7 Nanotechnology Mediated Detection and Control of Phytopathogens

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Abstract

In current environmental scenario, about 20–40% of crops are destroyed by pests and pathogens annually. Hence to control the plant pathogens, toxic pesticides are generally used which are harmful to both environment and human beings. In this context, nanotechnology provides harmless effect to pesticides as it reduces toxicity, increases solubility of less water-soluble pesticides and increases shelf-life. It also provides good impact on environment and soil. Nanoparticles are small particles which have size in between 1 and 100 nm. This chapter intends to discuss how nanoparticles can be used for the control of plant diseases either nanoparticle alone or acting as protestants or nanocarriers for insecticides, pesticides and fungicides. Nanoparticles which are synthesized by different methods can be used for agricultural applications. Nowadays although nanotechnology is progressing quickly, however its application in agricultural fields is insignificant to control pests and pathogens practically. Hence, agricultural applications can be developed by understanding the fundamental things of research and production of commercial nanoproducts to control plant disease.

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7.1 Introduction

Nowadays nanotechnology has become the new attraction point of research for most of the scientists. On the evening of ninth December 1959, an article entitled "There's plenty of room at the bottom" was delivered to American Physical Society by Prof. Richard Feynman (Feynman 1960). According to him, single atoms or molecules could be controlled, so that different possibilities of formulating new material could be generated, which lead the scientific community to discover the new word of "Nanotechnology". Later the term "Nanotechnology" was coined by Norio Taniguchi, Professor of Tokyo University of Science, Japan in 1974 to describe the superfine, refined particles (Bhattacharyya et al. 2009). Hence, the research or study related to the materials in the nanoscale is known as nanotechnology. The basic and fundamental unit of nanotechnology is nanoparticles. The particles whose size lies between 1 and 100 nm are called nanoparticles and mainly composed of carbon, metal, metal oxides and organic matter (Ealias and Saravanakumar 2017). Currently, nanotechnology is an exciting field for researchers as nanoparticles act as bridge between the physico-chemical gap between the atoms/molecules and bulk (macroscopic) material (Thakkar et al. 2010). This dissimilarity is due to the small size and high surface area to volume ratio of nanoparticles (Thakkar et al. 2010). So nanoparticles are used in various fields of science and technology due to these unique features. Hence researchers now have a keen interest in the synthesis of the nanoparticles using various techniques.

7.2 Synthesis of Nanoparticles

In the last decade, extensive research has resulted in enormous progress in the field of nanotechnology and also in synthesis of nanoparticles. Various methods were established to synthesize nanoparticles based on their physical, chemical, optical and mechanical properties (Cho et al. 2013). One of the most popular synthesis methods, widely known as the "top-down approach", was developed and established by Taniguchi et al. (Tarafdar and Adhikari 2015). The overall concept of this method is relatively simple and relies on the fact that most nanoparticles can be synthesized from larger molecules and later through a series of reactions can be converted into nano form (Abou El-Nour et al. 2010). Interestingly, almost 10 years after the introduction of the "top-down approach" to synthesize nanoparticles, a new concept was put forward by K. Eric Drexler termed as "bottom- up approach" (Drexled 1986). This concept envisages that nanoparticles can be synthesized from macromolecules based on the atomic and molecular composition. Most nanoparticles are synthesized by either physical and/or chemical methods. However, biological methods for synthesis of nanoparticles have gained more attention of the research community, due to its eco-friendly nature. In general eco-friendly strategy employs plant extract, bacteria, algae, fungi, etc. to synthesize different nanomaterials (Mittal et al. 2013). Furthermore, the synthesis of nanoparticles by these processes have some benefits over the chemical synthesis method, as the nanoparticles generated in such process are nontoxic by nature (Charitidis et al. 2014).

The ecological cycle is tightly regulated, with plants being placed as the primary producers to sustain the balance in the food chain. However, in the course of evolution, infections caused by plant pathogens have emerged as new players, threatening the genetic diversity and survival of plants. Plant pathogens and pests have created new issues and challenges in agriculture, resulting in decreased crop production. It is estimated that these pests and pathogens contribute to an overall 22–40% less production in crops per year globally (Worrall et al. 2018). Hence, pest management is utmost important. In layman's term pest management means use of pesticides such as insecticides, fungicides, herbicides, etc. to kill and destroy pest and increase crop yield. The use of pesticides although results in an overall increase in crop production, but comes with a price. For example, the advantages of the use of pesticides are (1) quick action for killing (2) lower in cost. On the contrary, the harmful effects of pesticides are (1) effect on the non-target organism, (2) increase in the evolution of resistant pest population, (3) adverse effect on health, (4) reduction in soil bio-diversity, (5) decrease in nitrogen fixation, etc. (Hayles et al. 2017). It is reported that 90% of pesticides are blown to the air during or after application, which causes adverse health issues (Stephenson 2003; Ghormade et al. 2011). Hence, various research groups are working to find out an alternative for the use of pesticides that could potentially be less harmful to the environment and precisely affect only the target pests in interest and enrich soil productivity.

In such a scenario, advancements in nanotechnology have emerged as a possible new strategy to overcome the traditional problems encountered in the agricultural field (Bramlett et al. 2019). Recently, nanotechnology has been implemented in plant hormone delivery, water management, seed germination, transfer of target genes and nano-sensors (Hayles et al. 2017). For the development of the new generation of pesticide, scientists are employing nanoparticles with desired size, shape and surface properties to provide better pest management (Khandelwal et al. 2016) (Fig. 7.1).

7.3 Early Detection of Phytopathogens Using Nanoparticles

Using conventional agricultural practices, it was daunting for phytopathologists to identify unknown phytopathogens causing different plant diseases. However, with the recent advancements in nanotechnology, phytopathologists can employ new strategies to detect the plant disease to increase better crop management (Rai and Ingle 2012). In this context, molecular and immunodiagnostic techniques in

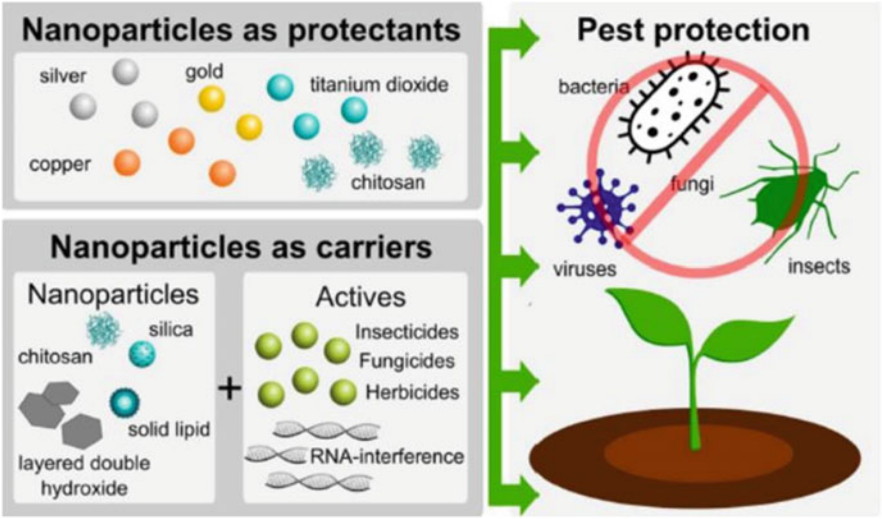

Fig. 7.1 Schematic diagram representing nanomaterial as either protectant or carrier for fungicide, insecticide or herbicide. Adapted with permission from Ref (Worrall et al. 2018)

conjunction with nanotechnology are used to characterize the pathogens for identification and detection of microorganisms. Polymerase Chain Reaction (PCR) is used for the detection of causative agent *Xanthomonas axonopodis pv*. *Punicae* in pomegranate (Mondal et al. 2012). ELISA test is used to detect *Xylella fastidiosa* (Sherald and Lei 1991). Advanced molecular techniques such as q-PCR is used to detect blight bacterial disease in pomegranate. These techniques have high sensitivity, reliability and specificity. Early detection of pathogens is the best method to prevent disease in plants. Phytosanitary analysis and plant quarantine etc routine surveys are performed to control the disease as these are reliable, fast and affordable process. Additionally, the lab-on-a-chip system is used for the detection of toxicity of water, nutrients in the water and control the quality of production of food (Gardeniers and Van den Berg 2004).

7.3.1 Action of Nanoparticles against Phytopathogens

7.3.1.1 Plant Disease Cycle

The process through which the development of disease occurs in plants is called the disease cycle. The events that are involved in disease development are inoculation, penetration, infection, incubation, reproduction and survival (De Wolf and Isard 2007). Pathogens are introduced into different plants by different methods. For example, some fungal pathogens release their spores into the air and these spores are spread by the air current and penetrate through the injury or wound and natural opening site of plants such as stomata and hydathodes during different environmental conditions such as moisture and temperature. When these pathogens enter into the plant tissue, they establish a parasitic relationship between pathogens and plants. Then pathogens undergo incubation or dormant period till the disease initiates (Agrahari et al. 2020). Plant pathogens reproduce sexually or asexually and survive a prolonged period till the favourable weather comes.

7.3.1.2 Host Pathogen Interaction

When the host enters into the plant tissue, immune elicitors stimulate plant defence mechanisms and hypersensitivity response occurs as reported by Stakman (Stakman 1915). Some scientists reported that host pathogen interaction is responsible for the process of apoptosis or programmed cell death in plants (Morel and Dangl 1997). Avirulence (Avr) genes are secreted by pathogens binde indirectly to the plant resistance gene (R). When both the R gene and corresponding Avr genes are present, then recognition takes place which leads to active resistance of the plant. If either Avr gene in the pathogen or R gene in the host is absent or mutated, then no recognition occurs, and the plant becomes diseased (De Wit 1995). As a result, putative reactions occur between two partners and transduction signal cascade is activated.

7.3.1.3 Generation of Reactive Oxygen Species (ROS)

Doke et al. reported that during plant–pathogen interaction, ROS is released and accumulated (Doke 1983). ROS production, otherwise known as ‟Oxidative burst", involves two phases. Phase I is rapid and nonspecific, while phase II is slower but yet yields a higher concentration of ROS (Wojtaszek 1997). ROS is a toxic intermediate that is generated by the reduction of molecular oxygen. Various enzymes are involved in the reaction. NADPH oxidase enzyme helps in the reduction of H_2O_2 under physiological conditions. At first, the reduction of O_2 forms superoxide anion (O^{2-}) and hydroperoxyl radical (H_2O•), and second reduction produces hydrogen peroxide (H_2O_2), and then it reduces to form hydroxyl radical (OH•) that is unstable, but H_2O_2 is more stable. H_2O_2 and OH• react with polyunsaturated lipids in the membrane and form lipid peroxide that results in the destruction of the biological membrane (Grant and Loake 2000).

7.3.1.4 Mode of Action

1. When nanoparticle is taken into the cells through the process of translocation and internalization, it helps in the degradation of intercellular ATP and DNA duplication (Lok et al. 2006).
2. Metal nanoparticles generate ROS and damage the cellular structure (Richards 1981).
3. Metal ion accumulates inside the cell and dissolves the bacterial membrane (McQuillan 2010) (Fig. 7.2).

Under stress conditions, the oxidation reactions occur in the cell, which leads to adverse effects on cell survival, signalling, death and generation of ROS (Mueller et al. 2005). Copper oxide, zinc oxide, silver nanoparticles also show antimicrobial activity against *Escherichia coli, Bacillus subtilis*, *Pseudomonas aeruginosa* and

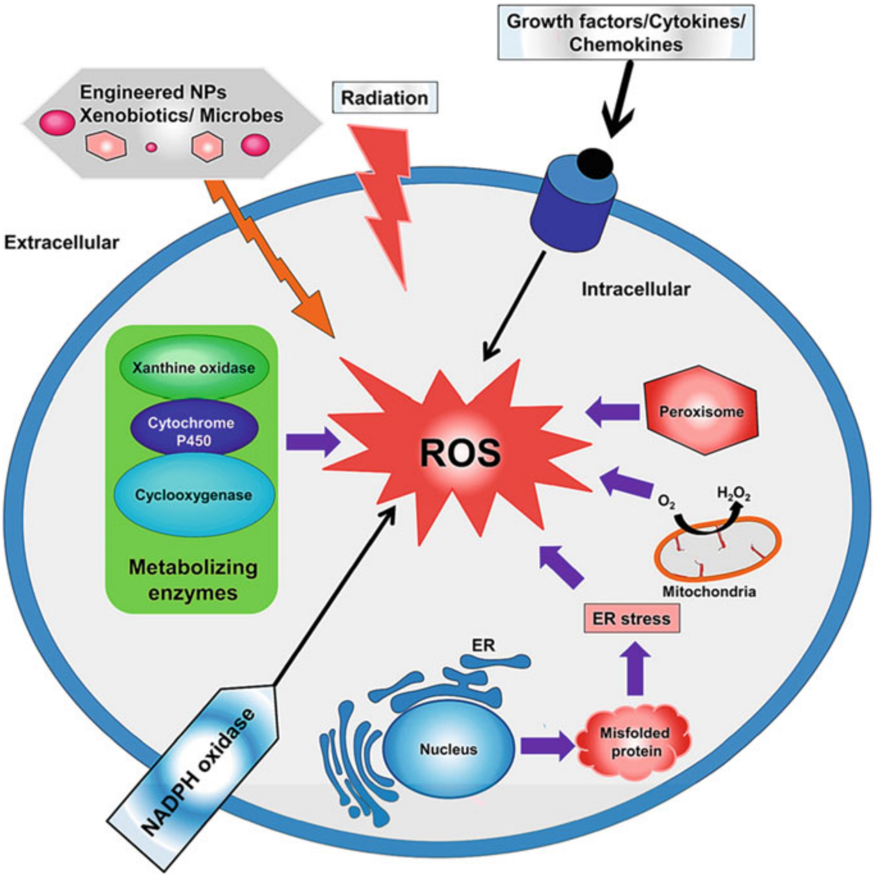

Fig. 7.2 Represents different sources of generation of ROS. Adapted with permission from Ref (Abdal Dayem et al. 2017)

Staphylococcus aureus (Viswanathan et al. 2006). Bacterial plasmolysis is a process in which subsequential degradation of components of cytoplasm and contraction of the plasma membrane take place. Upon treatment of nanoparticles with bacterial cells, nanoparticles form pores in the cell membrane, which lead to the release of intercellular glucose and trehalose into the suspension. Nanoparticles bind to the thiol groups of bacterial proteins and interfere with their activities. After binding to the cell membrane, they alter the cell electrical potential and disturbs the respiration process (Radzig et al. 2013). As a result, ROS is developed, which inhibits the respiratory enzymes (Park et al. 2009) (Fig. 7.2).

7.4 Nanoparticles in Controlling Phytopathogens

Different nanoparticles can be used to control plant pathogens. From different studies, it is reported that metalloids, metal oxides and non-metals are used as either bactericides, fungicide or nano fertilizers by suppressing foliar, stem, fruit or root rot pathogens (Kah and Hofmann 2014). To protect plants from pathogens nanoparticles can be used by two different ways: (1) Nanoparticle itself protects the production of crops (2) Nanoparticles act as a carrier for pesticides, e.g. double-stranded RNA (dsRNA) can be used by spray application, soaking into seeds, foliar tissue or roots. The advantages of nanoparticles as carrier are increased solubility of less water-soluble pesticides, decreased toxicity level, increased lifetime maintenance (Worrall et al. 2018).

Hence, this chapter focuses on nanoparticles mediated plant disease management, which can be used as protectant or carriers for insecticides, fungicides and herbicides. Although, few reports are available on prevention of plant pathogens using nanoparticles, however, the application of nanoparticles in crop pest management is not being explored well.

7.4.1 Nanoparticles Acting as Protectant

Nanoparticles are tiny materials having size in between 1 to 100 nm, and they have unique physical, chemical and biological properties in comparison to bulky material. Nanoparticles have the ability to protect the pathogen such as bacteria, virus, insect and fungus by suppressing the stem, fruit and root rot pathogen. Hence, researchers use different metals and metal oxide nanoparticles such as silver, Cu, ZnO, TiO_2, Au nanoparticles which have antifungal, antiviral and antibacterial properties (Kah and Hofmann 2014).

7.4.1.1 Ag Nanoparticle

Nowadays, the main focus is on green synthesis of Ag nanoparticles from plants, fungi, algae, etc. (Rafique et al. 2017). Initially, Ag nanoparticles were used for the control of pathogens in plants due to antimicrobial activity of Ag nanoparticles (Richards 1981). Kim et al. have investigated that double encapsulation of Ag with Ag^+ ions can be spread on the powdery mildew of roses, which can kill the fungus *Sphaerotheca pannosa* for at least 7 days (Kim et al. 2008). Lamsal et al. found that if 100 mg/mL of AgNP is applied on peppers, it suppress the Anthracnose (Lamsal et al. 2011). It was found that AgNP synthesized from the plant extract can be used for the treatment of banana at different concentrations for the post-harvest control of *Colletotrichum musae* (Jung et al. 2010). It was also found that AgNP has the antimicrobial activity against foliar fungal pathogens. When various concentrations of silver nanoparticles such as 10, 30, 50 and 100μg/mL were sprayed on the cucumber and pumpkin leaves, the suppression of powdery mildew was observed. AgNP can be used for the treatment of fungus, insects and virus (Lamsa et al. 2011). In this context, it is reported that 24μg/mL of silver nanoparticles can be used to treat total germination of *Bipolaris Sorokiniana* in greenhouse trials, which is the causative agent of spot blotch of wheat. From histochemical staining, new facts come out that due to green synthesis of nano-Ag, lignin deposits in the vascular bundles that is a unique and novel approach for disease management (Moussa et al. 2013). Soil-borne diseases which are caused by *Phytophthora parasitica*, *Fusarium spp*. and *Meloidogyne spp*. are suppressed by silver nanoparticle it is reported that, application of nano-silver inhibits the growth of *Sclerotium cepivorum* and *Colletotrichum Gloeasporioides* (Jung et al. 2010). Generally, nano-Ag can be used as anti-parasitic agent. Silver nanoparticle used for the inhibition of Juvelline stage of *Meloidogyne graminis* and its implementation reduces the root gall formation (Cromwell et al. 2014). Ocsoy et al. first developed a new product such as DNA-directed silver AgNP grown on graphene oxide, which has the capacity to suppress bacterial disease caused by *Xanthomonas perforans* on tomatoes. Application of 100 ppm of Ag@dsDNA@GO reduces the severity of bacterial spot disease in comparison to the conventional bactericide treatment (Ocsoy et al. 2013).

The most interesting point about AgNP is that it acts as biocontrol agent. Mallaiah et al. observed that if silver at nanoscale is combined with the biocontrol agents such as *Bacillus subtilis, Pseudomonas fluorescens, Trichoderma viride,* etc., it suppresses Fusarium wilt and increases flower yield from 5% to 12%, 14% and 15%, respectively (Mallaiah 2015). It is also helpful for the reduction of the quality of chemical and increases resistance of pathogens. From the studies, it is also reported that fluconazole has increased fungicide activity, i.e. *Alternaria alternata*, *Cladosporium herbarum* and *Fusarium oxysporum* (Bholay et al. 2013). But according to Gajbhiye et al., when silver nanoparticle biosynthesized from *A. Alternata* is combined with fluconazole, the antifungal activity is enhanced against plant pathogen *Phoma glomerata* (Gajbhiye et al. 2009). However, till now there is limited evidence about the defence mechanism of silver nanoparticle. Mainly silver nanoparticles provide obstacle for the production of toxicity and soil infertility.

7.4.1.2 Cu Nanoparticle

Mainly cu nanoparticles were found to have best antimicrobial activity to control plant diseases. Cu-based fungicides such as kocide 2000 35WG, kocide opti 30WG are used to treat *Phytophthora infestans* in tomato plant. Giannousi et al. monitored the symptoms of leaf lesions in every 10 days of tomato plants and found that when CuO nanoparticles at 150-340μg/ml were applied, they suppress the disease (Giannousi et al. 2013). According to Strager-Scherer et al. Cu nanoparticles, i.e. core shell cu, multivalent cu and fixed quaternary ammonium copper are used to inhibit the bacterial spot by *X. Perforans*. From the studies it was found that nano-CuO molecule can easily penetrate the bacterial membrane and leads to the busting of the cell (Strayer-Scherer et al. 2018). Copper nanoparticle acts as a potent fungicide. Some fungus such as *Fusarium solani*, *Fusarium oxysporum* and *Neofusicoccum sp*., etc. invade vascular tissue of plants and block the water transport system in xylem that forms foliage wilt (Yadeta and Thomma 2013). From the studies, it was found that copper nanoparticles of different concentrations show antifungal activities against *Fusarium solani, Neofusicoccum sp*. and *Fusarium Oxysporum*. Elmer et al. (Elmer et al. 2018; Elmer and White 2016) reported that copper nanoparticles can be used as nanofertilizer. From the studies it is reported that if CuO nanoparticle is treated with tomato and eggplant, then the harvest yield increases 24% than control (Evans et al. 2007). It is reported that when 10μg/L copper nanomaterial is used in maize plant, its growth increases to 51% than the control (Adisa et al. 2019).

7.4.1.3 Zn Nanoparticle

Similar to silver and copper nanoparticle, zinc nanoparticle has antibacterial, antiviral and antifungal activity. According to Paret et al., when photocatalyst technology is combined with nanotechnology, it increases antimicrobial activity and this technique is applied for Zn nanoparticle to treat bacterial spot on leaves of rose plant which is caused by *Xanthomonas spp*. (Paret et al. 2013). It was observed that when Zn nanoparticle was sprayed on leaves of lentil plant, it helps in improving plant growth, chlorophyll, carotene content and protects the cell membrane (Siddiqui et al. 2018). Treatment of Zn nanoparticle at 1000 ppm concentration enhances seed germination, plant growth and early flowering in peanut plant compared to the 2000 ppm concentration as it shows negative and toxic effect to plant (Prasad et al. 2012). It is reported that zinc nanoparticle or Zinkicide, i.e. SG6 when sprayed on leaves of sweet orange and grape fruit, subdue cracker lesion, citrus scab (*Elsinoe fawcettii*) and melanose (*Diaporthe citri*) respectively (Graham et al. 2016). Zinc nanoparticles are also used to treat fungal pathogen (Khan et al. 2016). It is reported that ZnO nanoparticles were used to treat fungus. From the observation, it was noted that different concentrations of ZnO nanoparticle, which is synthesized from zinc acetate at 9 mM/L impede growth and morphology changes of fungus like thinning of the fibres of hyphae takes place. Antifungal activity of zinc oxide nanoparticle was noticed at 12 mM/L against *Botrytis cinerea* and *Penicillium expansum* by distorting hyphae of fungus and inhibiting Conidiophore and Conidia (He et al. 2011).

Similarly, ZnO nanoparticles can act as biocontrol agent which enhances crop health. Zn nanoparticles at 500μg/ml combined with convention fungicide tetraconazole inhibit Cercospora leaf blight of sugar beet and enhance root yield and sugar content (Dimkpa et al. 2013). However, ZnO nanoparticle upon interaction with bio-controlling agent such as *Pseudomonas chlororaphis* O6 and *Fusarium Graminearum* in vitro enhances the efficiency of bio-controlling agent (Duffy 2007). Hence it is used in plant disease management and increases the production of the crop.

7.4.2 Nanoparticles Acting as Carrier

Different nanoparticles are commonly used for drug delivery and upon conjugation with active molecules can be implemented in agriculture to treat plant pathogen.

These nanoparticles can be used as carrier in fungicide, herbicide, insecticide and RNAi inducing molecule as described below:

7.4.2.1 Chitosan Nanoparticle

Chitosan is the deacetylated form of chitin which is found in the walls of fungus and the shells of crustaceans. One of the important properties of chitosan is: it is less soluble due to its hydrophobicity in the aqueous medium; hence it is used in the drug delivery (Kashyap et al. 2015). As a result, it is mixed with organic and inorganic compounds to improve solubility (Li et al. 2011). Chitosan has functional groups such as amine and hydroxyl which are modified and interacted with chitosan molecule. It is attached to the epidermis of leave and stem for long time and facilitates the uptake of the active molecule (Malerba and Cerana 2016).

7.4.2.2 Silica Nanoparticle

Silica nanoparticle has different physical and chemical properties, synthesized using different methods (Mody et al. 2014). They have particular size, shape, crystallinity and porosity which can be used for the drug delivery purpose. Different silica NP can be produced by chemical methods such as mesoporous particle, core shell particle porous hollow silica nanoparticles (PHSN) etc. In PHSN, the pesticide is loaded into inner core to protect the active molecule and delivery to the target area. Hence silica nanoparticles are used to treat plant tolerance against biotic and abiotic stress (Barik et al. 2008).

7.4.2.3 Titanium Nanoparticle

Titanium nanoparticle is also synthesized from different methods in huge amount which can be used in various applications. TiO_2 is polymorphic in nature, is widely used to control pests, nematodes in plants.

7.5 Nanopesticides

Pesticides are used to kill pests or pathogens to protect crop and to increase the yield. It is effective to control disease in plants, but its application has adverse effect to the environment and toxicity due to bioaccumulation. Hence nanopesticides serve as a better alternate to control the pest as well as keep the environment pollution free.

7.6 Insecticides

The coating of insecticides around nanoparticles was first started in the early 2000s. Earlier studies showed that insecticides are classified into contact insecticides and systemic insecticides. Furthermore, based on different physically affected organs such as nerve, muscle, growth, respiration, midgut, it is classified into 55 chemical functional groups (Sparks and Nauen 2015). Agrochemists are now focused to increase the activity of active ingredients of insecticides by reducing the particle

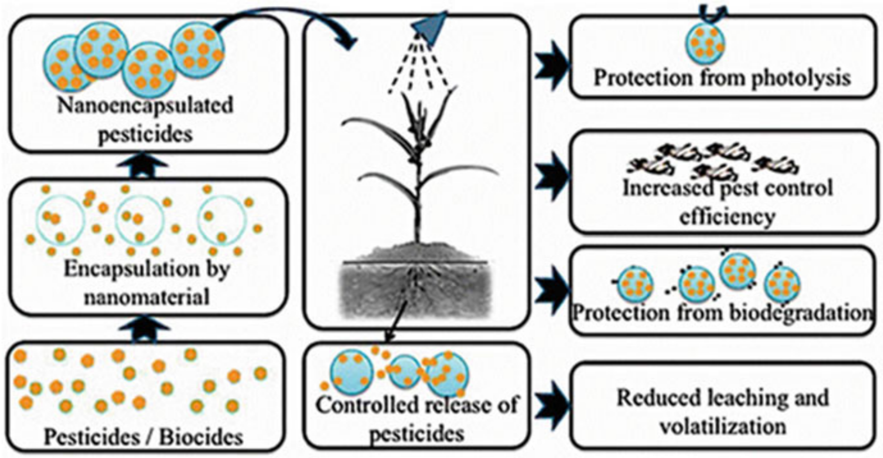

Fig. 7.3 Schematic diagram of nanoencapsulation in pesticide applications. Adapted with permission from Ref. (Nuruzzaman et al. 2016)

size or encapsulating active ingredients into the nanocapsule under normal temperature, alkaline and acidic moisture, etc. (Khan and Rizvi 2017). Hence mostly common nanoparticle carriers were chosen to target the pests or pathogens.

Generally, insecticides are less soluble in water. Hence the requirement of organic solvents is needed, but these results in increased toxicity. Nanoparticles are used to increase the solubility (Worrall et al. 2018). Besides, nanoparticles conjugated with conventional insecticides are used to inhibit the pathogens. Lu et al. reported that when the conventional thiamethoxam is loaded into dendrimer nanoparticles, it results in increased toxicity and mortality of *Hipposideros armiger* (Lu et al. 2013). Similarly, anacardic acid combined with ADH nanoparticle results in increased mortality of *S.litura* than that of anacardic acid-treated alone (Nguyen et al. 2014). Mainly nanoparticles are smaller than conventional/traditional chemicals. Hence it is important for release to the target side.

Evaporation is commonly used for the treatment of insecticides. Essential oil is used to treat but it is quickly evaporate in presence of sunlight, temperature and air (Lai et al. 2006). Hence the best method is encapsulation. If garlic essential oil is encapsulated into PEG (polyethylene glycol) and spread in harvested rice, it inhibits red flour beetles (*Tribolium castaneum*) as shown in Fig. 7.3. Imidacloprid synthesized from PEG and other aliphatic di-acids are used for encapsulation method to treat pest in different crops. Pepperman et al. reported that biodegradable microbial polymers are used to control the pests (Pepperman et al. 1991). Cameron et al. reported that there is a temperature-sensitive polymer such as intelimer, which releases pesticide according to the favourable temperature to protect the active ingredient (Cameron et al. 2018). A microcapsule is a reservoir system in which the target ingredient of pesticide is surrounded by a membrane. (Sotthivirat et al.

2007). Pest can be controlled using chitosan or alginate in this technique. SDS modified Ag/TiO_2 imidacloprid is prepared and applied on soya bean plants that are grown on soil having pH 6.2 and degraded faster within 8 days of treatment (Yan et al. 2005).

7.7 Fungicides

Fungicides are special group of pesticides that kill fungus and spores to provide improved plant protection. The word fungicide came into existence from France as early as 1938. Pierre-Marie-Alexis Millardet was the first to use fungicides to protect the vineyards from the pathogen phyllorexia. There are different types of fungus which cause disease in plants. With the advent of nanotechnology, conjugation of nanoparticles with fungicides has gained much attention. One of the recent techniques developed with the use of nanotechnology is "nano ghosts". According to Hatefalude et al. nano-sized bacterial ghost are taken from Gram-negative bacteria, attached to leave surface and improve the solubility in water of tebuconazole (Hatfaludi et al. 2004). Pyraclostrobin is another widely used fungicide that is conjugated with chitosan lactic co-polymer at different concentrations. After treatment, it inhibits *C. gossypii*, resulting in pest control within 7 days (Xu et al. 2014). Another low-soluble fungicide, Kaempferol, when loaded into chitosan or lecithin, shows 67% inhibition efficiency against *Fusarium oxysporum*. Another well-known method is encapsulation, which is mostly used as it is quick and commercial in nature. Similarly, Janatova et al. found that mesoporous silica nanoparticles (MSN) conjugated with essential oils show higher antifungal against *Aspergillus niger*, over a period of treatment of 14 days (Janatova et al. 2015).

Leaching is the most common method in which water and chemical move through the soil. It was observed that fungicide metalaxyn loaded with MNS showed an increase released rate of water. Similarly, nanoparticle encapsulated in Validamycin shows lesser efficiency than Validamycin alone (Qian et al. 2011). Kumar et.al reported that carbendazim loaded polymeric nanoparticle results in an increased rate of antifungal activity against *Fusarium oxysporum* and *Aspergillus parasiticus* than carbendazim (Kumar et al. 2017).

7.8 Herbicide

Herbicides are chemicals used to kill the herbs. Imazapic and Imazapyr are two widely used conventional herbicides (Maruyama et al. 2016). Interestingly, when these herbicides are conjugated with chitosan nanoparticle, it resulted in reduced toxicity and increased efficacy in inhibiting the *Bidens pilosa* weed. Similarly, when the conventional herbicides, i.e. Simazine and Atrazine were loaded into SLN nanoparticles it resulted in decreased toxicity and enhanced inhibition against *Raphanus raphanistrum*. (de Oliveira et al. 2015). According to Chidambaram et. al, rice husk can also be used as nanoparticle. 2,4-D is inserted into rice husk and it acts as the best herbicide agent against *Brassica species* than 2,4-D (Chidambaram

2016). Grillo et al. show that different concentrations of chitosan coated with polymeric nanoparticles help in attachment to the target plant and kill the pathogens more efficiently. These studies also show that paraquat-loaded chitosan helped to decrease toxicity against alga *Pseudokirchneriella subcapitata in* the presence of aquatic humic substances (Grillo et al. 2014).

7.9 Conclusion

Nanotechnology has emerged as a key tool in the field of agriculture and plant disease management. Conjugation of nanoparticles with pesticides or synthesis of new nanoparticles serving as biopesticides has gained much attention due to various advantages including increasing solubility of low-soluble water pesticides, precise target delivery, reduced toxicity and less pollution to the environment. The application of nanotechnology for plant disease management is yet to be explored in detail. Future works must be focussed to explore the use of nano-based pesticides to increase crop yield and productivity.

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