Chapter 9
Applications of Silver Nanoparticles in Plant Protection

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9.1 Introduction

The word "nano" is the one billionth of a meter or 10^{-9}. The term nanotechnology was coined by Professor Norio Taniguchi of Tokyo University of Science in 1974 who illustrated the precise manufacturing of materials at the nanoscale level (Taniguchi 1974). In green nanotechnology, microorganisms and plants are used for the synthesis of nanoparticles (NPs). It is well known that many microorganisms are capable of aggregating inorganic material within or outside the cell to form NPs. However, an enormous number of microbial species are capable of producing metal NPs, and the mechanism of NPs biosynthesis is very important. Microbial synthesis of NPs is an approach that interconnects nanotechnology and microbial biotechnology. Biosynthesis of many metals nanoparticles like gold, silver, gold-silver alloy, selenium, tellurium, platinum, palladium, silica, titania, zirconia, quantum dots, magnetite, and uraninite by bacteria, actinomycetes, fungi, yeasts, algae, and viruses

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has been reported (Narayanan and Sakthivel 2010; Prasad et al. 2016). Silver nanoparticles (AgNPs) have become one of the most commonly used nanomaterials in consumer products, and for several decades, silver (Ag^+) has been studied as an antimicrobial agent against various harmful microorganisms (Prasad 2014).

Due to plant disease agricultural production is reduced worldwide every year; therefore, millions of moneys have been invested in efforts to control the plant diseases. Various natural and artificial methods of control for protection of plants from these diseases have been applied. Among methods for disease control, use of pesticides is the most prevalent. In recent years, environmental hazards caused by excessive use of pesticides have been widely discussed; therefore, researchers in the agricultural field are searching for alternative measures against pesticides. Nanotechnological applicability in crop disease protection offers a great promise in the management of insects and pathogens. AgNPs are very effective against phytopathogens with low toxicity and lead to broad range of applicability in pesticidal activity. It is efficiently used for site-targeted delivery of important agrochemical products and for diagnosis purpose tools in case of prior detection of plant diseases (Chowdappa and Shivakumar 2013). AgNPs are the most studied and utilized NPs in the field of agricultural research to improve the efficiency, yield, and sustainability of agricultural crops. It has long been known to have strong pesticidal, antifungal, antiviral, and bactericidal effects (Chen and Schluesener 2008). Due to its broad spectrum of antimicrobial activities, AgNPs have the prospect to increase food quality, global food production, plant protection, detection and regulation of plant diseases, monitoring of plant growth, and pest control for "sustainable agricultural development" (Kim et al. 2012; Khan and Rizvi 2014; Prasad et al. 2017a, 2017b). AgNPs are highly stable and very well dispersive in aqueous solution. It is being used as foliar spray to inhibit the growth of fungi, molds, rot, and several other plant diseases (Singh et al. 2015). Moreover, AgNPs are also used as an excellent plant-growth stimulator. It provides novel tool for the management of diseases, rapid disease detection, and minimizing nutrient losses in fertilization through an optimized nutrient management (Pérez-de-Luque and Rubiales 2009). As an alternative to chemically manufactured pesticides, use of AgNPs as antimicrobial agents has become more common as technological advances make their production more economical. One of the important appliances of AgNPs is in the management of plant diseases. Silver displays multiple modes of inhibitory action against microorganisms; therefore, it may be used with relative safety for control of various plant pathogens, compared to synthetic fungicides (Aziz et al. 2016; Prasad et al. 2014, 2017a, 2017b).

Artificial chemical antimicrobials are widely used in modern agriculture to control plant diseases. Environmental hazards caused by excessive use of pesticides pose health problems as modern society is becoming more health-conscious. Therefore, agricultural scientists are searching for alternative eco-friendly and less capital-intensive approaches to control plant diseases. As an alternative to chemically manufactured pesticides, use of AgNPs as antimicrobial agents has become more common as technological advances make their production more economical. So, the focus of this chapter is to study the possibilities of using the synthesized AgNPs in plant protection (Fig. 9.1).

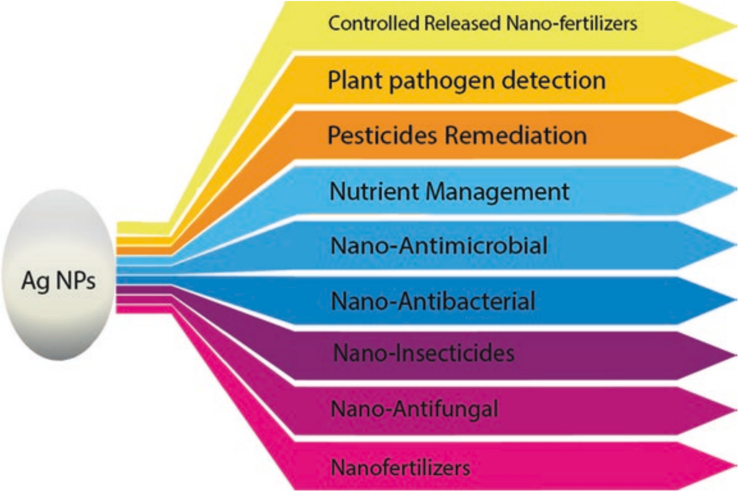

Fig. 9.1 Silver nanoparticle application in plant protections

9.2 Chemical and Biological Silver Nanoparticles (AgNPs)

The physical and chemical methods are numerous in number for the synthesis of AgNPs, and many of these methods are expensive or use toxic substances which are major factors that make them "not so favored" methods of synthesis. Various types of physical and chemical methods that are employed in the production of nanoparticles are top-down method, bottom-up method, reduction of silver metal salt, electroreduction of $AgNO_3$ in aqueous solution in the presence of polyethylene glycol, son decomposition, photoreduction in reverse micelle, and many more. The quest for such a method has led to the need for biomimetic production of silver nanoparticles whereby biological methods (using plants and microbes) are used to synthesize AgNPs. Biologically, various types of microbes and plants species are used for the biosynthesis of nanoparticles. In utmost cases, the chemical synthesis methods lead to some chemically toxic materials being absorbed on the surface and can hinder their practice in medical applications (Parashar et al. 2009; Swamy and Prasad 2012; Prasad and Swamy 2013; Prasad et al. 2016). Biological synthesis of AgNPs is a bottom-up method that typically involves reduction/oxidation reactions. The microbial enzymes or the plant phytochemicals with antioxidant or reducing properties act on the respective compounds and give the desired nanoparticles. The three major components involved in the biological synthesis of nanoparticles: solvent medium, the eco-friendly reducing agent, and a nontoxic stabilizing agent. The nanoparticles so produced have protein cap over it imparted by biological host. This capping helps in easy entry in pathogenic fungi. Presence of peak at 1654 in FTIR spectrum indicates the presence of amide bond in the sample indicating the presence of protein capping which is responsible for stabilizing the synthesized nanoparticles (Aziz et al. 2015).

9.3 Mechanism by Which Pathogens Cause Diseases

Pathogens cause infection via various mechanisms. Some common known mechanisms are:

1. Cell wall degradation: some pathogens have enzymes which can degrade the cell walls of plant leading to the easy access to the host plant.
2. Toxins: these chemicals are further categorized into host specific, which are specific for few plants, and non-host specific, which are active against all plants.
3. Effector proteins: these proteins interfere with the chemical signaling pathways of the host plant which results in reduction of phytochemical production (Winbo 2011).

9.4 Mechanistic Approach of antimicrobial activity of Silver Nanoparticles and Controls of the Growth of Pathogens

There are numerous mechanisms by which AgNPs control the growth of pathogens. The exact mechanism of AgNPs by which it causes antimicrobial effect is not evidently known and is one of the debated topics. Numerous theories on which action of AgNPs based by which it cause antimicrobial effects. Silver nanoparticles possess the ability to anchor the bacterial cell wall and penetrate it, causing structural changes in the cell membrane like cell membrane permeability and cell death. There are formation of “pits” and accumulation of the nanoparticles on the cell surface. The formation of free radicals by the AgNPs may be considered as another mechanism by which the cells die. The electron spin resonance spectroscopy studies suggest that there is a formation of free radicals when AgNPs contact the bacteria and these free radicals make membrane porous which ultimately leads to cell death (Danilcauk et al. 2006; Kim et al. 2007). It has also been proposed that there may be a release of silver ions by the nanoparticles (Feng et al. 2008) and these ions have inbuilt property to interact with the thiol (-SH) groups of many crucial enzymes and inactivate them (Matsumura et al. 2003). The bacterial cells come in contact with silver ions, which inhibit many functions and damage the cells which result in the generation of reactive oxygen species (ROS) that may also be produced possibly through the inhibition of a respiratory enzyme by silver ions and attack the cell itself. The action of these nanoparticles on the cell can cause the reaction to take place and subsequently lead to cell death. Another important fact is that the DNA contains sulfur and phosphorus as their major components; the nanoparticles can act on these components and destroy the DNA which ultimately lead to cell death (Hatchett and Henry 1996). The interaction of the silver nanoparticles with the sulfur and phosphorus of the DNA can lead to problems in the DNA replication of the bacteria and thus terminates their growth. It has also been found that the AgNPs can modulate signal transduction in bacteria. It is a well-known fact that phosphorylation of protein substrates in bacteria persuade bacterial signal

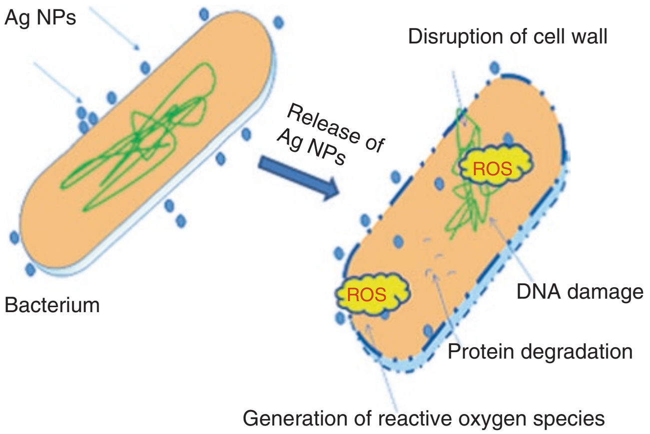

Fig. 9.2 Mechanistic approach of the antibacterial action indicating ROS generation induced by AgNPs. (Reprinted from Aziz et al. 2015)

transduction. Dephosphorylation is only reported in the tyrosine residues of gram-negative bacteria. The phosphor tyrosine profile of bacterial peptides is altered by the nanoparticles. It was found that the nanoparticles dephosphorylate the peptide substrates on tyrosine residues, which leads to signal transduction inhibition and thus the stoppage of growth. It is however necessary to understand that further research is required on the topic to thoroughly establish the claims (Shrivastava et al. 2007) (Fig. 9.2).

9.5 Effect of Silver Nanoparticles on Phytopathogens

Phytopathogens, viz., bacteria, fungi, viruses and nematodes, are key limiting factors in the production of food material. Several methods are used to control pathogens but not a perfect method to control of the disease. Hence, a great prospect exists for the manipulation of nanotechnology for the management of plant pathogens. Silver is considered the most capable nanomaterials with fungicidal, bactericidal, and viricidal properties owing to its wide-ranging effectiveness, low toxicity, ease of use, charge capacity, high surface-to-volume ratios, crystallographic structure, and adaptability to several substrates (Nangmenyi and Economy 2009). AgNPs act as robust antimicrobial agent due to strong inhibitory effects against various microorganisms (Clement and Jarrett 1994). Nanosilver exhibits high level of toxicity to the microorganisms and lower toxicity to the mammalian cells. It was observed that the microbe-killing effects of AgNPs were size dependent (Raza et al. 2017). The AgNPs/PVP were tested for fungicidal activity against different yeasts and molds such as *Candida albicans*, *C. krusei*, *C. tropicalis*, *C. glabrata*, and *Aspergillus brasiliensis*. The hybrid materials showed strong antifungal effects against the tested microbes (Bryaskova et al. 2011). Traditional microbiological plating, scanning electron microscopy, and Raman spectroscopy were used to study antifungal activities of AgNPs and to characterize the changes in morphology and cellular compositions of fungal hyphae. Aziz et al. (2016) observed the effect of biogenic nanoparticles from AgNPs on pathogenic fungi, *Candida albicans*, *Fusarium oxysporum*, and *Aspergillus flavus*, and these antimicrobial attributes were comparable to those of established fungicides (amphotericin B, fluconazole, and ketoconazole). Importantly, these nanoparticles show significant synergistic characteristics when combined with the antibiotics and fungicides to offer substantially greater resistance to microbial growth. Ocsoy et al. (2013) developed nanocomposite DNA-directed AgNPs grown on graphene oxide (Ag@dsDNA@GO). These composites effectively decrease *Xanthomonas perforans* cell viability in culture and on plants. At the very low concentration (16 ppm), composites show excellent antibacterial ability with significant benefits in improved stability and higher antibacterial activity. Also, in most cases, inhibition increased as the concentration of AgNPs increased. This could be due to the high density at which the solution was able to saturate and cohere to fungal hyphae and to deactivate plant pathogenic fungi (Kim et al. 2012). Synthesis of nanoparticles chemically requires chemical substances that are toxic in nature. Even after purification, there is a chance of chemical contamination which leads to unsafe use of nanoparticles. Chemical and physical methods need expensive chemicals and instruments, which leads to hike in production cost. However, biologically synthesized nanoparticles don’t undergo any toxic and expensive procedure. Laboratory test reveals that biologically synthesized nanoparticles are safest to use and these nanoparticles possess protein caps which allow easy excess to the pathogen cell membrane.

Nano Silver is one of the known strong bacteriostats and possesses broad-spectrum antimicrobial activity. It has been reported that well-dispersed nanosilver colloid is more adhesive to bacteria and fungi leading to enhanced antimicrobial activity (Kim et al. 2008). In nature many agricultural crops and forestry are attacked by many microorganisms resulting in loss of agricultural product and death of tree species. AgNPs came up as a new hope and control disease mechanism by damaging fungal hyphae, interference with nutrient absorption, and enhanced inhibition of fungal growth and germination. The mechanism involved may be the influence of silver ions and nanoparticles on spore formation and disease progression in plant pathogenic fungi. Hence, AgNPs prove to have high potential to be used as nanopesticides for controlling phytopathogens (Alghuthaymi et al. 2015).

9.5.1 Nano-antibacterial

Silver nanoparticles in agricultural soil affect several bacterial communities which are beneficial/harmful for plant and environment (Panyala et al. 2008). AgNPs act as strong antimicrobial agent due to strong inhibitory effects against various bacterial species (Clement and Jarrett 1994; Joshi et al. 2018). Kamran et al. (2011) reported that the nanosilver and nano-TiO_2 with a good potential may be used for removing the bacterial contaminants in the tobacco plant. AgNP exposure causes toxicity to bacteria, and treatment can prevent replication and protein synthesis (Chaloupka et al. 2010). Notably, the most common application problem involves the agglomeration and diffusion of these nanoparticles, which reduce antibacterial activity. These studies revealed that used various organic (Jo et al. 2009) and inorganic substances (Lamsal et al. 2011) as well as powerful carriers (Ouda 2014) to stabilize AgNPs. These substances can strongly influence the antibacterial activity and reduce the biological toxicity of nanoparticles. Also a synergistic antimicrobial effect is achieved when AgNPs are hybrid with other metal nanoparticles or oxides acting as a shell or a core to form bimetallic nanoparticles (Chou and Chen 2007). Chen et al. (2016) study revealed that bacteriostatic and bactericidal activity of the pure and surfactant-stabilized AgNPs (SDS-Tween 80-CTAB-PVP) capped silver nanoparticles. *Ralstonia solanacearum* (phytopathogenic fungi), which causes severe bacterial wilt in tobacco, is used to investigate the bacteriostatic and bactericidal activity of pure and surfactant-stabilized AgNPs. The surfactants affected the antibacterial activity of AgNPs toward *R. solanacearum* to different extents.

9.5.1.1 AgNP Antibacterial Mechanism

The accumulation of AgNPs in the cellular membrane led to an increase of its permeability and eventually to the death of bacterial cells. Also, they attempted to understand their mechanism of action. Presently, there are five main explanations that have been proposed to describe the antibacterial activity (Lemire et al. 2013):

1. Release of toxic ions that bind to sulfur-containing proteins – this accumulation avoids the proper functioning of proteins in the membrane and interfere in cell permeability (Sondi and Salopek-Sondi 2004).
2. They may be genotoxic – toxic ions can DNA destruction which leads to death of cell.
3. Interruption of electron transport chain, protein oxidation system, and collapse of membrane potential.
4. Generation of reactive oxygen species (ROS)-mediated cellular damage and different metal-catalyzed oxidation reactions might underlie specific types of DNA, protein, and membrane damage (Banerjee et al. 2010; Zeng et al. 2007; Aziz et al. 2015).
5. Interruption with uptake of nutrients (Pal et al. 2007).

These mechanisms might not operate separately which suggests that more than one mechanism occur simultaneously. These multiple targets of action might access NPs to fight effectively against different plant pathogens.

9.5.2 Nano-antiviral

For AgNP role against plant viruses, there are few reports documented. For instance, the effective control of bean yellow mosaic virus (BYMV), genus *Potyvirus*, family *Potyviridae*, would be of high interest for many African countries, which can suffer significant yield reductions in fava bean crops upon viral infection leading to considerable economic losses (Radwan et al. 2008). It has been suggested that AgNPs inhibit viral nucleic acid replication, while their antiviral activity depends on the particle size, as well as on the distribution of interacting ligand/receptor molecules (Lü et al. 2009; Papp et al. 2010). Elbeshehy et al. (2015) studied the effect of biosynthesized AgNPs on leaves of fava bean infected with BYMV which showed severe symptoms, including yellow mosaic, mottling, crinkling, size reduction, and deformation, symptoms that were absent from the non-infected leaves.

9.5.3 Nano-antifungal

The antifungal effect of AgNPs has established only insignificant attention and with very few publications (Roe et al. 2008; Kim et al. 2008). There are few studies available dealing precisely with their mechanism of action against clinical isolates and American type culture collection strains of *Candida* spp. and *Trichophyton mentagrophytes* (Li et al. 2012; Panáček et al. 2006; Min et al. 2009). The use of AgNPs as antimicrobial agents becomes more widespread as technological advances make their production more economical. Control of phytopathogens is one of the probable applications in which silver can be utilized in the management of plant diseases. Since silver displays a collection of modes of inhibitory action to plant pathogens (Park et al. 2006), it might be used as controlling agent for various plant pathogens in a moderately safer way as compared to synthetic fungicides. Ag-SiO NPs have a strong antifungal effect against *Botrytis cinerea* (Oh et al. 2006). The combined effect of fluconazole and AgNPs for their antifungal activity was evaluated by Gajbhiye et al. (2009) against *Phoma glomerata*, *Phoma herbarum*, *F. semitectum*, *Trichoderma* sp., and *Candida albicans* by disc diffusion technique. Ag_2S nanocrystals on amorphous silica particles show antifungal activity against *Aspergillus niger* (Fateixa et al. 2009). The phytogenic AgNPs were tested against three different plant pathogenic fungi such as *Rhizoctonia solani*, *Fusarium oxysporum*, and *Curvularia* sp. and showed antifungal activity against *R. solani* followed by *F. oxysporum* and *Curvularia* sp. (Balashanmugam et al. 2016). Amphotericin B showed moderate antifungal activity against the three plants pathogenic fungi (Fig. 9.3).

9.6 Nano-insecticides

Advance investigation highlighted the extensive application of AgNPs for insecticidal application to kill the mosquitoes and fleas. AgNPs possessed excellent antilice and mosquito larvicidal activity, having dynamic application in community

Fig. 9.3 Antifungal activity of phytosynthesized AgNPs against different plant pathogens on the third day (I) *Rhizoctonia solani*, (II) *Fusarium oxysporum*, (III) *Curvularia* sp. D-silver nitrate, S-phytosynthesized AgNPs, A-amphotericin-B, P-*C. roxburghii* aqueous leaf extract. (Reprinted from Balashanmugam et al. 2016)

health improvement. There are tremendous researches investigating the efficacies of biogenic synthesized AgNPs as mosquito larvicidal agent against different species of mosquitoes, i.e., *Culex quinquefasciatus*, *Heteroscodra maculata*, *Rhipicephalus microplus*, and *Anopheles subpictus*, and suggesting that it can be used as an ideal eco-friendly approach for their control (Marimuthu et al. 2011; Suman et al. 2013; Mondal et al. 2014). Jayaseelan et al. (2011) documented on the pediculicidal and larvicidal activity of synthesized AgNPs (from leaf extract of *Tinospora cordifolia*) against the head louse *Pediculus humanus* and larvae of *Anopheles subpictus* and *Culex quinquefasciatus* and showed maximum mortality. Rouhani et al. (2012) evaluated the insecticidal activity of AgNPs against the *Aphis nerii*. Soni and Prakash (2015) have described the larvicidal and pupicidal properties of biologically produced AgNPs (from fungal strain of *Aspergillus niger*) against the mosquito larvae of *Aedes aegypti*, *Culex quinquefasciatus*, and *Anopheles stephensi*.

9.7 Controlled Released Nanofertilizers

Silver nanoparticles are very stable and biodegradable and it also displays slow release of agrochemicals. So, it can be used for formation of nanocapsules for slow and optimized delivery of agrochemicals, pesticides, and fertilizers in agricultural practices (Chowdappa and Shivakumar 2013). Nanoencapsulated agrochemicals are designed to possess the desired properties including effective optimum concentration, time-controlled release, enhanced activity on target site, and least toxic effects (Tsuji 2001). It helps in slow release of agrochemical in controlled way to the particular host through dissolution, biodegradation, diffusion, and osmotic pressure with specific pH. Nanotagged agrochemicals reduce the damage to nontarget plant tissues and reduce risk of nonspecific chemical contamination in the surrounding environment (González-Melendi et al. 2008; Rai and Ingle 2012). Combinations of inorganic fertilizer mainly supply three nutrients, nitrogen (N), potassium (K), and phosphorus (P), to various crops at different growing conditions. This brings out the idea of developing encapsulated fertilizers, in which NPK fertilizers are entrapped within nanosilver to boost nutrient management.

9.8 Positive Effect on Plant Growth

Some nanoparticles have a beneficial effect on some plant species manifested by enhancing seed germination, enhancing crop yield, or suppressing plant disease (Servin et al. 2015; Arruda et al. 2015). As we know that NPs have both positive and negative effects on plant growth and development. Recently, Krishnaraj et al. (2012) studied the effect of biogenic AgNPs on hydroponically grown *Bacopa monnieri* growth metabolism, showed a significant effect on seed germination, and induced the synthesis of protein and carbohydrate and decreased the total phenol contents and catalase and peroxidase activities. Also, Savithramma et al. (2012) revealed that biologically synthesized AgNPs enhanced seed germination and seedling growth of trees *Boswellia ovalifoliolata*. AgNPs increased plants morphological (shoot and root length, leaf area) and biochemical attributes (chlorophyll, carbohydrate and protein contents, antioxidant enzymes) of *Brassica juncea*, common bean, and corn (Salama 2012; Sharma et al. 2012). However, Gruyer et al. (2013) reported AgNPs have both positive and negative effect on root elongation depending on the plant species. They reported that root length was increased in barley but was inhibited in lettuce. Also, Yin et al. (2012) studied the effects of AgNPs on germination of 11 wetland plants species (*Lolium multiflorum*, *Panicum virgatum*, *Carex lurida*, *C. scoparia*, *C. vulpinoidea*, *C. crinita*, *Eupatorium fistulosum*, *Phytolacca americana*, *Scirpus cyperinus*, *Lobelia cardinalis*, *Juncus effusus*) and found AgNPs enhanced the germination rate of 1 species (*E. fistulosum*). AgNPs induce root growth by blocking ethylene signaling in *Crocus sativus* (Rezvani et al. 2012).

Silver nanoparticles may have both a positive and a negative impact on plants, depending on size, concentration, chemical composition, zeta potential, stability, and the shape of nanoparticles (Mirzajani et al. 2013; Tripathi et al. 2015, 2017; Costa and Sharma 2016). Several studies have depicted a negative impact of nanoparticles on plants in the form of decrease in plant growth, productivity, and pigments (Tripathi et al. 2017). On the other hand, robust and smart engineered nanoparticles are also explored for the betterment of agricultural crop production, as growth stimulators, nanopesticides, nanofertilizers, soil-improving agents, or sensors for monitoring different agricultural parameters in the field (Fraceto et al. 2016; Prasad et al. 2016, 2017a). Due to the increased interest in the area, most of the research depicting the influence of industrial nanoparticles on plants has been performed in recent years. Recent studies have exposed that when AgNP was combined with different treatment/compounds, it may have a different impact on plants (Berahmand et al. 2012; Belava et al. 2017), due to the influence of other phenomena/compound on AgNPs. AgNP treatment in combination with magnetic field was observed to improve quantitative yields in *Zea mays* (Berahmand et al. 2012). In the wheat-pathogen phytosystem, an increase of lipid peroxidation was observed, when compared with NP or pathogen alone (Belava et al. 2017). The biosynthesized nanoparticles induced the protein and carbohydrate synthesis and decreased the total phenol contents, which can be considered as a positive effect; it may be due to the presence of altered size (2–50 nm) of nanoparticle or the different chemical property of biogenic NPs. The size of AgNPs of 200–800 nm was observed to enhance the plant growth (Jasim et al. 2016) whereas 35–40 nm observed to positively influence the root and shoot growth of different plant (Pallavi et al. 2016), which may be due to the inability of the penetration of large nanoparticles (Mirzajani et al. 2013). AgNPs (of comparatively small size, i.e., <30 nm) when applied in high concentration were observed to inhibit the root and shoot growth in different plants studied (Dimkpa et al. 2013; Vinković et al. 2017). Treatment of *Lupinus termis* seedlings with 100 ppm bio-AgNPs might improve the growth profile, while exposure of seedlings to high concentrations (300 and 500 ppm) resulted in a highly significant reduction in all growth parameters and growth indices (Al-Huqail et al. 2018) (Fig. 9.4 and Table 9.1).

9.9 Pesticide Remediation

As an alternative, AgNPs can be applied for the degradation of pesticides to overcome these problems. In modern nanotechnological research, applicability of AgNPs in pesticide mineralization is well reported. Nair and Pradeep (2003) confirmed the halocarbon mineralization and catalytic destruction by means of silver and gold nanoparticles. Manimegalai et al. (2011) reported the applicability of AgNPs for the removal of pesticides “chlorpyrifos” and “malathion” from water. These nanoparticles have been shown to completely remove the pesticides as it actively anchored the pesticide to its inert surfaces.

9.10 Plant Pathogen Detection

It is worth mentioning here that newly developed smart nanomaterials with special nanoscale characteristics offer tremendous breakthrough in plant pathogen detection and diagnosis technology (Khiyami et al. 2014). Striping voltammetry as an electrochemical technique can be applied to detect the metal nanoparticles directly making the assay simple to perform. Gold and silver nanoparticles can be used in these methods including different inorganic nanocrystals (ZnS, PbS and CdS) for analytic detection (Upadhyayula 2012). Schwenkbier et al. (2015) developed a helicase-dependent isothermal amplification in combination with on-chip hybridization for the detection of *Phytophthora* species. This approach allows efficient amplification of the yeast GTP-binding protein (Ypt1) target gene region at one constant temperature in a miniaturized heating device. The assay’s specificity was

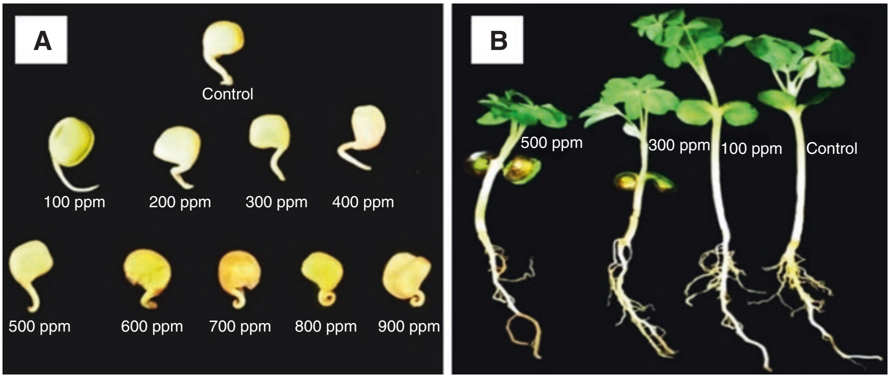

Fig. 9.4 (**a**) Effect of different concentrations of CSL-AgNPs (0–900 ppm) on *Lupinus termis* L. seed germination. (**b**) Effect of different concentrations of CSL-AgNPs (0, 100, 300 and 500 ppm) on growth parameters of *Lupinus termis* L. (Reprinted from Al-Huqail et al. 2018)

determined by on-chip DNA hybridization and subsequent AgNP deposition. The silver deposits serve as stable endpoint signals that enable the visual as well as the electrical readout. These advancements point to the direction of a near future on-site application of the combined techniques for a reliable detection of several kinds of plant pathogens.

9.11 Conclusion

Silver has been constantly superb antimicrobial (antibacterial and antifungal) and has been used for the purpose for ages. The unique physicochemical properties of AgNPs only increase the efficacy of silver. Chemical and physical methods of AgNP synthesis were being followed by several periods, but they are expensive, and the use of several toxic chemicals for their synthesis makes the biological synthesis the more desired possibility. Though microbial and plant extract sources can be used for AgNP synthesis, the easy availability, the nontoxic nature, the various options available, and the advantage of quicker synthesis make plant extracts the best and an excellent choice for biogenic AgNP synthesis. The uses of AgNPs are varied and many, but the most exploited and desired aspect is their antimicrobial and anti-inflammatory activities. The disadvantage of AgNPs is that they can induce toxicity at various degrees. It is recommended that higher concentrations of AgNPs are toxic and can cause innumerable health problems. It also revealed that the nanoparticles of silver can induce various ecological problems and disturb the ecosystem if released into the environment. Hence, this chapter concludes the application of silver nanoparticles in plant disease management, nanofertilizers, nanopesticides, pesticide remediation, and plant pathogen detections; with that there would be mechanisms devised to nullify any toxicity caused by nanosilver to humans and the environment so that the unique properties of this substance can be put to great use for human betterment without any controversies.

Table 9.1 Positive effect of AgNP on plant species

Size (diameter in nm)	Concentration	Exposure methodology	Plant studied	Impact	References
20	40 gha^{-1}	Field, through irrigation water, (nanoparticle applied with 10 mT magnetic field)	*Zea mays*	Combination of silver nanoparticles and magnetic field led to improved quantitative yields of fodder maize	Berahmand et al. (2012)
20 (polyvinylpyrrolidine-coated, PVP-NP) 6 (gum Arabic coated, GA-NP)	1, 10, 40 mg/L (toxic study performed with 40 mg/L in pure culture experiment)	Petri plates (treatment on seeds)	Eleven species of common wetland plants	PVP-NP significantly increases leave length in *Scirpus cyperinus* and *Carex lurida* whereas decreases in *Lolium multiflorum*. GA-NP shows a significant decrease in leave length except *Phytolacca americana* Root growth was observed to be positively affected by PVP-NP in *Phytolacca americana*, *Panicum virgatum*, and *Carex lurida*, whereas six other species have been observed to have negative effect of PVP-NP	Yin et al. (2012)
200–800	1 mg/L	Growth medium with Agar + pots with soil (treatment on germinated seeds)	*Trigonella foenum-graecum*	Enhancement in plant growth and diosgenin synthesis was observed	Jasim et al. (2016)

(continued)

Table 9.1 (continued)

Size (diameter in nm)	Concentration	Exposure methodology	Plant studied	Impact	References
35–40	50, 75 mg/L	Pots (foliar treatment on grown plant)	*Triticum aestivum*, *Vigna sinensis*, *Brassica juncea*	Relatively unaffected (wheat) The optimum growth promotion and increased root nodulation were observed at 50 ppm treatment (cowpea) Improved shoot parameters were recorded at 75 ppm (*Brassica*)	Pallavi et al. (2016)
12.9 ± 9.1 (90%) nanoparticles in ultrapure water	0.01, 0.05, 0.1, 0.5, 1 mg/L	Pots with soil (treatment on seedling)	*Capsicum annuum*	Concentration-dependent decrease in plant growth Concentration-dependent increase in cytokinin concentration	Vinković et al. (2017)
<100	1.5 mg/L	Hydroponic and pots (treatment on seeds)	*Triticum aestivum* (wheat-*Pseudocercosporella herpotrichoides* phytosystem)	In Myronivska 808 the lipid peroxidation was observed to be significantly high where nanoparticle was present with pathogen	Belava et al. (2017)

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