registering nano-Zn products (Zinkicide™) for crop disease management is encouraging and emphasizes the recognition of nanoparticles as a viable alternative to conventional strategies.

Less-studied metalloids, metallic oxides, and nonmetal nanoparticles. Considerably less information exists on how nanoparticles of Al, Au, B, Ce, Fe, Mg, Mn, Ni, and S affect plant disease. Much information summarized here originated from single reports, including some on in vitro activity, and further validation is necessary. Of these nanoparticles, the greatest attention with regard to disease management has been given to nanoparticles of MgO, SiO, and TiO.

For decades, the metalloid Si nanoparticles have found multiple uses in biosensors, optical materials, biocatalysts, electrochemistry, and immunochemistry; the first mention of nano-Si in plant pathology was an Ag-Si composite for suppression of powdery mildew. As discussed above, Park et al. (100) first constructed an Ag-Si that suppresses powdery mildew at 0.3 μg/mL. The study did not differentiate between the Ag and Si; thus, the individual elemental effects could not be assessed. However, maize grown in a loamy soil amended with nano-Si at 5, 10, or 15 kg/ha and compared to untreated control or bulked silica at 15 kg/ha had the lowest amount of disease caused by *Aspergillus niger* and *F. oxysporum* at the two higher nano-Si rates (126). Further investigations of total phenols, phenylalanine ammonia lyase, and peroxidase and polyphenol oxidase found that the biomolecules were more expressed at the higher nano-Si rates. Elmer et al. (36) included nano-SiO as a foliar spray to determine its effects on Fusarium wilt of watermelon and found no differences in plant weights or in disease severity rankings when compared to the untreated control.

Most researchers agree that the benefits of Si on plant resistance is greatest when the Si ions are continuously absorbed by the plant (28). Therefore, a single application of Si from a nanoparticle source may not be sufficient unless a very slow dissolution of Si from the product can be continuously released to the plant at a young age. Additional studies need to be designed with newer forms of nano-Si, perhaps as mesoporous nano-Si constructs that continuously release Si to susceptible plants over extended periods of time. Importantly, their unique hollow shape could also serve as a delivery vehicle for other plant chemotherapeutics (12, 130, 147).

Nanoparticles of MgO were examined after discovery of their antimicrobial activity (52). Imada et al. (53) demonstrated on tomatoes that MgO nanoparticles would increase systemic disease resistance against *Ralstonia solanacearum* if applied preventatively. Simultaneous treatment of tomato seedlings with nano-MgO (500 or 1,000 μg/mL) and *R. solanacearum* had only a marginal effect on disease, but when roots were pre-exposed to 350 μg/mL nano-MgO and then inoculated 4–8 days later with *R. solanacearum*, plants showed significant reduction in disease incidence. Liao et al. (78) reported suppression of bacterial spot on tomato with rates of 200 μg/mL nano-MgO. Imada et al. (53) reported that nano-MgO treated roots produce a rapid generation of reactive oxygen species along with upregulation of *PR1*, jasmonic acid, ethylene, and systemic resistance–related genes. Histochemical assays showed that β-1,3-glucanase and tyloses appeared in the vascular tissues of the hypocotyls of plants exposed to nano-MgO. Although less attention has been directed toward inhibiting fungal pathogens in vitro, Wani & Shag (139) demonstrated that the germination of conidia of *A. alternata*, *F. oxysporum*, *R. stolonifera*, and *M. plumbeus* was more inhibited by nano-MgO than nano-ZnO at 50 μg/mL. No published data currently exist for applied studies with nano-MgO and fungal pathogens.

The role of nano-Ti in photosynthesis has captured the attention of researchers (87). In fact, the photocatalytic activities of nano-TiO may contribute to its antifungal activity (17, 29). Pure TiO_2 is more active in the UV range, but when prepared in composites with metallic and nonmetallic elements, nano-TiO_2 becomes more active in visible light wavelengths. Paret et al. (99) developed a novel antimicrobial light-activated nano-TiO_2/Zn nanoparticle composite to suppress bacterial leaf spot on rose caused by a *Xanthomonas* sp. Because the composite also contained the disease-suppressing nano-ZnO, one cannot distinguish between the effects of TiO_2 and nano-Zn. However, this finding should stimulate more activity on nano-TiO_2 research for plant disease control given reports of disease suppression with Ti on cereals, rice, and tomatoes (19) as cited by Owolade et al. (97). Boxi et al. (17) and De Filpo et al. (29) found that nano-TiO_2 had strong antifungal activity; however, Elmer et al. (36) observed no disease-suppressive effect of nano-TiO on Fusarium wilt of watermelon.

Given the long history of S in fungicide use, it is surprising that to date only three reports discuss the effects of S nanoparticles on plant pathogens (22, 45, 109). One study directly compared nano-S to two other conventional forms of S fungicides (elemental S and Sulphur 80 WP) in vitro for suppression of powdery mildew of okra caused by *Erysiphe cichoracearum* (45). Using detached leaf assays, nano-S inhibited conidial germination and cleistothecia became sterile with distorted appendages. More importantly, the nano-S performed better than the commercial S fungicides, and delivered less total S per application than the conventional fungicide. Choudhury et al. (22) and Rao & Paria (109) presented in vitro data showing nano-S could inhibit *A. niger*, *F. oxysporum*, *F. solani*, and *Venturia inaequalis*. Studies designed to compare nano-S to conventional S fungicides in the greenhouse and field are needed.

Although there is much interest in how nano-Ce affects plants (111, 134, 146), only one report has investigated the effect on disease. Adisa et al. (2) found that nano-CeO applied at 250 μg/mL to roots or leaves of tomato inoculated with *F. oxysporum* f. sp. *lycopersici* reduced disease severity by 48% or 46%, respectively, when compared to noninfested plants. Compared to controls, chlorophyll, lycopene, catalase, peroxidase, polyphenol oxidase, fruit production, and total biomass were all increased by foliar and root exposure to 50 μg/mL of nano-Ce. Ce-acetate was less effective than nano-Ce at reducing disease severity, suggesting that the nanoscale size was a prevailing factor in its efficacy. The mechanism underlying nano-Ce facilitated disease resistance is very interesting. Rico et al. (111) demonstrated that an oxidative stress and antioxidant defense system in rice seedlings operated as a function of nano-Ce. In another study, it was shown that the release of Ce ions from a nano-CeO_2 application to roots forms Ce complexes with carboxyl compounds during translocation to the shoots, which may activate defense (146). Clearly, more research is needed to determine how nano-Ce might be employed to optimize potential treatment platforms.

Elmer et al. (36) examined nanoparticles of the metalloid B on watermelon in a single field trial to determine effects on Fusarium wilt. Seedlings that were sprayed twice two weeks apart with 500 μg/mL of metalloid B were grown in experimental field plots artificially infested with *F. oxysporum* f. sp. *niveum* soil or in noninfested soil. When compared to untreated controls, a reduction in disease rankings was observed, but no effect on yield was detected. When soybean seedlings (cv. Spencer) (3 to 4 leaf stage) grown in noninfested soil were sprayed with metalloid B nanoparticles, a 31% increase in fresh weight was observed when compared to controls, grown in noninfested potting soil, whereas a 34% increase was observed when treated seedlings were grown in potting soil infested with *F. virguliforme* (C.D. Perez & W.H. Elmer, unpublished results).

Despite the wealth of information on Mn and plant disease (128), the number of reports that demonstrate the effects of nano-MnO are few. Elmer and colleagues (36, 37) included MnO nanoparticles when assessing the effects of metallic oxides on wilt diseases of eggplants, tomatoes, and watermelon. Although not as effective as nano-Cu in suppressing disease, nano-Mn reduced the AUDPC values on tomatoes inoculated with *F. oxysporum* f. sp. *lycopersici*. Only a marginal effect was noted on suppression of *V. dahliae* on eggplant (37) or *F. oxysporum* f. sp. *niveum* on watermelon. A preliminary field experiment with potted chrysanthemums found nano-Mn (500 μg/mL) sprayed onto rooted cuttings provided season-long protection from Fusarium wilt caused by *F. oxysporum* f. sp. *chrysanthemi* and increased biomass by 24% (W.H. Elmer, unpublished data).

Although the role of Fe_3O_4 super paramagnetic nanoparticles is discussed below, there are a limited number of reports of nano-Fe in disease management. Sharma et al. (119) recently tested very high rates of a biosynthesized formulation of nano-Fe on okra to suppress *M. incognita*. When okra was grown in soil infested with *M. incognita* and treated with aqueous suspensions of 12% nano-Fe, no juveniles were observed and the plants performed better; however, the high rate of nano-Fe used in these studies may prevent its adoption by growers. Foliar application of FeO nanoparticles had no effect on growth or disease suppression when applied foliarly to tomatoes infected with Fusarium wilt (37).

Much information exists on the biosynthesis of nano-Au, but reports on disease suppression are absent. Jayaseelan et al. (59) produced nano-Au in extracts of *Abelmoschus esculentus* and showed the extracts inhibited *Puccinia graminis*, *Aspergillus flavus*, *A. niger*, and *Candida albicans*. Nanoparticles of AlO and NiO were included in the initial screen on Fusarium wilt of tomato by Elmer & White (37) and were found to have no effect. However, more information is needed concerning the use of Al or Ni nanoparticles on disease systems where these elements have previously shown efficacy (120, 142).

Carbon Nanomaterials

Given the allotropic character of carbon, nanomaterials composed of this element can be diverse in shape and function. Many types have been synthesized, but three categories are recognized here: carbon nanotubes (single walled or multiwalled), graphene oxides (oxidized and reduced forms), and fullerenes (C_{60}; buckyballs) (**Figure 1**). The use of carbon nanomaterials in engineering, textiles, paints, medicine, and electronics has been realized for decades. More recently, the antimicrobial activity of carbon nanomaterials has been demonstrated against bacteria (80, 136) and fungi (115, 135), along with positive effects on plant growth (69, 132, 137). A few scattered reports have illustrated the potential of nanocarbon in plant pathology, hinting at the development of an important management tool for disease control.

Carbon nanotubes are engineered from graphene oxide sheets to have a single cylindrical wall or multiple walls at the nanoscale and as noted above, potential antimicrobial activity has been explored. Radial measurements of *A. niger*, *Aspergillus oryzae*, and *F. oxysporum* on agar amended with graphene oxide (reduced form) were decreased significantly as concentration increased. The 50% inhibitory concentrations for *F. oxysporum*, *A. niger*, and *A. oryzae* were 50, 100, and 100 μg/mL, respectively. Wang et al. (135) found that single-wall carbon nanotubes were the most toxic to conidia of *F. graminearum* and *Fusarium poae* followed by multiwalled carbon nanotubes, graphene oxide (oxidized), and graphene oxide (reduced). Fullerenes exhibited no toxicity at concentrations of 62.5 to 500 μg/mL. Hao et al. (49) compared multiwalled carbon nanotubes, fullerene, and graphene oxide (reduced) along with three metallic nanoparticles of CuO, Fe_2O_3, and TiO_2, all at 50 and 200 μg/mL, for their ability to inhibit spores of *B. cinerea* in a detached leaf assay. After 72 h, the lesions were counted; and fullerene and nano-CuO at 50 μg/mL yielded the greatest suppression. Bacteria were equally sensitive to carbon nanomaterials. Graphene oxides at 250 μg/mL killed 95% of the bacterial cells of *Xanthomonas oryzae* pv. *oryzae* compared with 13.3% mortality when using the bactericide bismerthiazol (20). Additional practical applications of carbon nanotubes are beginning to be explored; for example, a novel phytosanitary treatment for *Xylella fastidiosa*–infected pecan is currently being developed using carbon nanotubes in a microwave (51).

Uncovering the mechanisms by which carbon nanomaterials inhibit microbial growth has opened up new areas of research. Many have speculated that the sharp nanoedges of graphene oxides allow direct contact with membranes and cell walls of microbes; here, reactive oxygen-containing functionalities of the graphene oxide could inactivate bacteria and react with chitinous compounds and other polysaccharides on the cell walls of fungi (14, 20, 80, 116, 135). Wang et al. (135) hypothesized that the mechanism for inhibition of *Fusarium* sp. by the single-walled nanotubes was due to an inhibition of water uptake and induction of plasmolysis. However, Berry et al. (14) provided evidence that it was the functionalized carboxylated forms of single-walled carbon nanotubes that activated disruptive oxidative enzymes in the white-rot fungi *Trametes versicolor* and *Phlebia tremellosa.* The purer noncarboxylated forms of nanotubes did not illicit the same level of enzymatic response.

There has been no field application Only a marginal carbon nanomaterials, and further validation of the mechanisms of action on pathogens and implications for nontarget species are required before any registration can be submitted. Furthermore, inhalation exposure to carbon nanotubes poses a real threat and can behave like asbestos (104). Yet the superior antimicrobial activity of carbon nanomaterials and the fact that certain forms can be mass produced at relatively low cost should excite researchers to expand their evaluation of carbon nanomaterials as a management tool and agricultural amendment.

Although a number of studies have attested to the ability of carbon nanomaterial to serve as a delivery vehicle for agents in plants (43, 69, 92), studies in which the payload has been directed to suppress plant diseases are few. However, some creative and noteworthy studies exist and serve as guidance for future developments. As described above, Ocsoy et al. (95) engineered a dsDNA-directed Ag nanoparticle onto graphene oxide to suppress the bacterial disease caused by *X. perforans* on tomatoes in the greenhouse. The graphene oxide decreased *X. perforans* cell viability at 16 μg/mL and enhanced stability, antibacterial activity, and improved adsorption. Sarlak et al. (115) used a different approach and polymerized citric acid onto the surface of oxidized multiwall carbon nanotubes to create water-soluble structures that, in turn, could trap the fungicides zineb and mancozeb. In vitro experiments indicated that the new nanocarbon structure was superior to the fungicide alone in causing more visual distortion on *A. alternata* hyphae when examined microscopically. Admittedly, the utility of carbon nanomaterials as vehicles for transporting material in plants and their pathogens is great given the hollow interior where fungicides, genetic material, and growth-promoting chemicals could be loaded. In time, these functionalized carbon nanotubes will likely have much greater application in plant disease management.

Liposomes

Liposomes are unique molecules composed of a spherical vesicle with at least one phospholipid bilayer. They vary in size, but many fall within the nano range and have an unfilled interior that can be housed with antimicrobial materials. Matouskova et al. (83) used liposomes to encapsulate antimicrobial components such as chitosan and herbal extracts that contained phenolics to inhibit gram-negative bacteria. The liposomes have long-term stability in water, which, in turn, offers a promise of using these products in crop irrigation treatments to suppress diseases. In fact, Pérez-de-Luque et al. (103) found that amphotericin that was bound to liposomes formed nanodisks that were effective in irrigation water at 10 μg/mL and delayed the onset of disease symptoms in chickpea affected by *F. oxysporum* f. sp. *ciceris*. Practical utilization of nanoliposomes has only begun to be examined.

Dendrimers

Dendrimers are branched, tree-like nanoparticles composed of a central core and dominated by attached functional groups (**Figure 1**). Multiple uses are being devised for these materials, but most relate to their efficiency as delivery agents. Their use in human medicine has been realized (21), but few reports exist for implementation in plant pathology. Khairnar et al. (67) showed on human fungal pathogens that drugs attached to dendrimers were twice as effective in increasing the zone of inhibition. Chemical agents and DNA conjugate to the dendrimer and are delivered to the diseased plant tissue via enhanced permeability. Dendrimers may be able to facilitate basipetal transportation of a disease-suppressing chemical to the roots following a foliar application and possibly allow for rescue treatments in established plantings. As dendrimers prove more useful as vehicles for transporting materials to tissues previously unreachable, their utilization in plant pathology will most certainly increase.

FUTURE OF NANOTECHNOLOGY IN PLANT DISEASE DIAGNOSTICS

The success of any sanitation/quarantine practice rests on the detection of infested or infected material before entry into the greenhouse, field, state, or country. Rapid detection of pathogens is essential for this practice to succeed. In this area of diagnostics, nanotechnology offers major advances through faster and more-sensitive pathogen probes. Intuitively, nanoparticles can be used as rapid diagnostic tools in disease detection for bacterial, fungal, nematodal, and viral pathogens (16, 144). Although human medicine has been utilizing nanotechniques for the past decade (40, 57), the number of reports in plant disease diagnostics is relatively few. The following provides a summary of how nanotechnology has enhanced the diagnostics of plant pathogens.

Advancing the use of super paramagnetic iron oxide nanoparticles to aid in pathogen detection has only recently been explored in plant pathology, although the techniques have been pursued in medicine and water purification for more than a decade (76, 127). Magnetic nanoparticles attach to biological tissue and to DNA, subsequently facilitating detection and/or extraction (4). Rispail et al. (112) found that super paramagnetic iron oxide nanoparticles differed from quantum dots after being applied to *F. oxysporum*. Although both nanoparticles attached to the hyphae, the super paramagnetic iron oxide nanoparticles attached only to the surface and were not absorbed where they could be visualized more readily. However, the quantum dots were taken up by the fungal hyphae and became toxic. Gorny et al. (46) developed a rapid and less expensive procedure for extracting DNA for quantification of *Meloidogyne hapla* from mineral soils. Adding the super paramagnetic iron oxide nanoparticles to the extraction lysate that combined a detergent lysis and polyvinylpolypyrrolidone, Gorny et al. (46) maximized DNA yield while minimizing contaminants. Given the broad applications of super paramagnetic iron oxide nanoparticles in detection of human bacteria and cancer cells (42, 143), its utilization in plant diagnostics will grow; one new area would be in rapid detection of pathogens in irrigation water (76).

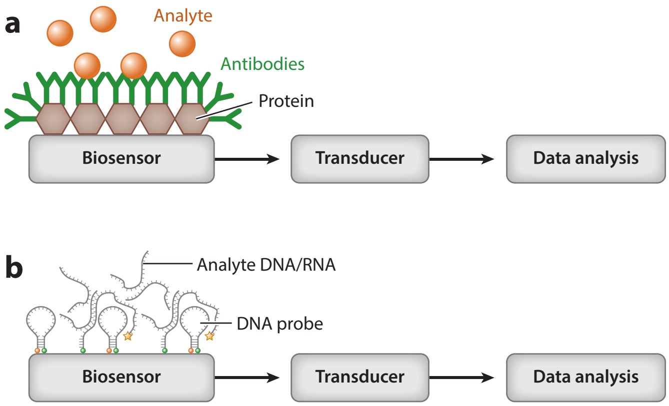

Figure 3

Schematic illustration of (*a*) antibody-based and (*b*) DNA/RNA-based biosensor for analyte (pathogen) detection. The specific combination of analyte and (*a*) immobilized antibody or (*b*) DNA/RNA probe produces a physicochemical change, such as in mass, temperature, optical property, or electrical potential. The change can be translated into a measurable signal for detection. Adapted with permission from Fang & Ramasamy (39).

Biosensors are nanoanalytical devices that use a biological sensing element integrated into a physicochemical transducer to produce an electronic signal when in contact with the analyte of interest (pathogen) (**Figure 3**). The biosensor can be loaded in sufficient quantity so that the electrical signal increases as a function of pathogen density. Thus, a biomolecular interaction is converted into a digital output. The construction of a biosensor is accomplished with metalloid or metal oxide nanoparticles and carbon nanomaterials (carbon nanotubes and graphene). However, more recent breakthroughs in nanotechnology have allowed biosensors to be prepared with different types of nanoparticles and nanostructures with fewer technical hurdles (39). A novel nanobiosensor used nano-Au functionalized with single-stranded oligonucleotides to detect as little as 15 ng of *R. solanacearum* genomic DNA in farm soil (68). During the past decade, several publications have demonstrated the capability of antibody-based biosensors for detection of plant pathogens (58, 79, 102). Nanobased biosensors have been developed for *Cowpea mosaic virus*, *Tobacco mosaic virus*, and *Lettuce mosaic virus* (58, 79). The sensitivity of detection has been increased by two orders of magnitude over traditional ELISA methods. Biosensors were used to detect gram-negative bacteria that use *N*-acyl homoserine lactones in quorum sensing (124). The bacterial biosensors phenotypically respond when exposed to exogenous lactones. Singh and colleagues (121, 122) have engaged in a program to develop a nano-Au-based dipstick for rapid detection of Karnal bunt disease in wheat in the field. Application of this dipstick could have far-reaching uses in enforcing quarantines. In addition to single biosensors directed at one pathogen, research has been ongoing to develop nanochip microarrays that contain multiple fluorescent oligo probes to detect small nucleotide changes in plant-pathogenic bacteria and viruses (81). Quantum dots are small semiconductor nanocrystals that have been used in biosensors to detect phytoplasma in lime trees (106). Using a fluorescence resonance energy transfer mechanism, a high sensitivity and specificity of 100% for approximately 5 *Candidatus* Phytoplasma aurantifolia per μL were achieved with consistent results. Quantum dot biosensors were useful in detecting rhizomania (*Beet necrotic yellow vein virus*) in *Polymyxa betae* (113).

This exciting new approach of using nanoenabled biosensors can be coupled with robotics and GPS systems to create smart delivery systems that detect, map, and treat specific areas in a field prior to or during the onset of symptoms. This technology could reduce agrochemical inputs and increase yield and profits (13, 70). Growers and scouts could perform diagnostics in situ once portable devices with biosensors are developed (64, 93). An extremely valuable use for fast and sensitive biosensors is at ports of entry, where quarantined pathogens could be intercepted with greater efficiency. The value of rapid analysis in detecting food pathogens and mycotoxins is obvious (8, 9, 131).

SUMMARY

Nanotechnology will enable the development of multiple new methods for suppressing disease in the greenhouse and field, enhance disease diagnostics, and create many new tools for molecular manipulations of plants and pathogens. Thus far, nano-Ag, nano-Cu, and nano-Zn make up most reports that pertain to disease suppression. One of the most striking and significant impacts of using nanoparticles in disease management is the vast reduction in metals being applied when compared to the conventional metallic fungicides. Many times, a single application to a young plant was associated with season-long health benefits. Furthermore, the relative unexamined possibility of using carbon nanomaterials in plant disease suppression needs to be further pursued. However, there are limitations that need to be addressed. For example, the health hazards of applying nanomaterials and their fate in the environment and food chain must be answered. A point made by Nair et al. (89) highlighted that many nanomaterials can behave differently on various hosts, which may dictate that each crop/disease system needs separate evaluations. In addition, the vast interactions with other stressors that arise with weather extremes and other pests may compromise or even negate many benefits achieved with nanotechnology. Nevertheless, nanotechnology has rightfully begun to make serious inroads into plant disease management. The use of nanoscale treatment in organic cropping systems is unresolved, but the Organic Material Review Institute allows copper hydroxide, copper oxide, and copper oxychloride to be used in a manner that minimizes accumulation in the soil; as such, the green synthesis of nano-Cu and perhaps other nanoparticles may be considered environmentally acceptable/sustainable and could provide a new weapon for fighting disease in organic systems. It is exciting to see how manipulation of mineral nutrition using nanotechnology can have such a marked effect on growth and yield. Chemists have played and will continue to play a major role in developing new nanocomposites that could deliver multiple nutrients and/or disease-reducing agents. Formulation chemistry will also need to be advanced to overcome the propensity of many of these nanoparticles to hetero/homoaggregate or agglomerate over time into larger particles that would likely lose efficacy.

In addition, the full use of nanotechnology in plant disease diagnostics and plant genetic research has only begun to be realized. Functionalizing metallic and carbon nanomaterials may replace many genetic delivery systems used today as more applications are explored. As new tools using biosensors and quantum dots are being adapted from medical applications and applied to plant pathogens, they may replace conventional assays such as ELISA. However, the struggle to integrate nanotechnology into phytopathology is currently being pursued by only a small number of laboratories, but as new research is directed to discover, adapt, and apply nanotechnology, we feel that the challenges to global food production can be lessened.

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Errata

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