SVU-International Journal of Agricultural Sciences
Volume 4 Issue (1) pp.: 211-219, 2022
Print ISSN 2636-3801 | Online ISSN 2636-381X
Doi: 10.21608/svuijas.2022.226644
RESEARCH ARTICLE

Efficiency of silver nanoparticles synthesized by *Pleurotus ostreatus* to manage fungal garlic cloves rot

Ahmed, Naglaa G. [1], H.A. Gouda [1] and M.A.M. Hussein [2]*

[1] *Plant Pathology Research Institute, Agricultural Research Center, 12619 Giza, Egypt.*
[2] *Plant Pathology Department, Faculty of Agriculture and Natural Resources, Aswan University, 81528 Aswan, Egypt.*

Abstract

Garlic is one of the most important vegetable crops in Egypt. It's naturally infected with fungal strains *Alternaria alternata Aspergillus niger*, *A. flavus*, *Fusarium oxysporum*, *F. proliferatum* and *F. solani* causes decayed garlic cloves. *F. oxysporum* was aggressive pathogen isolated from decayed cloves of the garlic (cv Balady). Silver nanoparticles which synthesized from fungal strain *Pleurotus ostreatus* was used in this study to manage such disease. The effect of three different concentrations of silver nanoparticles (40 ppm, 70 ppm and 100 ppm) was tested to assess their antifungal activities against the fungal causal pathogens. The results indicated that all three concentrations, of the nano-silver had inhibition rates and 100 ppm was the most efficient concentration and had the highest effect on the mycelial growth reduction of pathogenic fungi as well as enhanced the growth of garlic bulbs in *vitro* and in *vivo*.

Keywords: garlic cloves; *Fusarium oxysporum;* silver nanoparticles.

1. Introduction

Garlic (*Allium sativum* L.) is a crop of great importance in Egypt and throughout the world. Garlic has many culinary and medicinal properties (Galvez and Palmero, 2021). It is consumed in different forms. Its secondary metabolites have been demonstrated to have a significant impact on health and to aid in the prevention of a range of human diseases, due to its antioxidant, anti-inflammatory, and lipid-lowering properties (Galvez and Palmero, 2021). Many diseases infecting garlic either pre or post harvesting. Among the diseases, Fungal garlic cloves rot consider among important disease infecting postharvest garlic. Clove rot caused by a variety of fungus species, including *Fusarium culmorum*, *Fusarium proliferatum*, *Fusarium oxysporum*, *Aspergillus orchaceus*, *Aspergillus niger*, *Stemphylium botryosum*, *Botrytis allii*, *Rhizopus stolonifer* and *Pencillium purpurogenum* (Moharam *et al.*, 2013; Elshahawy *et al.*, 2017).

Moreover, *Fusarium* spp. pathogens are known to produce many toxins like fumonisins, fusaric acid and fusaproliferin, these toxins remain chemically stable during food processing and thus they consider a great risk to the health of humans (Mondani *et al.*, 2021; Tonti *et al.*, 2017)

Many strategies have been adopted to control this disease, including biological control, chemical control (Mondani *et al.*, 2021). Recently using Nanoparticles has emerged as new method to control plant diseases. Several reports have mentioned that using nanoparticles is useful method to control garlic gloves rot, and

***Corresponding author:, Mohamed A.M. Hussein**
Email: mmon3m@gmail.com
Received: March 2, 2022; Accepted: March 21, 2022;
Published online: March 23, 2022.

when seedling were treated with varied concentrations of AgNPs, Fresh Weight, root and shoot length, and vigour index were found to positively enhanced compared with non-exposed plants (Sharma *et al.*, 2012; Almutairi and Alharbi, 2015;)

And treatment with silver nanoparticles resulted to a significant increase in plant height, root length, bulbs diameter, plant weight compared with untreated garlic plants (Abd-Elbaky *et al.*, 2021), application of nanoparticles was very effective to control undesirable microbial in garlic (Elsaggan *et al*., 2018).

The objective of this study is to evaluate the efficacy of the biological synthesis of silver nanoparticles (AgNPs) in fungal garlic cloves rot management under greenhouse condition.

2. Material and methods

2.1. Garlic clove pathogens: isolation, purification, and identification

A hundred infected garlic bulbs (cv Balady) were collected from five storage sites located in the Assiut governorate in October 2020. Symptomatic diseased cloves were cut into small segments, soaking for three minutes in 3 percent sodium hypochlorite and then washing with distilled water (3 times) and finally dried with sterilized filter papers. four clove tissue pieces were added to the PDA plate. The emergent fungi were recognized based on their morphological features according to Moubasher (1993), Leslie and Summerell (2006), Ismail *et al*. (2015) and Domsch *et al*. (2015).

2.2. Pathogenicity tests

The pathogenicity of *Alternaria alternata* isolates *Aspergillus niger, A. flavus, Fusarium oxysporum,* and *F. proliferatum* was tested. As previously noted, the surface of healthy cvs Balady cloves was disinfected. Cloves were then cut near the apex to 4.5 mm depth and a diameter of around 1 mm, and artificially wounded then was filled with spore suspension (2.5×10^5 cfu/ml^3). As a control, non-inoculated cloves were used. Cloves that had been inoculated were kept in sealed plastic boxes for 30 days at 25 2°C (Dugan *et al*., 2007). Cloves were evaluated before and after the rotting portion was removed, and rot severity was calculated according Galal *et al*. (2002) using the formula as following:

Percentage of rot severity = (W1 − W2)/ W1 × 100

Where:

W1 is the total weight of the clove, and W2 is the clove's weight after decayed tissue has been removed.

2.3. Synthesis of silver myconanoparticles

Silver nanoparticles were synthesized using a *Pleurotus ostreatus*. One fungal strain agar plug (4-mm diam) was cultivated in a 250-ml Erlenmeyer flask with 100 ml potato dextrose broth (PDB) and incubated for 5 days at 25°C on a rotatory shaker at 120 rpm. The mycelial biomass was removed from the culture and washed with sterile distilled water to eliminate any medium components. By passing the culture extract through Whatman filter paper No. 1, the culture extract was prepared. In a 250 ml Erlenmeyer flask, 50 ml of 1 mM AgNO3 solution was combined with 50 ml of culture filtrate and stirred for 72 hours at 25°C (Basavaraja *et al*., 2008).

2.4. The effect of silver nanoparticles on pathogen mycelial development

Different concentration of silver nanoparticle *i.e*., 40, 70 and 100 ppm were tested with three replicates against isolates of *F. proliferatum*, *A. alternata* and *Aspergillus niger* on potato dextrose agar medium. Before plating in a petri plate (90 x 15 mm), one ml of different AgNPs concentrations were combined with the PDA medium. One agar plug 5 mm of tested fungi was injected at the center of each agar plate and incubated at 28°C for 10 days after medium containing silver nanoparticles was incubated at room temperature (28°C±2) for 48 hours. As a control, agar plates without Ag-nanomaterial were used. Each treatment was replicated three times. When the growth of the fungal mycelia in

control plates approached the petri dish's edge, the growth inhibition rate was computed (Kim *et al*., 2012). The following formula was used to compute the inhibition rate: (Shivapratap *et al*., 1996):

Inhibition percentage = (D1 – D2)/D1x100

D1 is the diameter of the pathogen colony in the control, while D2 is the diameter of the pathogen colony in the treatment.

2.5. Seed germination test

Garlic gloves were dipped in a 5 % sodium hypochlorite solution for 15 minutes to guarantee seed surface sterility before being soaked in a silver nanoparticles solution overnight. The gloves were also soaked in regular household water overnight as a control. The filter paper was then wetted with 5 mL silver nanoparticle solution and placed in petri dishes. The treated gloves were stored on petri plates on filter paper. The petri plates were covered and kept at room temperature for incubation. Germination was stopped after 36 hours, and the germination percentage was calculated.

2.6. Evaluation the effect of silver nanoparticles on garlic fungal incidence in greenhouse

During the 2020-2021 growth seasons, three concentrations of silver nanoparticles were evaluated against pathogenic fungus in a greenhouse setting. Two weeks before adding Ag-nanoparticles, sterilized pots (25-cm diam.) were filled with 3 kg sterilized sandy loamy soil and mixed with pathogenic fungi, *Alternaria alternata, Aspergillus niger, A. flavus, Fusarium oxysporum, Fusarium proliferatum,* and *Fusarium solani* at a rate of 3% (w/w). Cloves were either soaked for 5 min in AgNPs at 40, 70, or 100 ppm concentrations prior to sowing. Only the control pots, which were filled with sterilized soil and infested with tested fungi, were used. Each pot had five gloves sown in it, and three pots were used as duplicates. The percentage of symptomatic of cloves in the sample in respect to the total cloves was used to calculate the incidence of clove rot. According to Palmero *et al*. (2013) each bulb's disease symptoms were categorized into four arbitrary groups. The following formula was used to determine the severity of the rot:

RS=(N1×0) +(N2×1) +(N3×2) +(N4×3)/Number of total cloves

N1 indicates no symptoms; N2 indicates rotted patches; N3 indicates a 10-50 percent rotted clove; and N4 indicates a completely rotted clove.

2.7. Statically analysis

Split plot was designed, SAS Institute Inc., used for (ANOVA) analysis of variance. Least significant differences test at P 0.05 was used to compare the means (Gomez and Gomez 1984).

3. Results and discussion

3.1. Isolation and identification the fungi associated rotten garlic cloves

Isolation was obtained from the decaying tissues of the tested cv. Balady from five storage sites located in the Assiut governorate yielded five species of fungal belonging to three genera, *Alternaria alternata Aspergillus niger, A. flavus, F. oxysporum, F. proliferatum* and *F. solani* and these isolates were identified based on their colony characteristics and morphological features.

3.2. Pathogenicity tests

Tests of pathogenicity indicated that *F. oxysporum* was highly aggressive to garlic that has been wounded balady cloves by (75% rotted tissue) followed by *Fusarium proliferatum* (60.2%) and *Alternaria alternata* (60%). Other fungal species *Aspergillus niger* (35.1%), *A. flavus* (23%) and *Fusarium solani* (38.1) were less aggressive. According to Moharam *et al*. (2013), *F. oxysporum* caused the most severe clove rot. While Elshahawy *et al*., (2017) isolated *Aspergillus ochraceus, Aspergillus niger, Penicillium purpurogenum, Penicillium purpurogenum, Penicillium purpurogenum, F. proliferatum, Botrytis allii, F. oxysporum, S. botryosum, Penicillium purpurogenum,* and *R.*

stolonifer from rotting garlic cloves, with *Fusarium proliferatum* having a high prevalence of rot of garlic clove during the storage period. These results are in agreement with previous studies that found the most prevalent fungal pathogens causing cloves rot in garlic are *F. culmorum, F. proliferatum, Fusarium oxysporum, Aspergillus orchaceus, Aspergillus niger, Stemphylium botryosum, Botrytis allii, Rhizopus stolonifer and Pencillium purpurogenum* (Moharam *et al*., 2013; Elshahawy *et al*., 2017).

Table 1. Pathogenic capability of the isolated fungi isolated from rotted cloves Balady garlic cultivar.

Isolated fungi	rotted tissue (%)
Alternaria alternate	60.0
Aspergillus flavus	23.0
Aspergillus niger	35.1
Fusarium oxysporum	75.0
Fusarium proliferatum	60.2
Fusarium solani	38.1
LSD 0.05	6.12

3.3. Silver myconanoparticles synthesis

Silver nanoparticles were synthesized using a *Pleurotus ostreatus* isolate. The synthesis of myconanoparticles of Ag in the reaction mixture is indicated by the emergence of a brownish colour (Figure 1). The synthesis of AgNPs was indicated by the change in colour of the culture filtrates due to the reduction of silver ions, which ranged from pale yellow to brown (Bhainsa and D'Souza, 2006). When exposed to comparable conditions, 1 mM AgNO3, which was utilized as a control, did not change colour. Several studies reported production Nanoparticles NPs using microorganisms and plants (Iravani, 2011; Korbekandi *et al*., 2016), nanoparticle size under transmission electron microscope TEM was determined in from 20.4 to 30.8 nm as shown in figure 2, This finding is match the results obtained by many authors (Massoud *et al.*, 2018; Avan *et al.*, 2018)

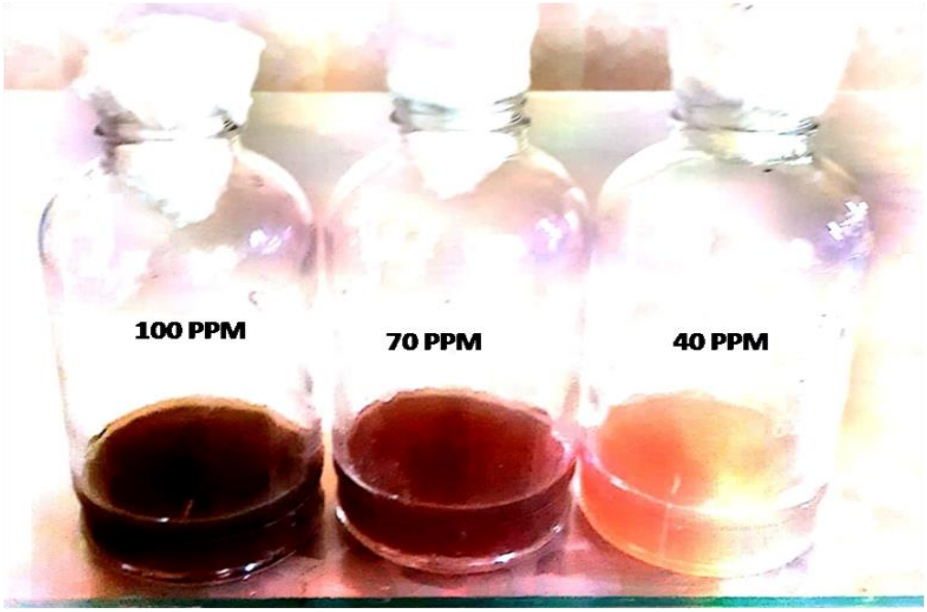

Figure 1. Filtrate of the *Pleurotus ostreatus* in aqueous solution of AgNO3 after 3 days of reaction.

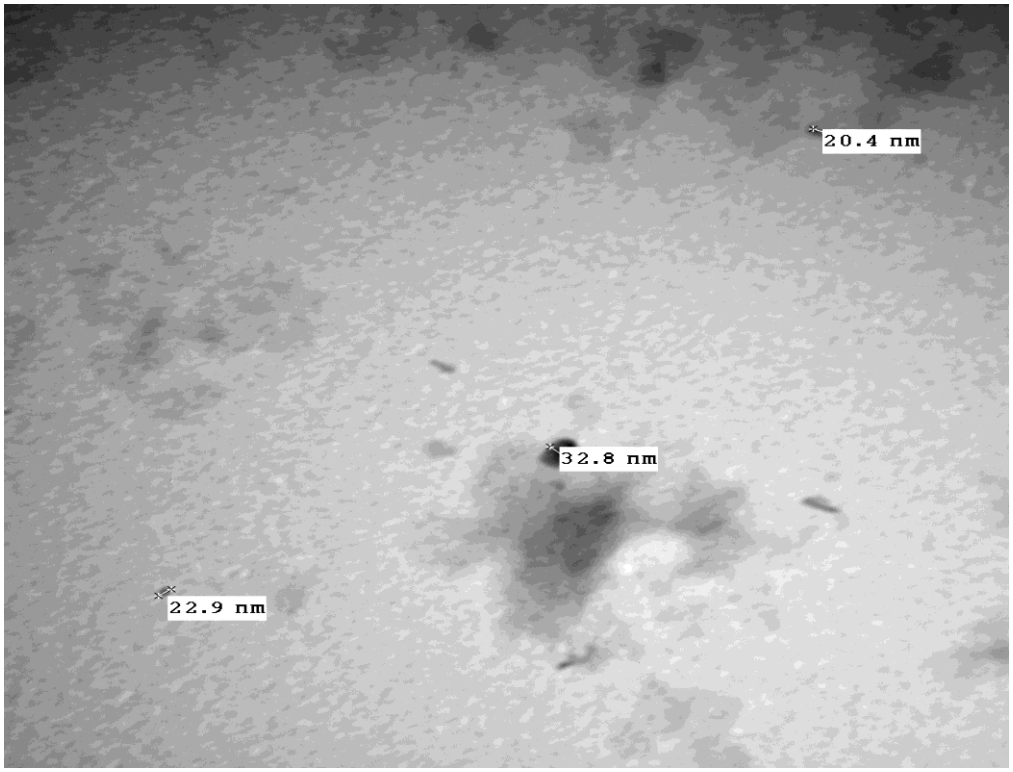

Figure 2: anoparticle size under transmission electron microscope TEM.

3.4. The effect of AgNPs nanoparticles on pathogens mycelial growth

The findings revealed that all AgNP concentrations synthesized by *Pleurotus ostreatus* inhibited the mycelial growth of the pathogenic fungi. The inhibition rate of *F. oxysporum* at 40 ppm was 71.6%, 70 ppm was 72.2 % and at 100 ppm was 80 % compared to control. The reduction rate of *Alternaria alternata* at 40 ppm was 55.5 %, 70 ppm was 77.7 % and at 100 ppm was 84.4 % compared to control while the growth inhibition of *Aspergillus niger* was 79 %, 70 ppm was 79.4 % and at 100 ppm was 83 % compared to control (Figure 3). According to Abd-Elbaky *et al.*(2021) a 100 percent concentration of silver nanoparticles had the greatest effect on the growth of *F. oxysporum* f.sp. *cepae* mycelia, which causes onion basal-rot. *In vitro*, Jung *et al*., (2010) found that all nano-silver liquid concentrations (1, 3, 5, 7, 10, 25, 50 ppm and 100 ppm) had more than 90% inhibition rates against *Sclerotium cepivorum*, the pathogen that causes green onion white rot. These results are in harmony with other studies as showed severely damaged cell walls, leakage of cytoplasmic and nucleic contents, and a swelling structure, all of which contributed to bacterial mortality (Elbeshehy *et. al*., 2015) In another study, biosynthesized AgNPs have antifungal activity against *Fusarium graminearum* as caused hyphae deformation and cell wall damage (Ibrahim, et al., 2020). Also, nanoparticles have similar effects on fungal phytopathogenic strains of *A. alternata*, *Trichosporon asahii* and *Botrytis cinera* (Xia *et al*., 2015).

Table 2. Silver nanoparticles growth inhibition percentage on the three fungal rot causal pathogens of garlic gloves.

AgNPS type	AgNPs concentration		
	40 ppm	70 ppm	100 ppm
Alternaria alternata	55.5	77.7	84.4
Aspergillus niger	79.0	79.4	83.0
Fusarium oxysporum	71.6	72.2	80.0
LSD 0.05	A Fungal strains 9.08	B AgNPs Concentrations 7.25	AB 1.22

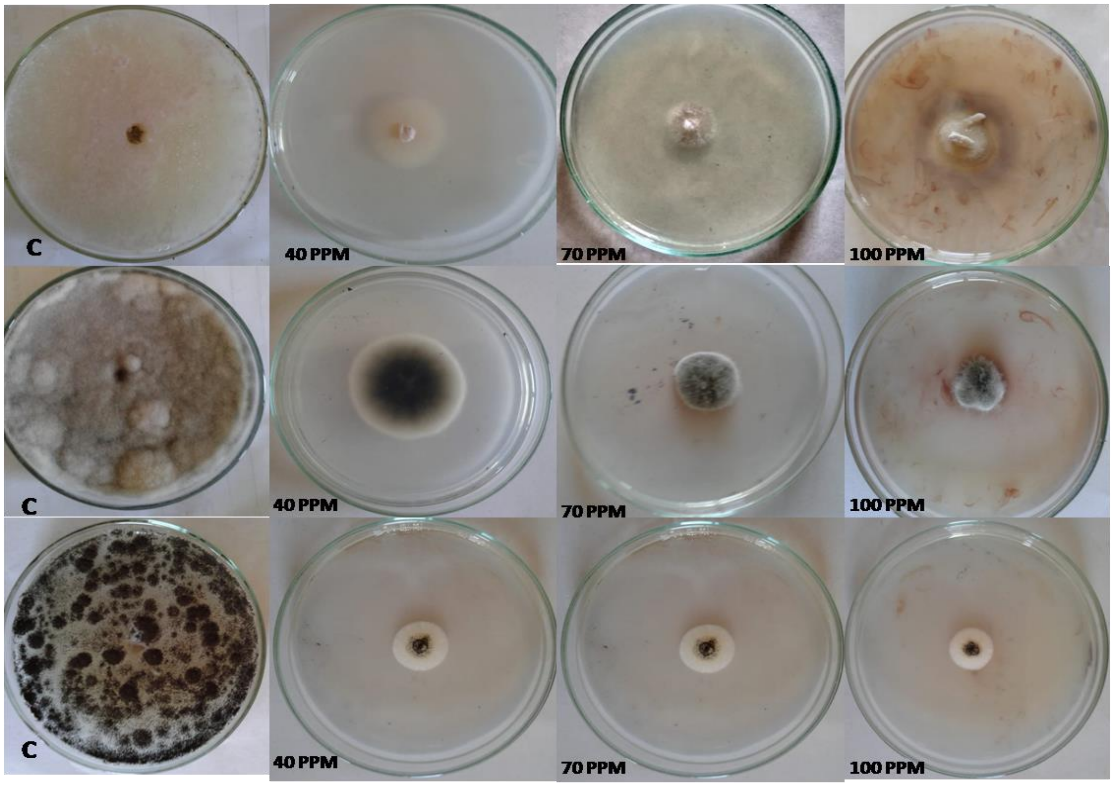

Figure 3. Effects of different concentrations of *Pleurotus ostreatus* synthesized silver nanoparticles on fungal pathogens (from top to down, *Fusarium oxysporum*, *Alternaria alternata* and *Aspergillus niger*).

3.5. Seed germination test

The results indicate that the highest germination percentage value (85.3%) and healthy seedling (88%) for garlic cloves which was observed when exposure to 100 ppm of AgNPs. Same results seen by (Savithramma *et al.*, 2012; Almutairi and Alharbi, 2015; Abd-Elbaky *et al*., 2021) that AgNPs increased germination value of seeds of crop plants significantly.

3.6. Greenhouse assessment of garlic's fungal pathogens response to silver nanoparticles

Under greenhouse conditions throughout the 2020 and 2021 growing seasons, the impact of soil that treated by using varied concentrations of Ag-nanoparticles on incidence of balady garlic rot fungal pathogens was studied. The data in table (3) showed that the treatment of soil with Ag-nanoparticles not only reduced the disease severity of fungal pathogenic garlic rot but also enhance the rate of growth of gloves and the data indicate that 100 ppm of AgNPs more effective than the other concentrations in both reduction of pathogenic fungi and also the growth rate of gloves seedling. Ag-nanoparticles had antimicrobial and antiviral agents (Burdusel *et al.*, 2018). According to Abd-Elbaky et al., (2021), the tested nanoparticle concentrations of 25, 33, 50, and 100 percent caused different values of antifungal effect on Fusarium mycelial growth of onion basal-rot, with the crude

concentration (100 percent) of silver nanoparticles having the greatest effect on *Fusarium oxysporum f.sp. cepae* mycelial growth. These results are in partial agreement with results obtained by Elamawi and Al-Harbi (2014) who found that nanoparticles of silver can effectively control *Fusarium* spp. pathogen.

Table 3. Effect of *Pleurotus ostreatus* nanoparticles to control garlic cloves rot

AgNPS type	AgNPs concentration							
	0 ppm		40 ppm		70 ppm		100 ppm	
	% Germination	% Healthy seedling	% Germination	% Healthy seedling	% Germination	% Healthy seedling	% Germination	% Healthy seedling
Alternaria alternata	50.8	54.6	65.4	66.4	68.0	70.3	88.2	90.4
A. niger	38.6	40.8	76.5	80.6	90.4	92.6	100	100
F. oxysporum	52.3	55.0	60.3	65.0	70.2	72.3	85.3	88.0
Control	100	100	100	100	100	100	100	100

LSD 0.05 A Fungal strains 7.68 B AgNPs Concentrations 5.42 AB 2.63

Table 4. Inhibition of growth rate (%) of fungal pathogens by silver nanoparticles various concentrations synthesized by *Pleurotus ostreatus*

Strains	AgNPs concentration					
	40 ppm		70 ppm		100 ppm	
	% reduction	% growth rate	% reduction	% growth rate	% reduction	% growth rate
Alternaria alternata	50.2	40	67	60	80.2	82.1
A. niger	60.3	55	63.2	66.4	85	75
F. oxysporum	65.3	55	63.4	60.3	75.2	80
Control	100	100	100	100	100	100

4. Conclusion

Alternaria alternata, *Aspergillus niger*, *A. flavus*, *Fusarium oxysporum*, *F. proliferatum* and *F. solani* are an important causal pathogens of garlic cloves rot. *F. oxysporum* was the most aggressive pathogen found in rotting garlic cloves. The results indicated that all three concentrations of silver nanoparticles which synthesized from *Pleurotus ostreatus*, had an inhibition effect on the causal pathogens. The most effective concentration was 100 ppm, which had the great effect to reduce pathogenic fungi growth *in vitro* and *in vivo*. This is a promising outcome achievement by using bio synthesized silver nanoparticles that could be used as a safe fungicide alternative.

Authors' Contributions

All authors are contributed equally in this research.

Funding

There is no fund in this research.

Institutional Review Board Statement

All Institutional Review Board Statement are confirmed and approved.

Data Availability Statement

Data presented in this study are available on fair request from the respective author.

Ethics Approval and Consent to Participate

This work carried out at Plant Pathology department and followed all the department instructions.

Consent for Publication

Not applicable.

Conflicts of Interest

The authors declare no conflict of interest

5. Reference

Abd-Elbaky, A.A., EL-Nahas, S.E., Hassan, M.E., Desoukey, A.F. (2021). 'Effect of Silver Nanoparticles on Fusarium Basal-Rot of Onion', *International Journal of Scientific and Engineering Research*, 12 (4), pp. 481.

Almutairi, Z.M., Alharbi, A. (2015). 'Effect of silver nanoparticles on seed germination of crop plants', *J. adv. Agric.*, 4(1), pp. 283-288.

Avan, E.D., Quadry, R.O., Ikenna-Ossai, C.N., Okolie, N.P. (2018). 'Effects of Annona muricata biofunctionalized gold nanoparticles on erythrocyte osmotic fragility and hematological profile in rat model', *Covenant J. Phys. Life Sci.*, 2(1), pp. 33–45.

Basavaraja, S., Balaji, S.D., Lagashetty, A., Rajasab, A.H., Venkataraman, A. (2008). 'Extracellular biosynthesis of silver nanoparticles using the fungus *Fusarium semitectum'*, *Materials Research Bulletin*, 43(5), pp. 1164-1170.

Bhainsa, K.C., D'souza, S.F. (2006). 'Extracellular biosynthesis of silver nanoparticles using the fungus *Aspergillus fumigatus*', *Colloids and surfaces B: Biointerfaces*, 47(2), pp. 160-164.

Burdusel, A.C., Gherasim, O., Grumezescu, A.M., Mogoantă, L., Ficai, A., Andronescu, E. (2018). 'Biomedical applications of silver nanoparticles: an up-to-date overview', *Nanomaterials*, 8(9), pp. 681.

Dennis, C., Webster, J. (1971). 'Antagonistic properties of species-groups of *Trichoderma*: I. Production of non-volatile antibiotics', *Transactions of the British Mycological Society*, 57(1), pp. 25-IN3.

Domsch, K.H., Gams, W., Anderson, T.H. (2007). *'Compendium of soil fungi. 2nd edition'*, IHW-Verlag, Eching.

Dugan, F.M., Hellier, B.C., Lupien, S.L. (2007). 'Pathogenic fungi in garlic seed cloves from the United States and China, and efficacy of fungicides against pathogens in garlic germplasm in Washington State', *Journal of Phytopathology*, 155(7-8), pp. 437-445.

Elamawi, R.M., Al-Harbi, R.E. (2014). 'Effect of biosynthesized silver nanoparticles on *Fusarium oxysporum* fungus the cause of seed rot disease of faba bean, tomato and barley', *Journal of Plant Protection and Pathology*, 5(2), pp. 225-237.

Elbeshehy, E.K., Elazzazy, A.M., Aggelis, G. (2015). 'Silver nanoparticles synthesis mediated by new isolates of *Bacillus* spp., nanoparticle characterization and their activity against Bean Yellow Mosaic Virus and human pathogens', *Frontiers in microbiology*, 6, pp. 453.

Elsaggan, M., Khater, M.S., El Desouky, G. (2018). 'Effect of spraying by nitrogen loaded on chitosan nanoparticles on growth and productivity of garlic under sandy soil conditions', *Egyptian Journal of Desert Research*, 68(1), pp. 135-155.

Elshahawy, I.E., Saied, N.M., Morsy, A.A. (2017). '*Fusarium proliferatum*, the main cause of clove rot during storage, reduces clove germination and causes wilt of established garlic plants', *Journal of plant pathology*, pp. 85-93.

Galal, A., Abdel Gawad, T., El Bana, A. (2002). 'Post-harvest decay of garlic cloves caused by *Bacillus polymyxa* and *Fusarium moniliforme*', *Egyptian Journal of Microbiology*, 37(1), pp. 71-88.

Galvez, L., Palmero, D. (2021). 'Incidence and Etiology of Postharvest Fungal Diseases Associated with Bulb Rot in Garlic (*Alllium sativum*) in Spain', *Foods*, 10(5), pp. 1063.

Gomez, K.A., Gomez, A.A. (1984). *'Statistical Procedures for Agricultural Research'*, John Wiley and Sons, 704 pp.

Ibrahim, E., Zhang, M., Zhang, Y., Hossain, A., Qiu, W., Chen, Y., Wang, Y., Wu, W., Sun, G., Li, B. (2020). 'Green-synthesization of silver nanoparticles using endophytic bacteria isolated from garlic and its antifungal activity against wheat Fusarium head blight pathogen *Fusarium graminearum*', *Nanomaterials*, 10(2), pp. 219.

Iravani, S. (2011). 'Green synthesis of metal nanoparticles using plants', *Green Chemistry*, 13(10), pp. 2638-2650.

Ismail, M.A., Abdel-Hafez, S.I.I., Hussein, N.A., Abdel-Hameed, N.A. (2015). *'Contributions*

to the genus Fusarium in Egypt with dichotomous keys for identification of species', Suchy Las, Poland. Publisher Tomasz M. Karpiński, 175 pp.

Kim, D.H., Kim, H.S., Choi, C.W., Kim, E.K., Kim, B.I., Choi, J.H. (2012). 'Risk factors for pulmonary artery hypertension in preterm infants with moderate or severe bronchopulmonary dysplasia', *Neonatology*, 101(1), pp. 40-46.

Korbekandi, H., Mohseni, S., Mardani Jouneghani, R., Pourhossein, M., Iravani, S. (2016). 'Biosynthesis of silver nanoparticles using *Saccharomyces cerevisiae', Artificial cells, nanomedicine, and biotechnology*, 44(1), pp. 235-239.

Leslie, J.F., Summerell B.A. (2006). '*Fusarium* laboratory workshops a recent history', *Mycotoxin Research*, 22 (2), pp. 73-74.

Massoud, K., Nafiseh, Jafar, (2018). 'Biosynthesis of gold nanoparticles using aqueous extract of stem of Periploca aphylla', *Plant Nano-struct.* 8(2), pp. 152–158.

Moharam, M.H., Farrag, E.S., Mohamed, M.D. (2013). 'Pathogenic fungi in garlic seed cloves and first report of *Fusarium proliferatum* causing cloves rot of stored bulbs in upper Egypt', *Archives of Phytopathology and Plant Protection*, 46 (17), pp. 2096-2103.

Mondani, L., Chiusa, G., Battilani, P. (2021). 'Chemical and biological control of *Fusarium* species involved in garlic dry rot at early crop stages', *European Journal of Plant Pathology*, 160(3), pp 575-587.

Moubasher, A.H. (1993). *'Soil fungi of Qatar and other Arab Countries.',* The Scientific and Applied Research Centre, University of Qatar, 566 pp.

Palmero D.L., Galvez L.P., Garcia M.D., Gil J.S., Benito S.S, (2013). 'The effects of storage duration, temperature and cultivar on the severity of garlic clove rot caused by *Fusarium proliferatum*', *Postharvest Biology and Technology*, 78, pp. 34-39.

Savithramma, N., Ankanna, S., Bhumi, G. (2012). 'Effect of nanoparticles on seed germination and seedling growth of *Boswellia ovalifoliolata* an endemic and endangered medicinal tree taxon', *Nano Vision*, 2(1, 2 and 3), pp. 61-68.

Sharma, R.K., Patel, S., Pargaien, K.C. (2012). 'Synthesis, characterization and properties of Mn-doped ZnO nanocrystals', *Advances in Natural Sciences: Nanoscience and Nanotechnology*, 3(3), pp. 035005.

Shivapratap, H.R., Philip, T. and Sharma, D.D., (1996). "In vitro antagonism of *Trichoderma* species against mulberry leaf spot pathogen, Cercospora moricola', *Indian Journal of Sericulture*, 35(2), pp. 107-110.

Taga, M.S., Miller, E.E., Pratt, D.E., (1984). 'Chia seeds as a source of natural lipid antioxidants', *Journal of the American Oil Chemists' Society*, 61(5), pp. 928-931.

Tonti, S., Mandrioli, M., Nipoti, P., Pisi, A., Toschi, T. G., Prodi, A. (2017). 'Detection of fumonisins in fresh and dehydrated commercial garlic', *Journal of Agricultural and Food Chemistry*, 65(32), pp. 7000-7005.

Xia, Z.K., Ma, Q.H., Li, S.Y., Zhang, D.Q., Cong, L., Tian, Y.L., Yang, R.Y. (2016). 'The antifungal effect of silver nanoparticles on *Trichosporon asahii*', *Journal of microbiology, immunology and infection*, 49(2), pp. 182-188.