Environment, Biodiversity & Soil Security
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Biosynthesis of Silver, Zinc, Titanium Nanoparticles Using Pathogenic Fungi: An Overview on Plant Disease supress, Toxicity and Safety

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NOWADAYS there was an attitude from fungi were positive in extra and intracellular production of nanoparticles (NPs) with different in their ability. In order, synthesis of silver, zinc and titanium nanoparticles by pathogenic fungi were became commercially practical and low-cost revival methods for separation of the particles from the fungal mat that can be used routinely in manufacturing procedures. Numerous mechanisms for the synthesis of NPs were reported. Many theories about nanoparticles antimicrobial mechanisms and nanotoxicity on plant ecosystem were proposed. Nanoparticles were considered a good alternative to control phytopathogenic fungi in agriculture. Different shapes and sizes have shown outstanding antifungal activities. Moreover, use of NPs in plant disease control as a novel and fancy approach that may prove effective in the future with the progress of application aspects of nanotechnology has been discussed in detail. Toxicity and safety of nanomaterials in plants and mammalian, future researches whichneeded also were discussed**.**

Keywords: Nanoparticles, biosynthesis, pathogenic fungi, application.

1. Introduction

In nature, most of fungal pathogens were saprophytic and parasitic. Kingdom fungi have been estimated about 1.5 million species of fungal pathogens belong to Basidiomycetes and Ascomycetes, which responsible for high proportion plant diseases and yield losses. Annual crop losses exceeded to 200 billion euros due to fungal pathogens (Gonzalez-Fernandez et al. 2010). Microbial diseases caused 16% losses in agricultural crops and more 70% count of this losses caused by fungal pathogens (Moore et al. 2011).Many of these chemicals are also too expensive for the resource of poor farmers. A reduction or elimination of synthetic pesticide applications in agriculture is highly desirable. In recent years, pathogenic fungi had grown increasingly resistant to commercially available antimicrobial agents. This promoted researchers to look for alternative means to combat microbial and fungal pathogens. Use of NPs in plant disease management is a novel and fancy approach that may prove very effective in the future with the progress of application aspects of nanotechnology. NPs may suppress the pathogen in away comparable to chemical pesticides .Secondly, the NPs can be used as a carrier of some fancy chemicals,viz. pheromones, SAR inducing chemicals ,polyamine synthesis inhibitors (Khan et al. 2014) and many biosensors ,such ones in plant disease identification .Different nanomaterials , i.e. metal oxide and metal, quantum dots, grapheme, carbon nanotubes, etc. can be used in nanobiosensors. For this respect, nanoparticles with antibodies were used for detection of *Xanthomonas axonopodis* (Yao et al. 2009) and Prunus necrotic ring spot virus (PNRSV), which causes disease in peach, plum, apricot, sweet cherry and almonds resulting into yield loss. Zong et al (2014) developed a new method for rapid detection of PNRSV using magnetic nanoparticle based reverse transcription loop mediated isothermal amplification which more highly specific and

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Received: 02/03/2023; Accepted: 07/05/2023
DOI: 10.21608/JENVBS.2023.197433.1211

sensitive than reverse transcription polymerase chain reaction. In the present review, we have discussed the different methods for biosynthesis of nanoparticles. Moreover, various-mechanisms of nanoparticles bioformation , antimicrobial theories of effect of nanoparticles ,schematic applications and aspects of nanotechnology in agriculture, Scope of biosynthesis of Ag, Zn and TiNPs by pathogenic fungi, their antimicrobial effect and application, finally toxicity and safety of this nanomaterials in plants and human had been discussed**.**

2. Synthesis of nanoparticles:

Over the past years, a number of nanofertilizers, nanosensors and nanopesticides products including the nanomaterials have been developed into agricultural practices. There are numerous methods for the synthesis of NPs as figure (1) i.e. sonochemical or sonolysis by powerful ultrasound radiation of 10-20KHz (Suslich et al. 1991); microwave radiation as form electromagnetic energy frequency in range of 300-MHz-300GHz (Komarneni 2003); electrochemical by an electrolyte supplied at 4V and 5A for 30 min by passing an electric between two electrode (Raja et al. 2008); solvothermal decomposition in pomp or autoclave under pressure and heating (Chen et al.2010) ;using chemical reduction by reducing agents, i.e. polyol (Park et al. 2007) ,alcohol, glucose, ascorbate, viz..vitamin C, etc.; microemulsion/collooidal ,i.e. water, oil ,(W/O) surfactant (Umer et al.2012);magnetic ways by physical vapor deposition, mechanical attrition and chemical route from solutions ;various biological materials as plant extracts (phytosynthesis) (Nadeem et al. 2018) and microorganisms, reducing agents, microfluids, marine organisms (Roberto et al. 2019).

3. Mechanisms of synthesis:

The stepwise mechanism of intracellular synthesis of NPs, in the preliminary step of bioreduction, trapping of metal ions takes place at the fungal cell surface. This is probably due to the electrostatic interaction of the positively charged groups in enzymes present on the cell wall mycelia (figure 2). In the next step, the metal ions are probably reduced by the enzymes within the cell wall, which leads to the aggregation metal ions and formation of NPs (Sastry et al. 2003).The extracellular biosynthesis of NPs by many fungi including three mechanisms: nitrate reductase action, electron shuttle quinones or both. The nitrate reductase was carried out by the reaction of nitrite with 2, 3-diaminophthalene, while several enzymes, NADPH-dependent reductases, nitrate-dependent reductases and an extracellular shuttle quinone (figure 2) were implicated (Duran et al. 2005, Kumar et al. 2007) and responsible for the reduction of aqueous silver ions into AgNPs (Rai et al. 2021). Jain et al. (2011) reported that, *A. flavus* synthesis Ag NP by 33kDa protein followed by a protein of cysteine and free amine groups by which stabilizes the NPs, forming a capping agent. Fungal cell wall and wall sugars played an important role in the absorption and reduction of metal ions in extracellular biosynthesis of NPs as reported by Deepa and Panda (2014). The biosource such as fungi that can catalyze specific reactions leading to inorganic NPs is a modern and rational biosynthesis strategy that is an alternative to other physical and chemical methods. In order, synthesis of NPs by fungi became commercially practical and low-cost revival methods for separation of the particles from the fungal mat that can be used routinely in manufacturing procedures, Alghuthaymi et al. (2015).

4. Antimicrobial theories:

Many theories about nanoparticles antimicrobial mechanisms as Figure (3) a , b ,i.e. (1) Disruption of transport systems, including ion efflux (Morones et al. 2005). (2) Generation of reactive oxygen species, which mediated cellular damage and different metal-catalyzed oxidation reactions could underlie specific types of protein, membrane or DNA damage (Zeng et al.2007). (3)DNA loses its ability to replicate resulting in inactivated expression of ribosomal subunit proteins, as well as certain other cellular proteins and enzymes essential to ATP production, membrane-bound such as in the respiratory chain (Kim et al. 2012). In a study, Lemire et al. (2013) and others proposed (4) accumulation prevents the proteins from properly functioning in the membrane, interfering in cell permeability and interfered with their microbial absorption by releasing of toxic ions, that can bind to sulfur which containing proteins. (5) They can be genotoxic - toxic ions that led destroy DNA and cell death. (6) Protein oxidation, interruption of electron transport and membrane potential collapse. (7) Interference with nutrient uptake. These mechanisms may not operate separately suggesting that more than one mechanism can occur simultaneously. The multiple targets of action could help NPs to fight effectively against different plant pathogens.

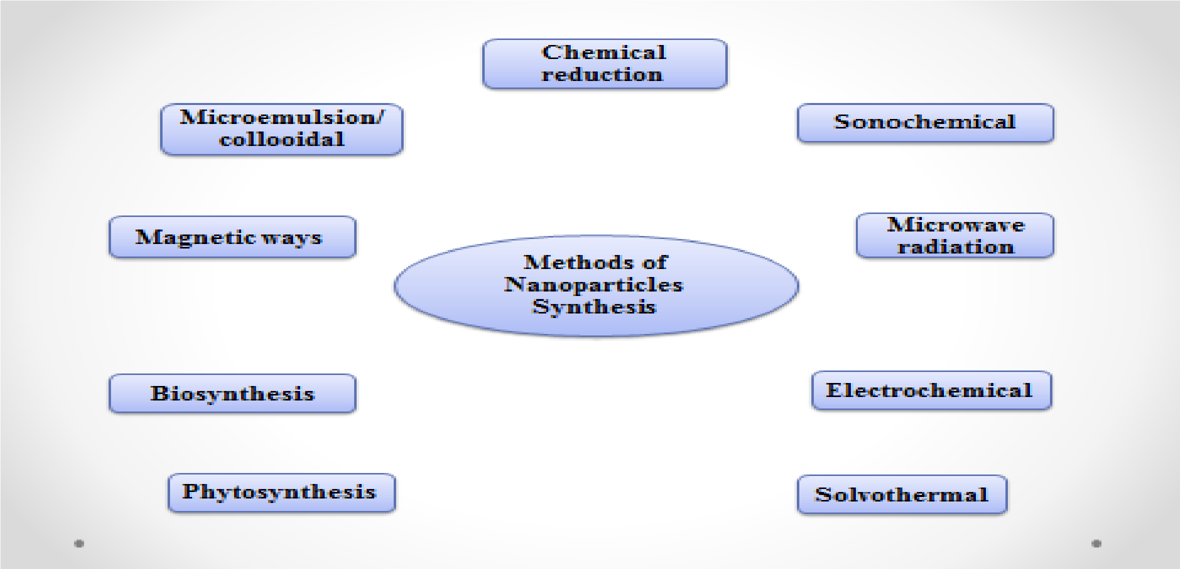

Fig. 1. Numerous methods for the synthesis of nanoparticles.

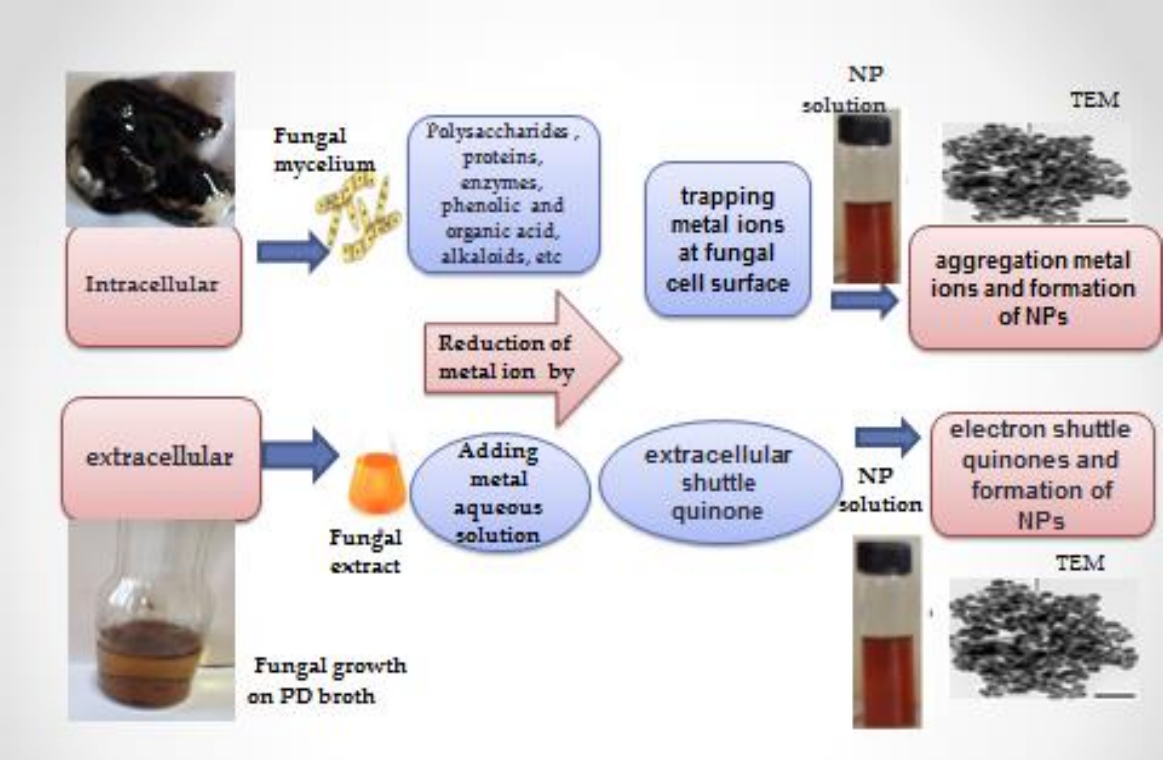

Fig. 2. Show different mechanisms of nanoparticles synthesis.

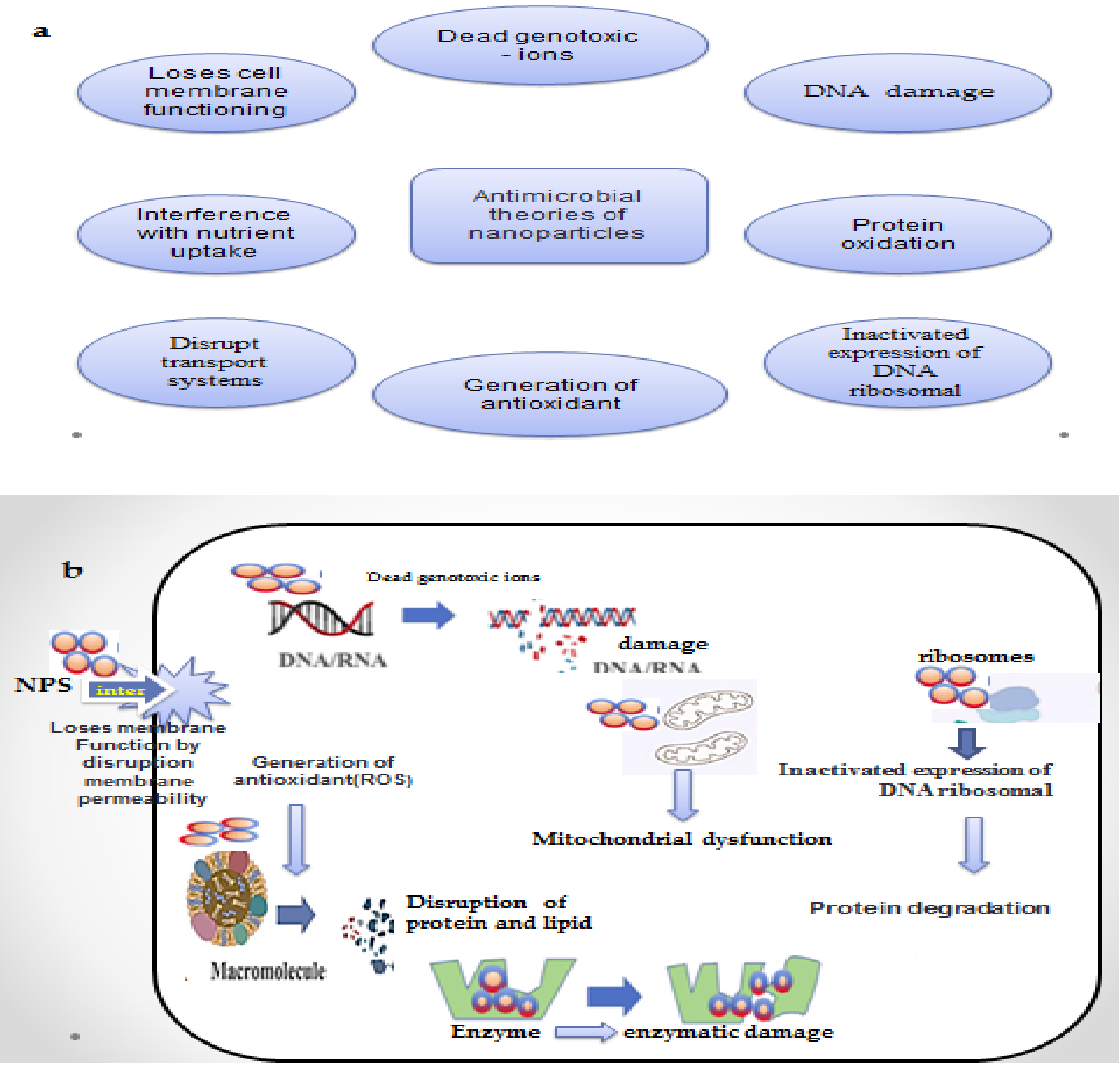

Fig. 3. Different modes of action of nanoparticles as antimicrobial (a), Mechanisms act on cell wall, leading to membrane damage, disruption causes loss of permeability after internalization NPs target main DNA, RNA in the nucleus, ribosomes, mitochondria and enzymes, causing fungal cell death (b).

5. Schematic and applications in agriculture:

There are a number of applications on nanotechnology in agriculture, which have many schematic representation and different aspects.

Nanotechnology assisted advancement in the plant resistance to environmental stresses such as drought, salinity and diseases, also in based gene sequencing of rapid, cost effective identification and utilization of plant gene trait resources (Branton et al. 2008).

5.1. Biosynthesis of nanoparticles:

Extracellular biosynthesis and bioproduction of nanoparticles by fungi with secreting enzymes are done, which reduce the metal salt of macro or micro scale into nanoscale diameter through catalytic effect. Negative electro kinetic potential of microorganisms enables to attract the cations and act as a trigger for biosynthesis of metal and/or metal oxide nanoparticles (Raliya and Tarafdar 2013).

5. 1 .1. Ag biosynthesis :

Microorganisms was constituted one of the vast and strong natural factories of biosynthesis of nanoparticles by harness beneficial effects through biotechnology (Khan 2007) and nanotechnology (Gurunathan et al. 2009). AgNPs solutions exhibited dark red to brown color, this due to reduction of aqueous solution by silver ions and formation of AgNPs (Gajbhiye et al. 2009). Ag^+ required the NADH dependent nitrate reductase enzyme for their reduction to Ago (Roh et al. 2001) or electron shuttle for *F. oxysporum* (Jain et al 2011) or both and secreted by the tested fungi in its extracellular. Intracellularly, AgNPs formed by *Verticillium sp* in range of 2–20 nm (Mukherjee et al 2001). Synthesize nano silver particles extracellulary by many numbers of fungal strains are capable to among which like, *F. oxysporum* (Ahmad et al. 2003), *A. fumigatus* (Bhainsa and D'Souza, 2006), *A. niger*, (Gade et al.2008), *F. semitectum* (Basavaraja et al. 2008), *Cladosporium cladosporioides* (Balaji et al.2009), *Phytophthora infestans* (Thirumurugan et al.2009), *A.clavatus* (Verma et al.2010) and *Amylomycesrouxii* fungus (Musarrat et al. 2010) have been previously described. El-Rafie et al. (2012) were used fungus of *F. solani* for biosynthesis of silver . Eleven different *Fusarium* species isolated from various infected plant have screened for the synthesis of AgNPs .Six of ones werescreened, i.e. *F. oxysporum, F. graminearum, F. solani, F. culmorum, F. tricinctum*and *F. scirpi* were synthesized smaller sized in AgNPs particles, which signifies their prominence synthesis. Among *fusaria* species, *F. oxysporum* was demonstrated a high potential for the synthesis of AgNPs (Gaikwad and colleagues 2013). From 18 different *Phoma* sp., *Ph. sorghina* reported to formation and synthesized of Ag NPs (Gade et al. 2013). In other case, the extracellular synthesis of AgNPs by *Phoma capsulatum*, *Ph. citri*and *Ph. putaminum* was reported by Rai et al. (2015). Moreover, Magdy et al. (2014) added that, six fungal species, i.e. *F. oxysporum*, *A. fumigatus* ,*A. ochraceus, P. italicum, Syncephalastrum racemosum* and *Candida albicans*were exhibited common ones in biosynthesis of silver NPs and reducing silver salt into silver nanoparticles. Devi et al. (2014) reported the *Penicillium funiculosum* GS2 and *Alternaria solani* GS1 as endophytic fungi in *Gloriosa superba* L. plants were used for AgNPs biosynthesis.

The authors outlined the biosynthesis an eco-friendly AgNPs using many different species of *Penicillium*, viz, *P. brevicompactum* (Shaligram et al. 2009), cell filtrate of *P. nalgiovense* AJ15 (Maliszewska et al. 2009), green extracellular AgNPs synthesis using the fungus *P. citrinum* (Honary et al. 2013), *P. brevicompactum* WA2315, *P. purpurogenum* NPMF(Nayak and Nanda 2014) and *P. notatum* which isolated from mould, as an adsorbed antimicrobial without any plausible toxicity (Desai et al.2015). The fungal cell filtrate of A. oryzae (MTCC No. 1846) synthesized AgNPs without using of chemical agents (Phanjom and Giasuddin 2015), *Fusarium* spp. and *Aspergillus spp*. (Rai and Kratosova 2015). The synthesized AgNPs mediated by fungus *A. versicolor* was observed by Elgorban et al. (2016). AgNPs successfully synthesized and characterized from *P. chrysogenum* NG85 and *F. chlamydosporum* NG30 and showed promising antifungal activity as in (Khalil and coworkers 2019). Efforts were made to synthesize AgNPs using mycellial aqueous extract of *Pythium oligandrum*, suggesting a strong candidate for industrial scale production of AgNPs by Noshad et al. (2020). In continuation, *Rhizoctonia solani*, *Sclerotium rolfsii*, *F. verticillioides*, *F. semitectum*, *A. alternate* and *A. niger* fungi were positive in extra than intracellular production of Ag NPs and *F. oxysporum* was the superior one in this respect as reported by Farahat (2021).

5.1.2. Antimicrobial and application:

Released Ag+ ions, which can inhibit several vital functions of cells (Feng et al. 2000). Also NP silvers possess different properties, which might come from morphological, structural and physiological changes (Nel et al. 2003). Upon exposure of AgNPs, microbial cells form a pit in the surfaces leading to AgNP accumulation, subsequently disrupt the permeability of cell membrane and ultimately cause cell death (Sondi and Salopek-Sondi 2004) . Nano sized silvers was shown that the efficiently penetrate into microbial cells, which implies that lower concentrations would be sufficient for microbial control,especially the less sensitive organisms to antibiotics, due to poor penetration into cells and disrupt transport systems, including ion efflux (Morones et al. 2005). The inhibition of fungi increased as the AgNPs concentration increased, due to high density of solution led to deactivate plant pathogenic fungi, cohere fungal hyphae, saturate, DNA loses its ability to replicate, resulting in inhibit the expression of proteins associated with ATP production as well as certain other cellular proteins and enzymes essential to ATP production and 100 ppm of AgNPs have a high inhibition effect (Yamanaka et al. 2005).Park et al. (2006) showed 100% growth inhibition of many fungi, i.e. *M. grisea*, *Colletotrichum gloeosporioides*, *P.ultimum*, *B.cinerea* and, *R. solani*, at 10 ppm of the nanosized silica-silver. Also, reactive oxygen species (ROS) via their reaction with oxygen are known to produce by silver ions, which are detrimental to cells, causing damage to proteins, lipids, and nucleic acids (Hwang et al. 2008). Powdery mildew incited by *Sphaerothecapannosa* Var. *rosae* in rose is a very widespread and common disease in greenhouse and field. It causes leaf curling, distortion, defoliation and reduced flowering. Spraying by 5000 ppm of

AgNPs solution faded out and did not recur for a week more than 95% of rose powdery mildew (Kim et al 2008).

The antifungal activity of silver nanoparticles was conducted against sclerotium-forming phytopathogens, *R. solani*, *S. sclerotiorum*, and *S.minor*, data showed that the nanoparticles strongly inhibited the hyphal fungal growth and sclerotial germination growth. The dysfunction of ion efflux can cause rapid accumulation of silver ions, interrupting cellular processes at their lower concentrations such as metabolism and respiration by reacting with molecules,seriously damaged the walls, resulting in the plasmolysis of hyphae (Min et al. 2009). Similarly, the authors reported the efficacy of Ag NPs *in vitro* on phytopathogen *Raffaeleasp.* fungal (causal of oak wilt), wherever Ag NP caused damage to fungal hyphae, interfered with their microbial absorption, and increased inhibition of fungal growth and conidial germination (Woo et al. 2009). AgNPs have been known strong antimicrobial activities and inhibitory effects in addition to a broad spectrum. Gajbhiye et al. (2009) tested thesynergistic effects of AgNPs from *A. alternate* combined with fluconazole against pathogenic fungi and found that, these combinations enhanced the antifungal against *Trichoderma sp*., *Ph. glomerata* and *Candida albicans* and the reverse was true with *F. semitectum* and *Ph. herbarum*. Silver is now an accepted agrochemical replacement, wherever eliminates unwanted microorganisms in planter soils and hydroponics systems and used as foliar spray to stop fungi, rot, moulds and several other plant diseases. Moreover, it is an excellent plant-growth stimulator, (Sharon et al 2010). AgNPs produced by the fungus *Amylomycesrouxii* showed antimicrobial activity against *F. oxysporum*, *Pseudomonas aeruginosa*, *Candida albicans* and *Bacillus subtilis* (Musarrat et al. 2010). Jung et al. (2010) tested the antifungal activity of Ag NPs against phytopathogens of white rot of the green onion caused by *S. cepivorum* and *C.gloeosporioides* (responsible for anthracnose in a wide range of fruit). The growth of *C. gloeosporioides* was significantly decreased in a dose-dependent manner (Aguilar-Mendez 2010). The studies on the applicability of nano-silver for controlling plant diseases has been limited until now. Silver nanoparticles have high of surface area, fraction of surface atoms and high antimicrobial effect as compared to the bulk silver. Several plant pathogenic fungi belonging to basidiomycetes and ascomycetes produced sclerotia, including *Sclerotinia sclerotiorum*, and *R. solani*. Sclerotia forming pathogens are world widespread, causing many important diseases in a wide host range of different genera plants. Diseases caused by sclerotium-forming fungi were difficult to control and diverse management ways has been used, i.e. chemical methods and genetic controls for the control. The antifungal activity of silver nanoparticles has a great potential against spore-producing fungal plant pathogens, the efficacy of silver is greatly influenced by time and preventive applications and it works better before fungal penetrate and colonize the plant tissues. (Lamsal et al. 2011a). They added alsoAg NPs inhibited the activity of *Colletotrichum* spp. causal of anthracnose pathogen, in field trials. Application of Ag NPs (4-8 nm) enhanced the disease suppression, suggesting that alternative mechanisms such as induction of resistance mechanisms may be reported. Ag NPs penetrated and damaged the cell membrane, subsequently reducing infection (Jo et al. 2009). Lamsal et al. (2011b) also, reported that,testing of 100 ppm AgNPs showed maximum inhibition for the growth of fungal hyphae and conidial germination *Golovinomyces cichoracearum* or *Sphaerothecafusca*, in *in vivo* tests. In the field, application of 100 ppm AgNPs showed the highest inhibition rate of powdery mildew disease on cucumbers and pumpkins. Scanning electron microscope results indicated that the silver nanoparticles caused detrimental effects on both mycelial growth and conidial germination. Interestingly, 15 mg of Ag NPs showed excellent inhibitory activity against fungal plant pathogens, i.e. *M. phaseolina*, *R. solani*, *A. alternata*, *Sc. sclerotiorum*, *B.cinerea*and *Curvularialunata* (Krishnaraj et al. 2012). It has also been hypothesized that Ag^+ primarily affects the function of membrane-bound enzymes, such as those in the respiratory chain (Kim et al. 2012) and exhibited strong antimicrobial activity against *A.flavus* as the plant pathogenic fungus (Jayaseelan et al. 2012) and *F. oxysporum* at 8 gm/L (Gopinath and Velusamy 2013). Before using of silver nanoparticles, phytotoxicity and fungicidal effects in hosts tissues must be conducted until the possibility use of it as an alternative to chemical pesticides. Silver nanoemulsion caused strongly growth inhibition of *Sc.rolfsii* and promoted the growth of mung bean plants. Agrawal and Rathore (2014) reported that, silver ions and nanoparticles against the devastating phytopathogens *Bipolaris sorokiniana* and *Magnaportha grisea*. Higher concentrations and higher toxic effects of silver nanoemulsion were observed, so toxic levels and an optimum concentration should be studied before recommendation to use in agricultural fields. Nano silver eliminates unwanted microorganisms in plants, soils and hydroponics systems and used as a foliar spray against fungi, moulds, rot and several other plant diseases .Moreover, silver is an excellent plant-growth stimulator and it is stable in the environment in the form of a metal or oxide and an accepted agrochemical replacement. Nano silica silver showed antifungal activity and controlled powdery mildew used by *Erysiphe cichoracearum* of pumpkin at 0.3 ppm in both field and greenhouse tests (Patel et al. 2014). In addition (Alghuthaymi et al. 2015) reported that, NPs can offer green and eco-friendly alternatives for plant disease management and mediated gene transfer would be useful for improving of crops resistant to pathogens .The synthesized AgNPs by *Aspergillus versicolor* mediated against *Botrytis cinerea* and *Sclerotinia sclerotiorum* in strawberry plants by Elgorban et al. (2016), *Helmenthosporium oryzae*, *F. oxysporum* and *A. niger* in rice by Elamawi et al. (2016). *R. solani*, *A. tereuse, A.flavus, A. niger*, *P.notatum*, *F.solani*, *F. oxysporum Pythium spinosum* and *Verticillium dahlia* plant pathogenic fungi *in vitro* were suppression by AgNPs of different extents in a range of 70- 100, µg / l and minimum fungicidal concentration were varied due to different types of used fungi . The antifungal activity of the AgNPs is useful in solving different problems in crops production as well as in animals nutrition (El-Saadony et al. 2019).Cruz-Luna et al. (2021) reported that, nanoparticles were considered a good alternative to control phytopathogenic fungi in agriculture and Ag NPs have been the most investigated one due to their good antifungal activities. Different methods have been used to produce these nanoparticles with different shapes and sizes, which have shown outstanding antifungal activities. Akpinar et al. (2021) added that, maximum antifungal effect against *F. oxysporum f. sp. radicis-lycopersici* strains was achieved by decreasing nanosize and increasing concentration of AgNPs. Mycelium growth was decreased about 50- 90% by AgNPs treatment with 3 nm sizes at 25- 50 ppm. The productivity of fungal biomass was found to be too limited at the 25-37.5 ppm of AgNPs concentrations with all sizes. In addition, both septation number, dimensions of micro- and macroconidia were found to be gradually decreased with the application of silver nanoparticles .So, low concentration of AgNPs could be used as potential antifungal and applied for control of phytopathogens. Moustafa et al. (2022) used Ag-NPs paracetamol formula [Ag $(Para)_2(NO_3)_2$] as antifungal at concentration 25-150 mg/ml, which recorded significant reductions of forming colonies with efficiencies to 79.4- 81.8%, against *V. dahlia* (*Verticillium* wilt in olives), *F. acuminatum* and *A. brassicae* ,indicating possibly to using for controlling an agricultural pests. Different modes of antifungal activity of Ag nanoparticles were illustrated in Figure (4).

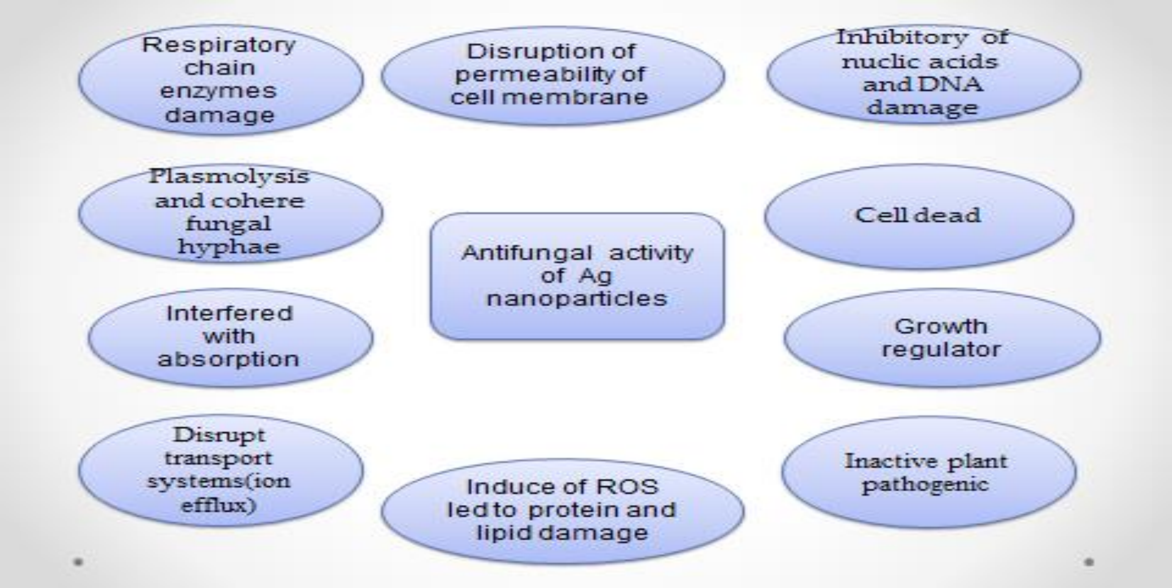

Fig. 4. Different modes of antifungal activityof Ag nanoparticles on fungal cell.

5.2. Zn

5.2.1. Biosynthesis

To date, biological synthesis of Zn NPs have been successfully exploited by several fungi, it is evident that different species of Aspergillus are the prime choice for scientists. Raliya et al. (2013) and Raliya and Tarafdar (2014) demonstrated that, *R. bataticola*, *A. flavus*, *A. niger*, *A. fumigatus*, *A. tubingensis*, *A. oryzae* and *A.terreus*were suitable for Zn NPs biosynthesis. The results suggested that CZR 1 isolate of *A. terreus* showing the maximum 1480.98 g mL^{-1} extracellular protein contents and have potential for nanoparticle synthesis of Zn. The 32kDa protein was responsible for the synthesis of Zn from its precursor compound ZnO. A previous study exhibited the potential for extracellular synthesis of ZnO NPs was by *A. aeneus* fungus (isolated from the soil) and high zinc metal tolerance ability (Jain et al. 2013). In addition, *A. alternate* cell filtrate was used to synthesis of ZnO NPs (Sarkar et al. 2014). Moreover, culture filtrate of *A. niger* ATCC 16404 subjected to the synthesizing of ZnONPs (Kalpana et al. 2018). Five isolates of *A.niger*, *A.tubulin*, *A.fumigatus*, *P.citrinum*and *F. oxysporum* fungi were successfully been extracellular synthesis of ZnO nanoparticles (Hefny et al. 2019). The overall findings of Gao et al. (2019) suggested that *A. niger* had a potential for the biosynthesis of ZnNPs as an alternative biomethods for future application as an antioxidant and antimicrobial compound. *F. solani* isolated from *Chonemorpha fragrans* plant as endophytic strain was capable to synthesize nanoparticles with anticancer activity (Clarance et al. 2020) and ZnNPs successfully entophytic synthesized by *A. tenuissima* fungus culture filtrate, as eco-friendly and a rapid methods (Abdelhakim et al. 2020). Also, silver and zinc NPS had been synthesized by *F. solani* to combat multidrug-resistant pathogens (El-Sayed and El-Sayed 2020). The promising study by Elsayim et al. (2021) found that the ZnONPs synthesized from *A. niger* as biological method.

5.2.2. Antimicrobial and application:

Zinc compounds are mainly used in agriculture as fungicides (Waxman 1998). Recent interest lies in their NP forms. It is believed that the smaller the size of ZnO was the stronger antimicrobial activity it has (Yamamoto 2001). 240 mg/kg of ZnO is the 50% lethal dose (LD50) of oral toxicity for rats (South 2002). ZnNPs had been effective to control plant pathogens by generation of ROS (Seven et al. 2004). ZnNPs had antifungal and /or fungicidal effects as figure (5) due to induce intracellular the generation of reactive oxygen species (ROS). Additionally, it was reported to affect membrane potential by induce lipid peroxidation and overall disruption of membrane function causing cell death (Brayner et al. 2006, Xia et al. 2008). Abd-El-Hai et al. (2009) added that, Mg and Zn NPs suppressed the damping off in charcoal rot disease in sunflower. Lin et al. (2009) added that, ROS stress induced fungicidal mode of action of ZnNPs in three different way, a-biochemically, supported by enzyme assay, b-biophysically, supported by electron micrograph ,c-microarray-based analysis of related ROS genes, in addition, the DNPH binding assay of carbonyl content. Ruffolo et al. (2010) studied potential biocidal efficacy of ZnO and $ZnTiO_3$ NP powders against *A. niger* fungus , moreover, $ZnTiO_3$ NPs showed higher growth inhibition efficiency than ZnO (Jo et al. 2009). Also, ZnNPs significantly inhibited the fungal growth greatly of many fungi, i.e. *B.* formed *cinerea* unusual bulges on the surface of fungal hyphae and hyphae lost their smoothness,that indicating inhibition the growth by deforming the fungal hyphae structures. Moreover, conidia germination of *P. expansum* was completely inhibited and suppressed the conidial development. So, ZnNPs was affecting by cellular function , which led to deformation of fungal hyphae and death of fungi , eventually prevented development of conidiophore and conidia , He et al. (2010). Patra et al. (2012) found that, SEM of *A. niger* hyphae treated by ZnNPs showed dramatic changes of cell surface and structural deformities in the out layers of cell wall leading cell membrane damage. Dimkpa et al. (2011), Lipovsky et al. (2011) and Patra et al. (2012), showed that, ZnNPs have bactericidal and fungicidal due to induce ROS generation intracellular. Panwar et al. (2012) confirmed that, Zn plays a vital role for various metabolic pathways in plant diseases concerned to its deficiency, improved growth and yield. Although, increase of ROS which effective to control plant pathogens (Wani and Shah 2012), lipid peroxidation and reduced glutathione (Muthurman et al. 2014). Moreover, Mahendra et al. (2012) and Abd–Elsalam (2013) reported that application of NPs grower–friendly in agriculture need to be readily use for protection crops and avoiding loss to plant pathogens diseases.ZnNPs efficiently inhibited fungal growth of *F. graminearum* **(**Dimkpa et al.2013), *A. flavus, A. niger, A. fumigatus, F. oxysporum* and *F. culmorum*

(Rajiv et al.2013).

Hamza et al. (2013) added that, ZnNPs can be promising to control of late wilt disease in maize by increasing of PO enzyme with enhancement of yield. Khan and Rizvi (2014) added that, NPs have found suppressive to fungi. Zn plays a vital role for various metabolic pathways in plant system and plant diseases concerned to its deficiency, and improved growth and yield. In addition to Raghu et al. (2014), Anusuya and Sathyabama (2015) reported that, NPs were significantly alteration of ROS (PO,PPO, glucanase and protease inhibitors) and protected turmeric plants against rot disease. Also, Zn NPs promoted plant height, root length and biomass fruit yield and increase of chlorophyll content (Raliya et al. 2015a). Additionally, Graham et al. (2016) showed that, ZnNPs (zinkicide) spraying led to reduce of grapefruit canker lesion development and incidence than bacteriocide cuprous and Zn oxide effective against fungal disease i.e. grape fruit scab and melanose. Jamdagni et al. (2016) conducted that ZnONPs was effective against *A. alternata*, *A. niger*, *B. cinerea*, *F.oxysporum*, *P. expansum* fungi and *A. niger* was the lowest sensitive one. Under *ex-vitro* condition, NPs of Zn caused to reducing of cercospora leaf spot disease severity percentage of sugar beet plants and enhancement of TSS and sucrose contents compared to protected plantsand recorded high enzymes activity values of peroxidase, and polyphenoloxidase, so exhibited as alternative mechanism in defense against the disease (Farahat 2018). Spherical ZnNPs can be internalized more efficiently by cells than ZnNPs other shapes (like hexagonal structure), have more antimicrobial property and ZnNP (chemically biosynthesis) was much more effective than ZnNP (green biosynthesis) in controlling *A. solani* (Yadav et al. 2021) a phytopathogenic fungus, causing “early blight” disease in a number of plants such as tomato, potato, eggplant (Chaerani and Voorrips 2006). SEM clearly show disturbed fungal hyphae of pathogens of *F. oxysporum f. sp.lycopersici* and *A. solani* grown in ZnONPs amended media and adverse effect of NPs on pathogens. Maximum increase in plant growth (shoot dry weight), photosynthetic pigments and proline content and reduction in disease indices (up to 1%) were observed by foliar application of 0.20mL/L ZnO NPs followed by seed priming. Substantial management of above mentioned fungal diseases may be obtained by foliar application of ZnO NPs (Parveen and Siddiqui 2021).

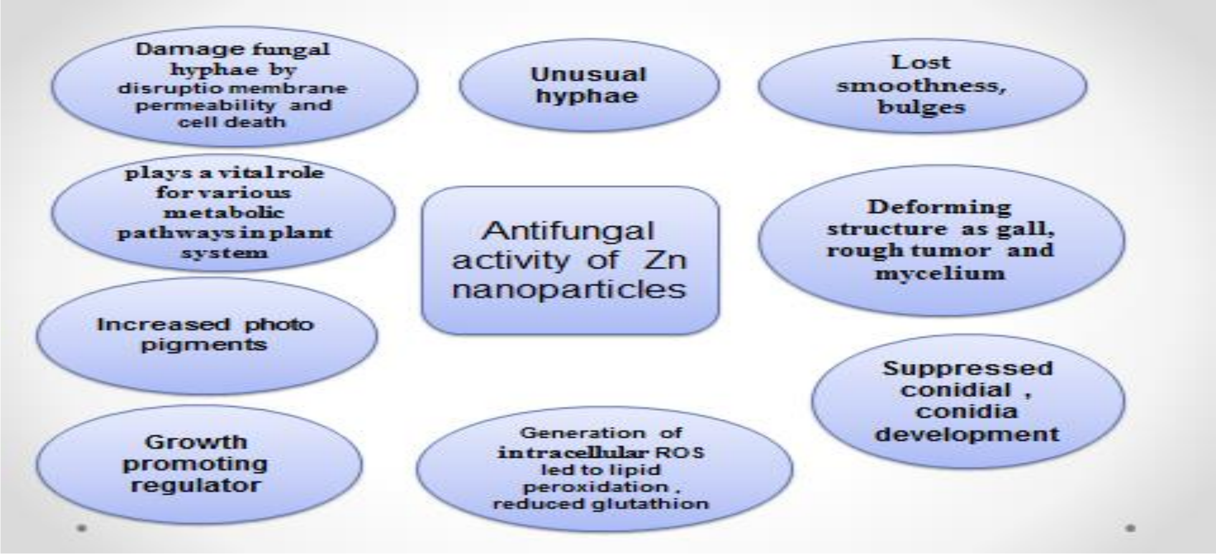

Fig. 5. General antifungal activity of Zn nanoparticles on fungal cell.

5.3. Ti

5.3.1. Biothynsize:

Bansal and coworkers (2005) showed that titania particles may be produced by the fungus *F. oxysporum* with aqueous anionic Ti and the fungus induce the secretion of extra proteins of molecular weight 24 kDa to exposure. Consequently, microorganisms contain proteins; act as reducing and capping agents forming stable and shape-controlled TiO_2 NPs. The biological method of TiO_2 NPs production provides rates of synthesis faster than chemical methods and can be potentially used in various human contacting areas such as foods, cosmetics and sunscreen products (Quadros and Marr

2010).Using of microorganisms in producing of titanium NPs are preferable compare to physical and chemical methods in aspects of costs, energy and security (Azhar et al.2011). TiO_2 nanoparticle (NPs) biosynthesis is a low cost, ecofriendly approach developed using the fungi *A. flavus* TFR 7(Richardson and Simpson 2011). The biosynthesis of TiO_2 NPs was achieved by using *A. flavus* as a reducing, capping agent and biodegradable, a novel and convenient procedure (Rajakumar et al. 2012). Raliya et al. (2013), Raliya and Tarafdar (2014) demonstrated that, *A. flavus, A.terreus, A. niger,* and were *A. oryzae* suitable for Ti NPs biosynthesis and *A. terreus* showing the maximum extracellular protein contents and have potential for synthesis of Ti from its precursor compound TiO_2. The results claim that *A. niger* releases enzymes capable of synthesizing and develop safer TiO_2 nanoparticles (Durairaj et al. 2014). *F.oxysporum* has been effectively in the biosynthesis of TiO_2NPs by Senapati et al. (2014), Ganpathy and Siva (2016). The biotechnologically synthesize of TiO_2 NPs distributed between 50-100 nanometers by *Aspergillus sp*. TK4 was successfully extracellularly from the precursor from TiO_2 (Şahin et al.2020).

5.3. 2. Antimicrobial and application:

Figure (6) concluded the different modes of antifungal activity of Ti nanoparticles on fugal cell. TiO_2 combined with Al and Si was effective in controlling powdery and downy mildews of grapes possibly through stimulation of plant physiological defenses, interference with recognition of plant surface and direct action on the hyphae (Bowen et al. 1992). Application of TiO_2NPs on food crops has been reported to reduce disease severity, enhance yield by 30%, promoting plant growth and increase the photosynthetic rate. It has been reported to show excellent efficacy against Curvularia leaf spot disease incidence and severity in maize. Also, it significantly reduced the incidence of rice blast and tomato mold with increase in yield weight due to growth promoting effect (Mahmoodzadeh et al. 2000). Lu et al. (2002) added that, TiNPs increase of nitrate reductase and stimulation antioxidant effects. TiO_2 NPs has received much attention for the application in the fields due to photocells due to its stability, low cost (Ogura et al. 1987) and photo catalytic activity for degradation of pesticides (Hattori et al. 2006, Pelaez et al.2012). Moreover, Oshira et al. (2008) added that, releasing of ROS depends on the surface area of the semiconductor, which high antimicrobial activity was conducted by smaller TiO_2 NPs, resulted in more oxygen species and higher hydrogen peroxide production at the surface. Secondly, evaporate of water from TiO_2NPs solution, with the progression of time and the remaining unabsorbed TiO_2NPs was found to make plants resistant to external stress. Likewise, a portion of the TiO_2 NPs increases the ability of plants to use solar energy in photosynthesis, which subsequently increases plant growth and yield.

The photo catalytic of TiO_2 NPs and plant protection because it does not form toxic and dangerous compounds hence possess great pathogen disinfection efficiency by dye doping and other suitable has shown (Yao et al. 2009). Small diameter of TiNPs which approximately the same size exhibited a greater antibacterial effect when the zeta potential was more positive and zeta potential plays a significant role in a particle’s ability to penetrate into cell bodies. The reactive oxygen species presence was observed for all titanium dioxide nanoparticle size, therefore, the most significant contributing factor to the antibacterial mechanism was interference with membrane integrity (Simon-Deckers 2009). The activation of defense mechanism and modulating the biosynthesis of phytohormones such as cytokinins and gibberellin by TiO_2 NPs may play a significant role (Mandeh et al. 2012). In addition, TiO_2NPs is useful as protective encapsulating agents that increase adhesive force in the interspecies relation between bacteria and plants (Webster et al. 2008 and Chowdhury et al. 2012). Moreover, higher growth inhibition efficiency of $ZnTiO_3$ NPs showed than ZnO (Jo et al. 2009). .Ruffolo et al. 2010 , Song et al. 2014) added that $ZnTiO_3$ and Zn hydroxide carbonate NPs showed higher growth inhibition efficiency of *A. niger* and fungal activity against cotton *Verticillium, Rhizopus, Mucorales* than ZnONPs, respectively. Size of the spherical (about 3.5 nm) thus resembling the nanoparticles obtained from TiBALDH have been proven to be biocompatible with lung cell cultures and pollen grains and TNs seem environmentally benign and convey properties that might make them useful as protective encapsulating agents that increase adhesive forces in the interspecies relationship between bacteria and plants (Groenke et al.2012). Moreover, TiO_2 have been improved crop yield through nitrogen photo-reduction with beneficial physiological responses and to incorporated into fertilizer as a photocatalytic bacteriocid (Larue et al .2012). TiO_2NPs have become one of the most important substances in nanotechnology, which caused plant growth promoting to roots of oilseed rape and protected the plants against *A. brassicae* infection and adhesive effects on bacteria, *Bacillus amyloliquefaciens* (Palmqvist et al. 2015). TiO_2 NPs is suitable for nutrient and application at 10 mg/L on the leaves of mung bean plants, leading to significant improvement in shoot length, root length, root area and nodule, chlorophyll content and total soluble leaf protein. Also, microbial rhizosphere population were increased by 21.4-48.1% and activity of acid phosphatase (67.3%), alkaline phosphatase (72%), phytase (64%) and dehydrogenase (108.7%) enzyme was observed over control in plants owing to application of TiO_2 NPs. A possible mechanism has also been hypothesized for TiO_2 NPs biosynthesis. Also, Ti NPs promoted plant height, root length and biomass fruit yield and increase of chlorophyll content (Raliya et al.2015a,b). TiO_2 NPs had potential antimicrobial for management of pathogenic microorganisms affecting agriculture crops (Rai and Kratosova 2015). It is speculated that, TiO_2 NPs as nanocide can enter into the cells by crossing the cell layer and can defeat the resistance of microorganisms and fungi may not become resistant to such physical mechanism (Hamza et al. 2016a).They also in 2016b added that, TiO_2NPs was exhibited the most effective treatments for cercospora leaf spot disease suppression in the field application. Yield and growth characters of treated sugar beet significantly increased in comparison to control. So, TiO_2NPs may offer alternative control for leaf spot in sugar beet.

Additionally, Farahat (2018) added that in field conditions, NPs of Ti caused to reducing of cercosporaleaf spot disease severity in sugar beet plants followed by enhancement of TSS and sucrose contents, activation of oxidative enzymes of peroxidase and polyphenoloxidase. Antifungal activity of TiO_2NPs was evaluated against *Ustilago tritici* pathogenic plant fungus of wheat rust, which is one of the major causes of serious loss in crops. It was inhibited at different concentrations and exhibited a safer option due to less adverse effects (Ahmad et al. 2018). Exogenous application of synthesized TiNPs conferred to induced antioxidant genes and enzyme systems like, superoxide dismutase and catalase and tolerance to arsenic oxidative injuries (Katiyar et al. 2020).Foliar spray with 0.20 mL/L TiO_2 NPs provided management of fungal *F. oxysporum f. sp. lycopersici* and *A. solani* diseases of tomato and useful in increasing defense enzymes, viz. superoxide dismutase,ascorbate, phenylalanine ammonialyase, peroxidase , catalase activities and chlorophyll content, proline and plant growth. SEM revealed the absorption of TiO_2 NPs by trichomes, stomata and seed surface (Parveen and Siddiqui 2022). Hamzat et al.(2022) added that, foliar applications of TiO_2 NPs, treatments in the field recorded significantly reduction in the disease incidence (9.75 and 10.88%) and severity (1.00 and 1.06) for Curvularia spot and leaf blight of maize, respectively. Also, application of 625 cc ha^{-1} of TiO_2 at 4 and 8 WAP combined with NPK fertilizer increased the yield by 30%.

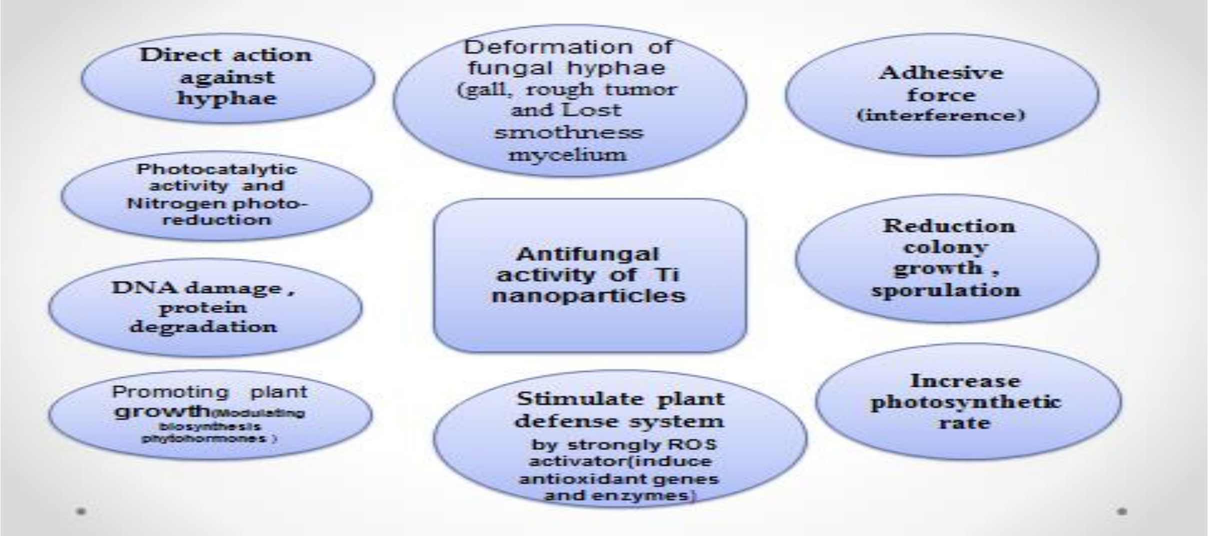

Fig. 6. Different modes of antifungal activity of Ti nanoparticles on fugal cell.

Table 1 and 2 concluded the bioproduction and antimicrobial activity of nanoparticles of siliver, zinic and titanium by plant pathogenic fungi.

Table 1. Bioproduction of nanoparticles of siliver, zinic and titanium by plant pathogenic fungi.

Nano materials	**Pathogenic Fungi produced**	**Product type**	**References**
AgO	*Verticillium sp*	Intracellular	Mukherjee et al. (2001)
	Fusarium oxysporum	Extracellular	Ahmad et al. (2003)
	Aspergillu. niger	Extracellular	Gade et al. (2008)
	Fusarium semitectum	Extracellular	Balaji et al. (2009)
	Cladosporium cladosporioides	Extracellular	Basavaraja et al. (2008)
	Phytophthora infestans	Extracellular	Thirumurugan,et al. (2009)
	Aspergillus clavatus	Extracellular	Verma et al. (2010)
	Amylomyces rouxii	Extracellular	Musarrat et al. (2010)
	Fusarium. graminearum, Fusarium. solani, Fusarium oxysporum, Phoma sp., *Fusarium tricinctum, Fusarium culmorum, Fusarium scirpi, Phoma sorghina*	Extracellular	Gaikwad et al. (2013)
	Phoma capsulatum, Phoma citri, Phoma putaminum	Extracellular	Gade et al. (2013)
	Fusarium oxysporum, Aspergillus. fumigatus, Aspergillus.ochraceus,	Extracellular	Rai et al. (2015)
	Penicillium italicum, Syncephalastrum racemosum, Candida albicans	Extracellular	Magdy et al. (2014)
	Penicillium funiculosum, Alternaria solani		Devi et al. (2014)
	Aspergillus. versicolor	Extracellular	Elgorban et al. (2016)
	Penicillium chrysogenum, Fusarium chlamydosporum	Extracellular	Khalil and coworkers (2019)
	Pythium oligandrum	Extracellular	Noshad et al. (2020)
	Rhizoctonia solani, Sclerotium rolfsiiFusarium verticillioides,	Both	Farahat (2021)
	Alternaria alternate Aspergillus niger, Fusarium oxysporum, Fusarium semitectum	Extracellular	
ZnO	*Aspergillus flavus, Aspergillus terreus, Aspergillus tubingensis , Aspergillus niger, Aspergillus fumigatus,*	Extracellular	Raliya et al. (2013) ,Raliya and Tarafdar (2014)
	Rhizoctonia bataticola; Aspergillus. oryzae; Alternaria alternate	Extracellular	Sarkar et al. (2014)
	Aspergillus niger	Extracellular	Kalpana et al.(2018)
	Aspergillus niger, Aspergillus tubulin, Aspergillus fumigatus, Penicillium citrinum, Fusarium. oxysporum	Extracellular	Hefny et al. (2019)
	Aspergillus niger	Extracellular	Gao et al.(2019)
	Fusarium solani	Extracellular	Clarance et al. (2020)
	Alternaria tenuissima	Extracellular	Abdelhakim et al. (2020)
	Fusarium solani	Extracellular	El-Sayed and El-Sayed (2020)
	Aspergillus niger	Extracellular	Elsayim et al. (2021)
TiO_2	*Fusarium oxysporum*	Extracellular	Bansal and coworkers (2005)
	Aspergillus flavus	Extracellular	Richardson and Simpson (2011)
	Aspergillu. flavus	Extracellular	Rajakumar et al. (2012)
	Aspergillus flavus, Aspergillus terreus, Aspergillus niger, Aspergillus oryzae	Extracellular	Raliya et al. (2013)Raliya and Tarafdar (2014)
	Aspergillus niger	Extracellular	Durairaj et al. (2014)
	Fusarium oxporum	Extracellular	Senapati et al. (2014)
	Fusarium oxporum	Extracellular	Ganpathy and Siva (2016)
	Aspergillus sp.	Extracellular	Şahin et al. (2020)

6. Toxicity and Safety in plants and human:

Nanoparticles interfere with the plant transport pathways as a physical barrier rather by inhibiting through the blockage of the intercellular spaces in the plant cell wall or cell wall pores. On the other hand, the chemical nanotoxicityis related to the excessive production of reactive oxygen species (Nel et al. 2006). There are many gaps in our knowledge on the agricecotoxicity of NPs, many unresolved problems and new challenges concerning the biological effects (Monica et al. 2009). Plants interactions with nanoparticles have two likely modes of nanotoxicity, ie. physical and chemical. Physical nanotoxicity is closely associated with the restricted flow ofnutrients as a direct consequence of apoplastic or symplastic trafficking (Ma et al.2010). ZnO at 100 - 1000 mg/L15 mg/L was stunted root growth in *Oryza sativa* ,50 % inhibitory concentration to root growth of *Allium sativum* (Boonyanitipong et al.2011).The ecotoxicological effects of nanomaterials on plants , plant-soil interactive systems and soil micro-organisms are still largely unknown and need widely investigation (Vittori-Antisari et al.2011 , Lee et . al. 2012).

6.1. Ag

Ag was environmentally safe and even beneficial to human health (Yau et al. 2004) and nontoxic to humans (Yeo et al. 2003,Elchiguerra et al. 2005). Since Ag is environmentally safe and even beneficial to human health, the charge of nanosized silica-silver is much less in commercial fungicides; it is believed that the formulation is very important in the management of various fungal plant diseases in eco-friendly sustainable agriculture (Yau et al. 2004). So, it is has antimicrobial activity against pathogens and a wide applications in metal or NP form. The charge of nanosized silica-silver was very important in the management of various fungal plant diseases in eco-friendly sustainable agriculture and successful applied as a thin film to boost cereal germination and reduce fungal growth. AgNPs may be less toxic to animals and humans than synthetic fungicides. Moreover, the toxicity that nanoparticles may be coupled with some positive effects in plants, algae and fungi, (Sondi et al. 2004). Nanosilver may be controlled of various plant pathogens in a moderately safer way compared to synthetic fungicides (Oh et al.2006). However, Nano-Ag used as a disinfectant drug also has some risks as the exposure to silver can cause argyrosis or argyria; it can be toxic to mammalian cells (Gong et al. 2007).

Table. 2. Antimicrobial effect and application ofsiliver, zinic and titanium nanoparticles against phytopathogenic fungi.

Nano materials	Activity against Pathogenic Fungi	Impact	References
AgO	*Sphaerothecapannosa Var rosae*	Powdery mildew	Kim et al. (2008)
	Rhizoctonia solani, Sclerotinia sclerotiorum Sclerotinia.minor	*In vitro*	Min et al.(2009)
	Raffaelea sp.	*In vitro*	Woo et al.(2009)
	Fusarium oxysporum, Pseudomonas aeruginosa	*In vitro*	Musarrat et al. (2010)
	Candida albicans Bacillus subtilis	*In vitro*	
	Sclerotiorum. cepivorum	*In vitro*	Jung et al. (2010)
	Sclerotinia sclerotiorum, Rhizoctonia solani.	*In vitro*	(Lamsal et al. (2011)
	Alternaria alternata, Sclerotinia sclerotiorum, Macrophpmina phaseolina,	*In vitro*	(Krishnaraj et al.(2012)
	Botrytis cinerea, Curvularia lunata, Rhizoctonia solani,	*In vitro*	
	Aspergillus flavus	*In vitro*	Jayaseelan et al.(2012)
	Fusarium oxysporum	*In vitro*	Gopinath and Velusamy (2013)
	Bipolaris sorokiniana, Magnaportha grisea	*In vitro*	Agrawal and Rathore (2014)
	Erysiphe cichoracearum	Powdery mildew	Patel et al. (2014).
	Botrytis cinerea, Sclerotinia sclerotiorum	Fruit rot	Elgorban et al. (2016)
	Rhizoctonia solani, Aspergillu. tereuse , Aspergillus flavus, Aspergillus niger, P.notatum	*In vitro*	(El-Saadony et al.(2019)
$Ag(Para)_2(NO_3)_2]$	*Fusarium.solani, Fusarium oxysporum ,Pythium spinosum, Verticillium dahlia*	*In vitro*	
	Fusarium oxysporum f. sp. radicis-lycopersici	*In vitro*	Akpinar et al. (2021)
	Verticillium dahlia, Fusarium acuminatum Alternaria. brassicae	*In vitro*	Moustafa et al. (2022)
ZnO	*Aspergillusniger*	*In vitro*	Patra et al.(2012)
	Aspergillus flavus, Aspergillus fumigatus, Aspergillus niger, Fusarium culmorum,	*In vitro*	Rajiv et al.(2013)
	Fusarium oxysporum Fusariumgraminearum	*In vitro*	Dimkpa et al. (2013)
	Cephalosporium maydis	Late wilt	Hamza et al. (2013)
	Aspergillusniger	*In vitro*	Ruffolo et al.(2010)
	Botryris cinerea Penicillium expansum	*In vitro*	He et al. (2010)
Zinkicide	*Xanthomonas. citri subsp. Citri* *Elsinoe fawcetti,*	Grape fruit canker Grape fruit scab	Graham et al.(2016)
ZnO	*Alternaria alternata, Aspergillus niger, Botrytis cinerea, Fusarium oxysporum, Penicilliuem xpansum*	*In vitro*	Jamdagni et al. (2016)
	Cercospora beticola	Cercospora leaf spot	Farahat (2018)
	Alternaria solani	*In vitro*	Yadav et al. (2021)
	Fusarium. oxysporum f. sp. lycopersici, Alternaria solani	Wilt, leaf blight	Parveen and Siddiqui (2021)
Ti ($ZnTiO_3$) **TiO_2**	*Aspergillus niger,Verticillium, Rhizopus,*	*In vitro*	Song et al. (2014)
	Alternaria brassicae	Alternaria spot	Palmqvist et al. (2015).
	pathogenic microorganisms	Agriculture crops	Rai and Kratosova (2015)
	Cercospora beticola	Cercospora leaf spot	Hamza et al. (2016b), Farahat (2018)
	Ustilago tritici	Rust	Ahmad et al.(2018)
	Fusarium oxysporum f. sp. lycopersici, Alternari solani	Wilt, leaf blight	Parveen and Siddiqui (2022).
	Curvularia lunata, Heminsporium turcicum	Curvularia spot, leaf blight	Hamzat et al.(2022)

The hemolytic activity of Nano-Ag was investigated as an indicator of its cytotoxicity to mammalian cells. The toxicity of silver nanoparticles (mean diameter = 10 nm) to human fibroblast cells performing viability assays at 24 hours with 25- 400 µg/mL, which led to reduce cell viability to 100 - 60%, respectively, compared to control. Ag NPs at or above 200 µg/mL reduced viability to less than 50% of control at the 72-hour time point. The dose-dependent toxicity of Ag NPs to the generation of reactive oxygen species and DNA damage were reported by AshaRani et al. 2009). The result suggested that Nano Ag could be applied to- therapeutic agents regarding human fungal diseases with low cytotoxicity (Lee et al. 2010). AgNPs disrupted cell division process causing chromatin Bridge, stickness and cell disintegration (Kumari et al. 2010). It has also been successful applied as a thin film to boost cereal germination and reduce fungal growth. Silver at 40 mg/L led to completely inhibited root hair formation, deformation of roots in *Allium cepa* (Yin et al.2011). The safety and efficacy of nanosilver products had spent millions of dollars by the American Biotech Labs on testing. Their studies have showed that nanosilver products are not toxic to animals or humans at low concentration. Moreover, resistance pathogens development were prevented by antimicrobial activity of AgNPs. So, it can be used safety in the control of plant pathogens as an alternative to synthetic fungicides, due to these properties (Loo et al.2018). The toxicity of AgNPs depends upon the size, shape, surface charge and used concentration (Rai et al. 2021).

6.2. Zn

Comparing to previous study which established the cytotoxic threshold for ionic zinc 10 ppm and 32 ppm were found (Borovansky et al. 1989, Palmiter et al. 2004). In particular, ZnO NPs are environment friendly, non-toxic, biosafe and biocompatible making them an ideal candidate for biological applications (Rosi and Mirkin 2005). Cytotoxicity of ZnO NPs on a model neural cell showed 10, 25, 50, and 100 µg/mL concentrations were reduced cell viability to 90, 80, 55, and 10%, respectively of control (Jeng et al.2006). The cytotoxicity of ZnO NPs (average diameter 19 nm) was tested on human mesothelioma MSTO-211H and rodent 3T3 fibroblast cells by Brunner et al.(2006) and showed that 3.75 µg/mL did not significantly reduce viability after 3 days followed by 7.5 µg/mL reduced viability of both cell types to 75%, and at or above 15 µg/mL killed nearly all cells present. The toxicity of ZnO NPs partially attributes to the release of zinc ions and note that it is unclear whether ion release before or after nanoparticle uptake by the cells is most relevant. They also note,15 µg/mL was critical amount of zinc and present to radically inhibit cell viability. The harmful impact of nanomaterials on living cell have addressed by several reports, but low concentrations of ZnO are nontoxic to eukaryotic cells (Zaveri et al. 2010). In one study, phytotoxicity of nanoparticles of zinc and zinc oxide on seed germination and root growth of radish, rye grass, rape, lettuce, corn and cucumber as a higher plant species were studied and reported that, seed germination was not affected with except of the inhibition of ZnNPs on corn at 2000 mg/L and on ryegrass. Root growth inhibition varied greatly among nanoparticles and plants, and 2000 mg/L ZnO or ZnNPs practically terminated root elongation of the tested plant species. IC50 of ZnO and ZnNPs were estimated to about 20 mg/L for rape and rye grass, 50 mg/L for radish. This results are disposal of engineered nanoparticles and significant in terms of use (Lin and Xing 2007). Additionally, good biocompatibility to human cells to ZnONPs had been reported by Padmavathy and Vijayaraghavan (2008). Another study by Lai et al.(2008) investigated the ZnO NPs (diameter ≤100 nm) on viability of human astrocytoma U87 cells and exposed to varying concentrations of nanoparticles added to the cell culture media. At 1 µg/mL or less, cell viability was not affected but at 10 µg/mL cell viability was reduced to about 55% of control and at or above 25 µg/mL reduced cell viability to less than 5 %. Application of ZnNPs mediated cytotoxicity on various tissues of a dipocytes and most of cells are dead at higher concentrations (Lipovsky et al. 2011, Patra et al. 2012).Moreover, ZnO NPs could be used as food safety and an effective fungicide in agricultural applications (He et al. 2010, Krishnaraj et al. 2012). Hernandez et al. (2013) reported that, Zn NPs did not accumulate in the grains thus were safe to use as a nutrient. Zinc is a mineral element essential to human health and ZnO is a form in the daily supplement for zinc. Additionally, US Food and Drug Administration (FDA 21CFR182.8991) recognized ZnO as generally as safe (FDA 2015) due to its nontoxic properties (Pulit-Prociak et al.2016).

6.3. Ti

In spite of, TiO_2 NPs appears to be safe on plant surfaces with high oxidizing power (Frazer 2001).The synthesized TiO_2NPs do not restrict any toxicity, proliferation and provide evidence of biocompatibility in MG63 cell lines (Pinkerton 2009). Moreover, TiO_2 NPs are used in areas related to human health such as pharmaceutical industries as well as in bone tissue engineering because of their nontoxic and biocompatible properties (Malarkodi et al. 2013). Schiling et al. (2010) demonstrated that, TiNPs are considered to be save up to 30% in products of food colorant. Ahmad and Rasool (2014) added that ,NP TiO_2 considered harmless and nontoxic when used in food products up to1 % of the product mass .The ecological toxicity of TiO2 is depended on the exposure concentration and the crystal structure of the NPs, also the exposure routes such as dental, oral and inhalation (Varner2010). Hamaza et al. (2016b) found that, TiO_2 NPs had no toxicity or mild compared to the high dose offered orally to the treated rats and it is expected that people will not be exposed to this level under any conditions. TiNPs showed that reducing health risk associated with its contamination in the food chain, thereby lowers lead uptake and bioaccumulation in *Oryza sativa* ,Cai et al. (2017). Ti and Fe NPs dioxide are included in human food pigments and coloration, respectively (McClements and Xiao 2017).

7. Conclusion and future research:

In agricultural crops, microbial diseases caused 10-20% losses, resulting in billions of dollars of losses to agriculture and more 70% count of this losses caused by fungal pathogens. In last years, pathogenic fungi had resistant to available antimicrobial agents. This promoted to look for alternative means to combat fungal pathogens. Use of NPs synthesized by pathogenic fungi in plant diseases management is a novel and fancy approach that may prove very effective in the future with the progress of application aspects of nanotechnology. Eco-green synthesis of NPs using plant pathogenic fungi as microorganisms is environmentally friendly and a promising approach for different agricultural applications such as fungal diseases control, nano-fertilizers, nano-biosensors and nano-pesticides. In the future, given their potential wide spread use, it is likely that large volumes of NPs produced by different methods will enter ecosystems. Crop disease management by a novel platforms are essentially needed to be a central component for long time strategy in increasing or sustaining agricultural production. NPs may suppress the pathogen in away comparable to chemical pesticides. The potential of applications of nanotechnology particularly in agriculture has generated revolution. The biosynthesis of nanoparticles using pathogenic fungi is a novel and emerging field of bio eco-friendly and sustainable nanotechnology. Application of nanoparticles would open a vista of research in integrated manner such as pathogens control, plant-pathogen interactions, integrated pest management and many more. However, several aspects of nanoparticles with relation to plants viz., half-life in soil, toxicity effects on plants and optimum dosage of application in open field needs to be determined. Future research should be targeted to optimize treatment success and maximize yield. Investigations of mechanistic effects in the crop or cropping system so as to address concerns over risks and food safety. There are many questions remaining to be addressed, as the exact mechanisms of interaction of nanoparticles with fungal cells and how nanoparticles influencing and killing. Many pathogenic fungi affected in vitro, controlled and /or reduced by application of nanoparticles in the green house and in the field, in the same time many pathogenic fungi have economic values in producing different nanoparticles but this substances will effect against themselves or effect on other microorganisms only. The potential use of NPs materials to address, these needs has been a topic of discussion for many years. The little information exists to accurately assess NPs hazards in the environment, and the use of NPs to suppress plant diseases and enhance yield should be proceeded with cautions. Uptake studies of phytotoxicity on seed systems, i.e. Germination, adsorption, root length and accumulation of NPs into the plant systems exposed to different NPs is needed (Kumari et al. 2012). As well as, phytotoxicity on flowers, fruits, green fresh leaves which used directly. Many efforts are needed to clarify the interaction between nanosubstances, plants, microflora, phylloplane endophytes and soil microorganisms, both pathogenic and plant health. Also, further studies needs to estimate the effects of different NPs on agricultural environment, also recommend models for this estimation, i.e. biological models of mechanisms of interaction between different organisms could be proposed to study the effects of NPs on biological systems such as those represented by plants and associated microbes (Thul et al. 2013).

NPs as Ag, Zn and Ti are known to play critical roles in plant disease resistance by directly inhibiting disease causing organisms, through enzymes activation for defense barrier and/or by affecting the systemic acquired resistance pathway. However, NPs availability in soil and poor intra-plant translocation inherently limit the utility of amendment strategies. At the nanomaterials acquire unique physical and chemical characteristics not observed in equivalent application, availability and transport in biota, including plants, as a function of nanometer particle size. Preliminary suggested significant potential for nanomicronutrients, either by foliar, root application and seed treatment, to suppress disease and decrease crop yield losses. The applications of different nanoparticles as antimicrobials have attracted the interest of the researchers globally. Further studies of the mechanism of the synthesis of nanoparticles should be needed because it remains warrants and unclear. The process is bio eco-friendly and economically viable nanoparticles synthesized by pathogenic fungi have huge potential in agriculture as fungicidal agents of the new generation. Looking at the potential applications of nanoparticles, the toxicity is a major issue that depends up on size, shape, surface charge, and the dose of nanoparticles used. Thus, it can be recommended that pathogenic fungi are a promising for the biosynthesis of nanoparticles and its potential should be needed in agricultural applications. Nanomaterials have given a promising future for the modern agriculture practices, i.e. precision delivery of fertilizers, enhanced nutrition, plant growth and plant disease resistance at an early stage. Nanomaterials can improve fungicides application through allowing the slow, sustained release of the active substances and providing better penetration to plant pathogens. So, nanotechnology can provide bio, efficient and eco-friendly strategy for plant pathogens management in agriculture. The advanced nanotechnology techniques can improve the way identification, detection and forecasting of plant pathogens in the promising future in the upcoming age of agricultural mechanizes, as well as, help us in controlling its, increase the yield and decrease food losses in quality and quantity.

Ethics approval and consent to participate: This article does not contain any studies with human participants or animals performed by any of the authors.

Consent for publication: All authors declare their consent for publication.

Funding: There is no external funding.

Conflicts of Interest: The author declares no conflict of interest.

Contribution of Authors: All authors shared in writing, editing and revising the MS and agree to its publication.

Acknowledgement: We would like to express thanks and gratitude to staff members in Plant Pathology Dept. Sakha Research Station for kind help, faithful effort and advising us during this research.

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