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Antifungal activity of cassia plant extract and silica nanoparticals (NPs) against *Fusarium oxysprum* and *Rhizoctonia solani*

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Abstract

The activity of Ridomil gold plus, *Cassianodosa* extract, bio silica NPs and chem silica NPs investigated against *Fusarium. oxysporum and Rhizoctonia solani.* The results indicated that all tested compounds decreased the growth of *F.oxysporum and R. solani with* increased concentrations. *F.oxysporum* more sensitive to all tested compounds except chem silica NPs. The fungicide Ridomil is the most effective against both fungi followed by *C. nodosa* extract and bio silica NPs. In contrast, the chem silica NPs less effective in inhibiting the growth of *F.oxysporum* and *R. solani*. The fungicide Ridomil at 1000 ppm resulted in the emergence of irregular and unstable forms in fragments. The use of *C. nodosa* extract at 2100 ppm led to the appearance of deformations and degradation of the fragile filaments. While there was clear decomposition and deformation in the fragments from the use of bio silica NPs in the microscopic field of 400, these distortions increased in the magnified field 1000_x with the presence of lacunae in the hypo filaments of both *F. oxysporum* and *R. solani.*

Keywords: *Fusarium oxysporum*, *Rhizoctonia solani*, Ridomil, Plant Extracts, Nanoparticles, Growth, Inhibition

Introduction

Plants subjected to many biotic stresses that lead to severe losses in yield, such as fungi. Some of these fungi attack the aerial parts of plants such as leaves, stems, flowers and fruits and another type of fungi that attacks the non-aerial parts at the soil surface such as roots and stems. In most cases, it is difficult to notice these fungi except in critical stages of infection. *F. oxysporum* and *R. solani* are both of the most dangerous fungal diseases that affect large numbers of crops caused losses reached 20: 100% of the yield. *F.oxysporum* is one of soil borne fungus affecting many plants by causing fusarium wilt. There are many ways to spread the fusarium such as, tools of farm, transferring of infection planting materials, also water and air can play role in spread fungi Gamliel [1]. *R. solani* one of the fungi has ability to infect a wide host range *R. solani* Can attack plant seeds at the soil and also can infect leaves ,steams, roots and pods *R. solani* is causing damping off or preventing seeds to germinate . In some case it can kill seedling after emerge from the soil. The seedling is sensitive to *R. solani* in early stages Wibberg [2]. There are many

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ways to combat *F. oxysporum* and *R. solani*. Chemical fungicides one of these ways to control the infection of fungal diseases. The pesticide Ridomil stands out as one of the most widespread fungicides in Egypt. Ridomil is fungicide with systemic function. It is not a specialized pesticide on fusarium and rhizoctonia but many studies have investigated the possibility of using Ridomil eliminate on of fungi. Ullah [3] pointed that Ridomil gold is the best fungicides against *F.oxysporum*, *R.solani* and caused completely inhibited growth of *R. solani*. Fungicides give effective in reducing the spread of fungal diseases, but they cause great environmental hazardous. Many studies evaluate the activity of plant extracts in reducing the spread of fungal diseases. Yadav [4] found that extracts of *Cassianodosa* leaves effective against *Aspergillusflavus*, *Aspergillus niger*, *Fusarium moniliformae* and *Rhizoctonia bataticola.* Nanotechnology considered one of the latest sciences and the most used in all fields of applied sciences, especially agricultural sciences. Nanotechnology is an environmentally friendly that has gained popularity recently as a safe alternative to pesticides and chemical fertilizers. In this study, investigated the efficiency of bio silica NPs and chem silica NPs against *F. oxysporum* and *R. solani*. Park [5] found that, silica-silver NPs was effective against *Botrytis cinerea*, *R. solani* and *Colletotrichumgloeosporioides*. Therefore, The aim of this study evaluate the efficiency of bio silica NPs, chem silica NPs and *C. nodosa* extract as safe alternative to chemical fungicides Ridomil in controlling *F. oxysporum* and *R. solani* at laboratory condition.

Experimental Technique:

The present study carried out at Itay El-barud Agriculture Research Station laboratory

Chemical used

*****Fungicide: Ridomil gold plus 71.5 WP (Mefenoxam 2.5% + copper oxychloride 69%)-Syngenta.

Chemical name:

a) Methyl N-(methoxyacetyl)-N-(2,6-xylyl)-D-alaninate; methyl(R)-2-[(2,6-dimethylphenyl) methoxyacetyl]amino]propionate.
b) Dicopper chloride trihydroxide.

**Cassianodosa* extract (crude).

*Bio silica NPs: obtained from Research Center of Kafr El-Sheikh.

*Chem silica NPs: obtained from Research Center of Kafr El-Sheikh.

Fungi used:

Fusarium oxysporum, Rhizoctonia solani obtained from Plant Pathology Institute.

Extraction of *C.nodosa*:

Stem bark of *C. nodosa* obtained from the Faculty of Agriculture, Tanta University. *C. nodosa* identified by Prof. Dr. Mohammed Ibrahim Fotoh, Professor of Ornamental Horticulture and Landscape Design, Faculty of Agriculture, Tanta University, Tanta, Egypt. Stem bark were air dried and powdered. The powder enter the oven to remove moisture the plant was extracted by using methanol 95% by using ultrasonic device after that extracts were concentrated at vacuum. The extract was stored in brown bottle in the freezer until used.

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In vitro assay:

Ridomil at concentrations from 50 to 1000 ppm, *Cassianodosa* extract at concentrations from 50 to 2100 ppm, bio silica and chem silica NPs at concentration from 10 to 200 ppm investigated against *F. oxysporum* and *R. solani*. These compounds added to PDA media before solidification into petri dishes (9 cm). The fungi discs (0.5 cm) added into the medal of all petri dishes. Control without these compounds. All of petri dishes incubated at 28 $\pm$ 2a. When control completely with colonized fungi. Then the growth of mycelium of fungi measured in all treatment. The percentages of inhibition calculated according to the method **of** Taisan [6] by using this formula

MGI%

R is the radial growth of fungi in control plate

r is the radial growth of fungi in treatment plate

Examined the effect of different antifungal treatments in *F.oxysporum* and *R. solani* cells at electronic microscope.

The changes in *F. oxysporum* and *R. solani* cells at different antifungal treatments characterized by scanning electron microscopic (SEM) observation (JOEL, JSM 5300) with high resolution at an accelerating voltage of 120 Kev. An aliquot of each material coated on copper grid and scanned for its size and shape. Where, the tested cellssubjected to X-ray Electron Dispersive Analysis (EDA) using an X-ray Oxford detector unit (model 6697, England).

Characterization of NPs

The prepared NPs subjected for Scanning Electron Microscope (SEM) (Fig 1). Biosynthesized SiO_2NPs showed amorphous particles with irregular shapes ranged from 16.67 to 27.78 nm. On the other hand, chemically synthesized silica NPs showed nearly spherical shape in the range 27.8-59.7 nm

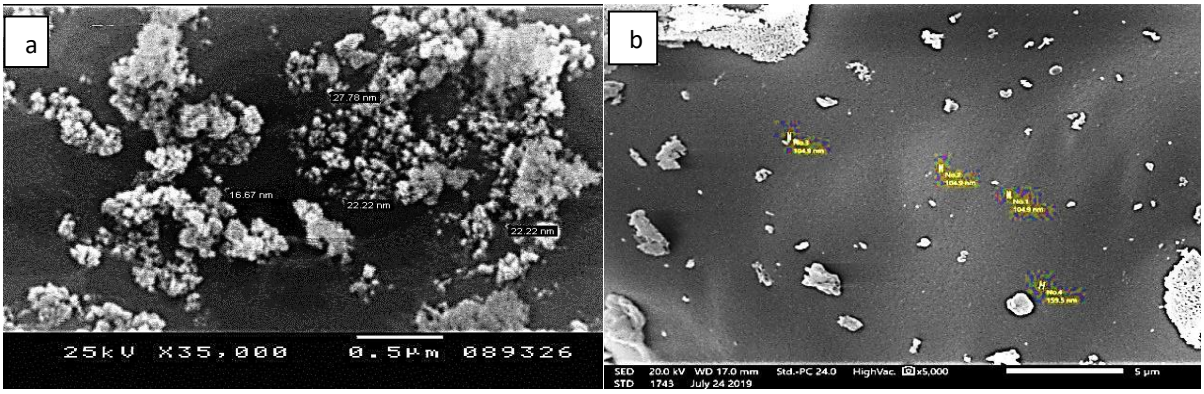


Fig 1: SEM images of (a) bio-synthesized silica NPs and (b) chemically-synthesized silica NPs that visualized at 35000 and 33000$_X$, respectively.

Statistical analysis:

The experiment was set up in a complete randomized design. The obtained data analyzed by using ANOVA. Analysis of variance (ANOVA) using cost at 6.3111 software 1998-2005 and Duncan, s multiple range test at P < 0.05 level used for means separation Winer [7].

Results and discussion

Effect of fungicide Ridomil against *F. oxysporum* and *R. solani*.

The results in Table 1 indicated that both *F. oxysporum* and *R. solani* growth gradually decreases with the increase of Ridomil concentrations. *F. oxysporum* was more susceptible to Ridomil than *R. solani* where *F. oxysporum* scored lower growth 4.26 cm than *R.solani* 5.15 cm at all concentrations. *F. oxysporum* growth were 9.00 cm at the control treatment then decreased to 5.37 cm at 50 ppm Ridomil with a growth inhibition of 40.37% then reached their lowest value 1.77 cm at 1000 ppm of Ridomil with a growth inhibition of 80.37%. With respect to *R. solani* the obtained data indicated that *R. solani* growth was 9.00 at the control treatment and reduced to 7.62 cm at 50 ppm of Ridomil with a growth inhibition of 15.37% then reached to 2.42 cm with a growth inhibition of 73.15% at 1000 ppm of Ridomil. In general, the mean growth rate of the both tested fungi species was 9.00 cm in the control treatment and gradually decreased with the increase of Ridomil concentration. The mean average of growth was 6.49 cm at 50 ppm with a growth inhibition of 27.87% and reduced to 4.09 cm with a growth inhibition of 54.54% at 500 ppm. The lowest value at 1000 ppm of Ridomil was 2.09 cm this reduce in growth rate associated with the highest growth inhibition 76.76%. According to the annual report of food and agriculture organization of the united nation (FAO STAT, 2018), the yield losses of field and vegetable crops due to different fungi pathogenic ranged between 15:22% from the total yield productions caused a large loss in the economic return of farmers. In this study all Ridomil concentrations showed a large inhibitory of both *F. oxysporum* and *R. solani* colony diameters. These findings are in the same way with those of Fravel [8] who found that Ridomil Gold Bravo significantly reduced the size of colon, *F. oxysporum* at 50 ppm or greater while Njiru [9] found a higher inhibition on fungal hyphal growth and Hyphae incorporated on media with Ortiva had larger diameter. According to the study of Saad [10] revealed that both Ridomil gold MZgold MTridexxTridexT high inhibition of *A. solani* reached a complete reduction (100%) of growth mycelial at 400 ppm as compared to other fungicides. In addition, Gade [11] indicated that 0.2% of Ridomil had high effect in redaction the growth of *F. solani*, which cause rot of rhizome on ginger. In the study of Banu [12] 0.20% of mancozeb resulted in the maximum inhibition of *F. oxysporum* (62.2%) followed by 0.15% which recorded 57.7% inhibition.

Table 1: Effect of Ridomil against in *F. oxysporum* and *R. solani*.

Conc. (ppm)	*F. oxysporum*		*R. solani*		Mean	
	G. mean (cm)	GI%	G. mean (cm)	GI%	G. mean (cm)	GI% mean
Con	9.00[a]	0.00[h]	9.00[a]	0.00[f]	9.00[a]	0.00[i]
50	5.37[b]	40.37[g]	7.62[ab]	15.37[ef]	6.49[b]	27.87[h]
100	5.30[bc]	41.11[fg]	6.87[b]	23.67[e]	6.09[bc]	32.39[gh]
200	5.16[bc]	42.67[fg]	6.45[b]	28.33[e]	5.81[bc]	35.50[gh]
300	4.82[bc]	46.48[fg]	6.25[bc]	30.56[de]	5.53[cd]	38.52[fg]
400	4.55[cd]	49.44[ef]	4.97[cd]	44.81[cd]	4.76[de]	47.13[ef]
500	3.88[de]	56.85[de]	4.30[de]	52.22[bc]	4.09[ef]	54.54[de]
600	3.45[ef]	61.67[cd]	3.83[de]	57.41[bc]	3.64[fg]	59.54[cd]
700	3.05[fg]	66.11[bc]	3.65[def]	59.44[abc]	3.35[fgh]	62.78[bcd]
800	2.47[gh]	72.59[ab]	3.48[ef]	61.30[ab]	2.98[gh]	66.94[bc]
900	2.25[h]	75.00[a]	2.93[ef]	67.41[ab]	2.59[hi]	71.20[ab]
1000	1.77[h]	80.37[a]	2.42[f]	73.15[a]	2.09[i]	76.76[a]
LSD 5%	0.79	8.78	1.41	15.67	0.79	8.73
Mean	4.26[b]	52.72[a]	5.15[a]	42.81[b]		

G. mean: mean of growth, GI%: percentage of growth inhibition.

Effect of *C. nodosa* extract against *F. oxysporum* and *R. solani*.

The obtained data in Table 2 revealed that both *F. oxysporum* and *R. solani* growth negatively associated with *C. nodosa* concentrations where the growth of both fungus gradually decreases with the increase of concentrations of *C. nodosa* extract. *F. oxysporum* was more sensitive to *C. nodosa* extract than *R. solani* where *F. oxysporum* scored lower mean growth 3.95 cm than *R.solani* 5.73 cm at all concentrations with mean growth inhibition of 56.13% and 36.30% for the both fungus, respectively.

The results indicated that *F. oxysporum*growth were 9.00 cm at the control while decreased to 5.73 cm with growth inhibition of 36.37% at 50 ppm and reached their lowest value 1.82 cm at 2100 ppm with a growth inhibition of 79.81%. With regard to *R. solani* the obtained data showed that *R. solani* growth was 9.00 cm at the control and reduced to 7.85 cm with growth inhibition of 12.78% at 50 ppm and decrease again until reached to the lowest value 2.93 cm with growth inhibition of 67.48% at 2100 ppm of *C. nodosa* extract. The results also confirmed that the mean growth rate of the both tested fungi species was 9.00 cm in the control treatment and gradually decreased with the increase of *C. nodosa* extract concentrations. Across the both fungi, the average growth of both *F. oxysporum* and *R. solani*was 6.79 cm at 50 ppm of *C. nodosa* extract with a growth inhibition of 24.57% and the average growth of the both fungi reduced to 3.99 cm with a growth inhibition of 55.69% at 900 ppm. The average growth of the both fungus reached to the lowest value at 2100 ppm of *C. nodosa* extract 2.37 cm this reduce in growth rate associated with the highest growth inhibition 73.65% . The prevailing trend in the world during the last both decades has become to reduce use of pesticides in controlling pests due to their harmful effect on the environment and public health. Our findings are in agree with those of; Bowers [13] who, found that commercial formulations of cassia extract had high efficacy to control muskmelon wilt caused by *F. oxysporum f. sp. melonis*. In addition, Mogle [14] reported that, *C. siamea* was highly inhibitory for *F. oxysporum* and *R. solani*. In the same line, Yadav [4] indicated that *C. nodosa* extracted by various organic solvents had a great inhibitory against *F. moniliformae* and *R. bataticola*. Ibrahim [15] showed that, the maximum antifungal activity noticed in different extracts of *C. fistula* against the three fungal strains*; F. oxysporum, Fusarium solani* and *R. solani* with significant differences comparing with the other extracts. Finally, Abid [16] indicated that *A. squamosa, O. basilicum* and *C. fistula* leaves, stem and fruit powder were effective at the concentration of 1% for the enhancement of growth of okra.

Table 2: Effect of *C. nodosa* extract against *F. oxysporum* and *R. solani*.

	F. oxysporum		*R. solani*		Mean	
Conc. (ppm)	G. mean (cm)	GI%	G. mean (cm)	GI%	G. mean (cm)	GI% mean
Con	9.00^{a}	0.00^{e}	9.00^{a}	0.00^{e}	9.00^{a}	0.00^{f}
50	5.73^{b}	36.37^{d}	7.85^{b}	12.78^{d}	6.79^{b}	24.57^{e}
100	5.53^{b}	38.59^{d}	7.40^{bc}	17.78^{cd}	6.46^{b}	28.19^{e}
300	3.80^{c}	57.78^{c}	7.15^{bc}	20.56^{cd}	5.48^{c}	39.17^{d}
600	3.47^{cd}	61.48^{bc}	6.83^{c}	24.15^{c}	5.15^{c}	42.81^{d}
900	2.80^{cde}	68.89^{abc}	5.18^{d}	42.48^{b}	3.99^{d}	55.69^{c}
1200	2.62^{cde}	70.93^{abc}	4.48^{d}	50.26^{b}	3.55^{de}	60.59^{bc}
1500	2.50^{cde}	72.22^{abc}	3.43^{e}	61.93^{a}	2.96^{ef}	67.07^{ab}
1800	2.23^{de}	75.26^{ab}	3.10^{e}	65.56^{a}	2.66^{f}	70.41^{a}
2100	1.82^{e}	79.81^{a}	2.93^{e}	67.48^{a}	2.37^{f}	73.65^{a}
LSD 5%	1.40	15.55	0.82	9.12	0.785	8.70
Mean	3.95i$^{b.}$	56.13 $^{a6.}$	5.73 ^{a}	36.30		

G. mean: mean of growth, GI%: percentage of growth inhibition.

Effect of silica NPs against *F. oxysporum* and *R. solani*.

Effect of bio-silica NPs against *F. oxysporum* and *R. solani*

The results in Table 3 showed that that both *F. oxysporum* and *R. solani* growth decreased with the increase of bio-silica NPsconcentrations. *F. oxysporum* was more susceptible to bio-silica NPsthan *R. solani* where *F. oxysporum* scored lower mean growth 3.50 cm than *R. solani* 5.36 cm at all concentrations with growth inhibition of 61.15% and 40.45% for the both fungus, respectively.

The obtained data revealed that that *F. oxysporum* growth was 9.00 cm at the control and decreased to 4.55 cm with growth inhibition of 49.55% with the increase of bio-silicaNPsconcentration to 10 ppm and reached the lowest value 1.65 cm at 200 ppm of bio-silica NPswith growth inhibition of 81.67%. With respect to *R. solani* the data in Table 3 showed that *R. solani* growth was 9.00 cm at the control treatment and reduced to 8.08 cm with growth inhibition of 10.26% with the increase of bio-silica NPsconcentration to 10 ppm. The growth decrease again until reached 2.28 cm with growth inhibition of 74.70% with the increase of bio-silica NPsconcentration to 200 ppm of bio-silica NPs. The mean growth rate of the both tested fungi species was 9.00 cm in the control treatment and gradually decreased with the increase of bio-silica NPsconcentrations. Across both fungi, the average growth of both *F. oxysporum* and *R. solani* was 6.31 cm at 10 ppm of bio-silica NPswith growth inhibition of 29.85% and the average growth of the both fungi reduced to 3.82 cm with growth inhibition of 57.56% at 100 ppm of bio-silica NPs. The average growth of the both fungi reached the lowest value at 200 ppm of bio-silica NPs1.96 cm this reduction in growth rate associated with the highest growth inhibition 78.19%

Table 3: Effect of difference concentrations of bio-silica NPsin *F. oxysporum* and *R. solani*.

Conc. (ppm)	*F. oxysporum*		*R. solani*		Mean	
	G. mean (cm)	GI%	G. mean (cm)	GI%	G. mean (cm)	GI% mean
Con	9.00^{a}	0.00^{h}	9.00^{a}	0.00^{g}	9.00^{a}	0.00^{i}
10	4.55^{b}.	49.44^{g}	8.08^{b}	10.26^{f}	6.31^{b}	29.85^{h}
20	3.82^{c}	57.59^{f}	7.67^{b}	14.81^{f}	5.74^{c}	36.20^{g}
40	3.58^{cd}	60.19^{ef}	7.59^{b}	15.63^{f}	5.59^{c}	37.91^{g}
60	3.21^{de}	64.33^{de}	6.20^{c}	31.11^{e}	4.71^{d}	47.72^{f}
80	3.00^{ef}	66.67^{cd}	6.05^{c}	32.78^{e}	4.53^{d}	49.72^{f}
100	2.82^{efg}	68.63^{bcd}	4.82^{d}	46.48^{d}	3.82^{e}	57.56^{e}
120	2.77^{efg}	69.26^{bcd}	4.06^{de}	54.89^{cd}	3.41^{ef}	62.07^{de}
140	2.68^{efg}	70.26^{bcd}	3.38^{ef}	62.48^{bc}	3.03^{fg}	66.37^{cd}
160	2.51^{fg}	72.11^{bc}	2.65^{fg}	70.56^{ab}	2.58^{gh}	71.33^{bc}
180	2.37^{g}	73.70^{b}	2.55^{g}	71.67^{a}	2.46^{h}	72.69^{b}
200	1.65^{h}	81.67^{a}	2.28^{g}	74.70^{a}	1.96^{i}	78.19^{a}
LSD 5%	0.57	6.37	0.77	8.60	0.47	5.19
Mean	3.50$^{b.}$	61.15^{a}	5.36^{a}	40.45^{b}		

G. mean: mean of growth, GI%: percentage of growth inhibition.

Effect of chem-silica NPs against *F. oxysporum* and *R. solani*.

The presented data in Table 4 showed that that both *F. oxysporum and R. solani* growth decreased with the increase of chem-silica NPs concentrations. *R. solani* more sensitive to chem-silica NPs than *F. oxysporum* where *R.solani* scored lower mean growth 3.42 cm than *F. oxysporum* 4.61 cm over all concentrations with growth inhibitions of 62.02% and 48.83% for the both fungus, respectively.

The growth of *F. oxysporum*were 9.00 cm at the control and decreased to 4.98 cm with a growth inhibition of 44.63% at 10 ppm reached the lowest value 3.23 cm at 200 ppmwith a growth inhibition of 64.07%. *R. solani* growth was 9.00 cm at the control and reduced to 4.98 cm at 10 ppm with a growth inhibition of 44.63% at 10 ppm. The growth decrease again to 1.95 cm with a growth inhibition of 78.33% with the increase of chem-silica NPs concentration to 200 ppm. The mean growth rate of the both tested fungi species was 9.00 cm in the control and gradually decreased with the increase of chem-silica NPs concentration. Across the both fungi, the average growth of both *F. oxysporum* and *R. solani* was 4.98 cm at 10 ppm with a growth inhibition of 44.63% and the average growth of the both fungus reached to the lowest value at 200 ppm of chem-silica NPs 2.59 cm with growth inhibition 71.20%.

Table 4: Effect of chem-silica NPs against *F. oxysporum* and *R. solani*.

	F. oxysporum		*R. solani*		Mean	
Conc. (ppm)	G. mean (cm)	GI%	G. mean (cm)	GI%	G. mean (cm)	GI% mean
Con	9.00^{a}	0.00^{f}	9.00^{a}	0.00^{f}	9.00^{a}	0.00^{h}
10	4.98^{b}	44.63^{e}	4.98^{b}	44.63^{e}	4.98^{b}	44.63^{g}
20	4.60^{bc}	48.89^{de}	4.17^{bc}	53.70^{de}	4.38^{c}	51.30^{f}
40	4.48^{bcd}	50.19^{cde}	3.73^{cd}	58.52^{cd}	4.11^{cd}	54.35^{ef}
60	4.42^{bcd}	50.93^{cde}	3.13^{de}	65.19^{bc}	3.78^{de}	58.06^{de}
80	4.38^{bcd}	51.30^{cde}	2.80^{def}	68.89^{abc}	3.59^{de}	60.09^{de}
100	4.35^{bcd}	51.67^{cde}	2.50^{ef}	72.22^{ab}	3.43^{ef}	61.94^{cd}
120	4.32^{bcd}	52.04^{cde}	2.28^{ef}	74.63^{ab}	3.30^{efg}	63.33^{bcd}
140	4.17^{cde}	53.70^{bcd}	2.25^{ef}	75.00^{ab}	3.21^{efg}	64.35^{bcd}
160	3.77^{def}	58.15^{abc}	2.15^{ef}	76.11^{ab}	2.96^{fgh}	67.13^{abc}
180	3.57^{ef}	60.37^{ab}	2.07^{f}	77.04^{a}	2.82^{gh}	68.70^{ab}
200	3.23^{f}	64.07^{a}	1.95^{f}	78.33^{a}	2.59^{h}	71.20^{a}
LSD 5%	0.72	8.01	0.99	1.99	0.59	6.61
Mean	4.61a	48.83b	3.42b	62.02a		

G. mean: mean of growth, GI%: percentage of growth inhibition

Nanotechnology considered one of the latest sciences and the most used in all fields of applied sciences, especially agricultural sciences. Many of the previous studies were in agreement with our findings, such as those of: Park [5] found that, silica-silver NPs was effective against *Botrytis cinerea, Colletotrichum gloeosporioides* and *R. solani*. In the same way, Suriyaprabha [17] indicate that Silica nanoparticles used as an alternative potent antifungal agent against phytopathogens. In addition, Nejad [18] indicated that the activity of the SNPs aganist *R. solani* are dependent on sprayed, mycelia growth and the quantity of SNPs. Kanmani [19] showed that one possible cause is the reaction of AgNPs with phosphorous and sulfur containing materials inside and outs ide of cells. In addition, positive charge-containing AgNPs believed to bind with negative charge-containing fungal membranes and disrupt cell walls and then destroying the membrane lipid bilayer, leading to induce the intracellular ion efflux resulting in cell death. Raghunath [20] revealed that copper oxide nano-sheets tend to there is non-specifically wrap the microbial cells. It suggested that, CuONPs might be interact with molecules containing P and S either outside or inside the fungal cells. On the other hand, positively charged CuONPs may bind to negatively charged fungal membranes, and this electrostatic attraction may cause disruption in the fungal cell wall functions with damage in the protein synthesis profile and the intracellular ion exchanges, and finally resulting in fungal cell death. In addition, El Shewy [21] confirmed that nanoparticles exhibited inhibitory effect of *R. solani*.

Effect of different antifungal treatments in *F. oxysporum* and *R. solani* growth.

F.oxysporum:

In case of *F.oxysporum*, all treatments exhibited significant declines in hyphal morphology, respect to control (Fig 2). Image of control group at 400X (Fig. 2A) showed regular hyphal and extensively growth. At magnified image at 1000x, it was obtained regular hyphal (Fig 2A*). However, treatment with fungicide, Ridomil at concentration (1000 ppm) exhibited slightly irregular shape and some adsorbed hyphal (Fig. 2B). In magnified field at 1000x, it was obtained adsorbed and irregular shape (Fig 2B*). Significant deformation was noted in case of *C. nodosa* treatment (2100 ppm) (Fig. 2C), while magnified field (1000x) displayed adsorbed and deformed hyphal (Fig. 2C*). Regarding bio-silica NPs exhibited significant lysis, deformed and adsorbed hyphal morphology (Fig. 2D) (at 400x), followed by magnified field (1000x) displaying these alterations with a bright focusing (Fig. 2D*). Significant destructed and

adsorbed hyphal were obtained in case of chem-silica NPs (Fig. 2E) at 400x, and as described above the bright focusing at 1000x documented these alterations (Fig. 2E*).

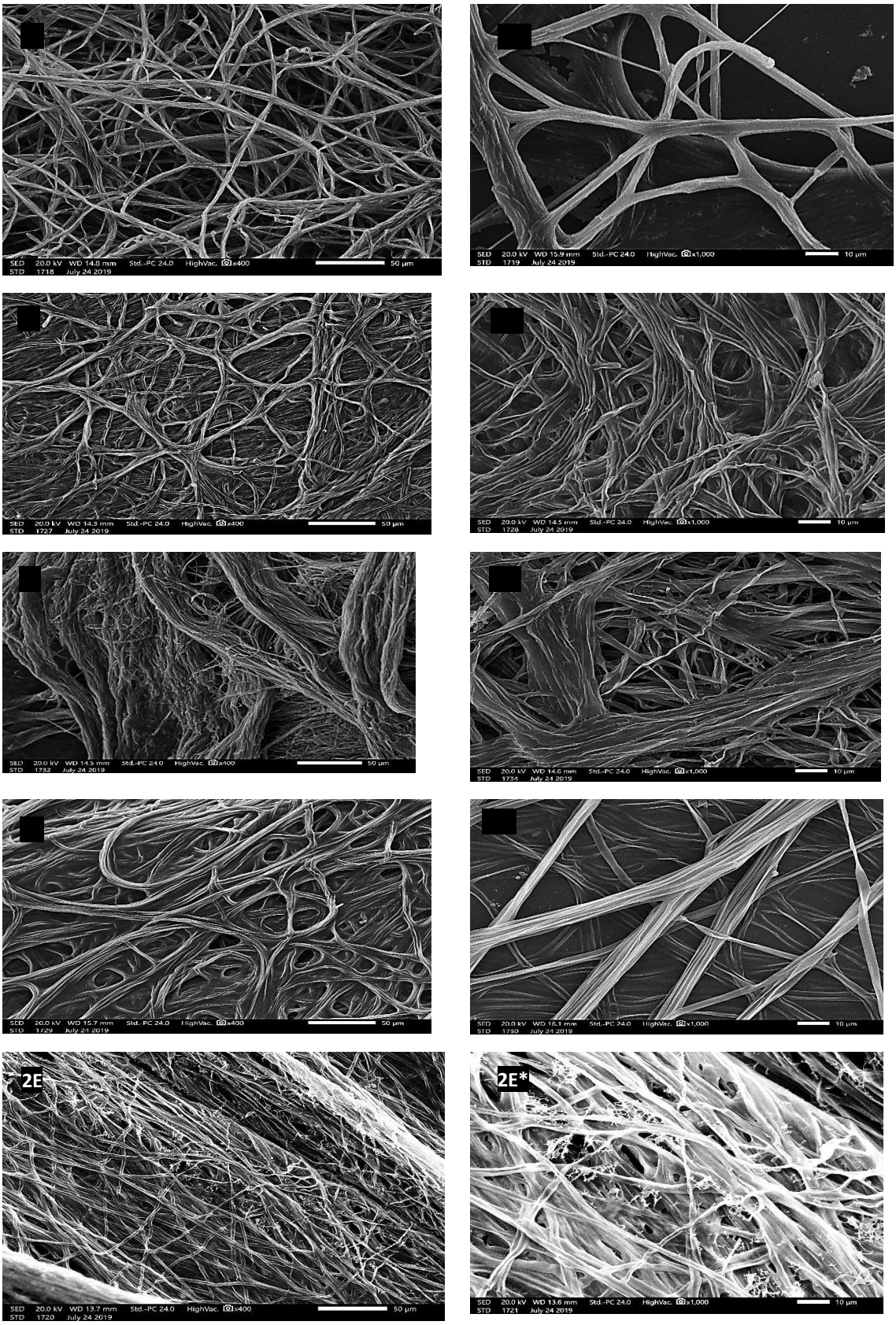
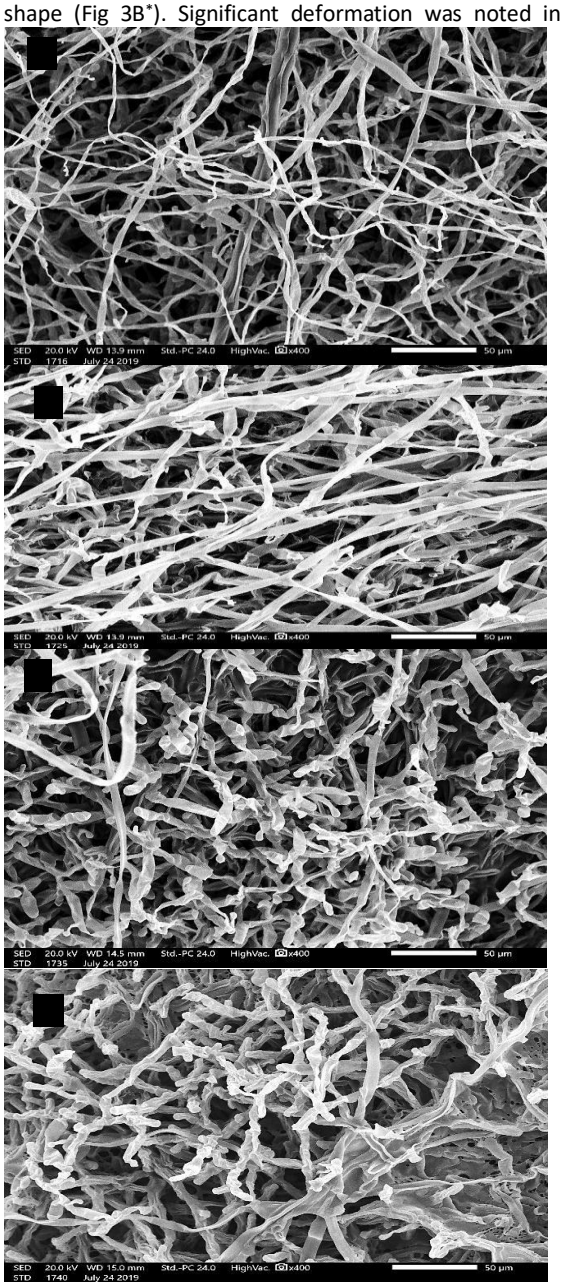
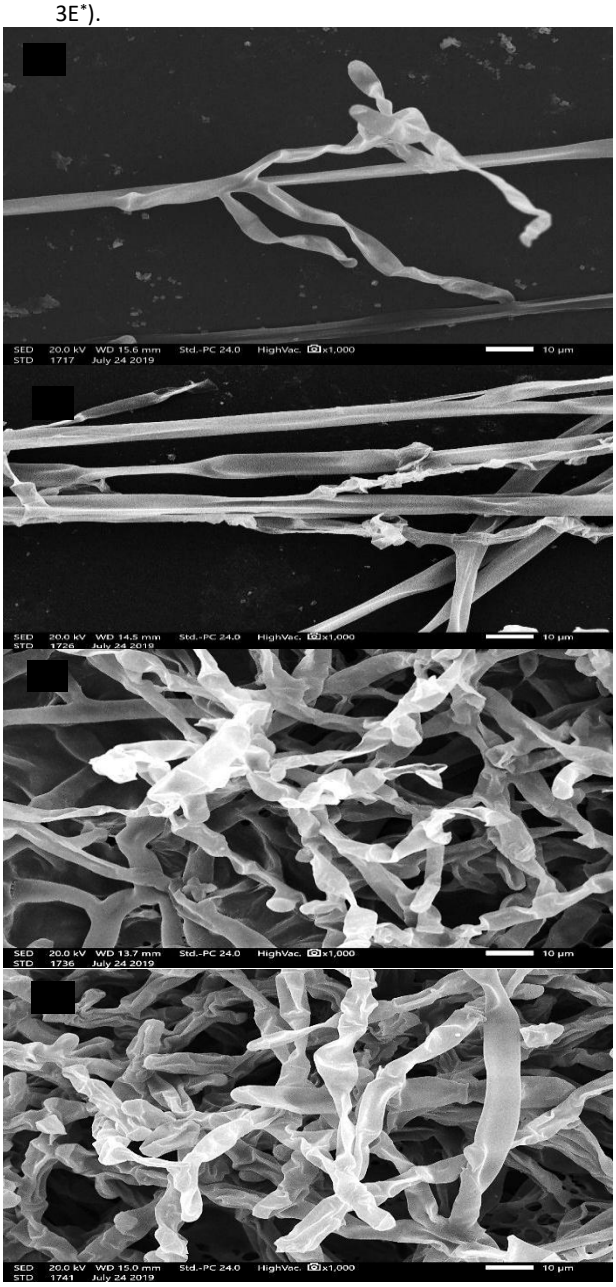


Fig. 2: Effect of different antifungal treatments in *F.oxysporum* growth at lab conditions by using SEM visualization technique [A =Control, B= Ridomil, C= *C.nodosa*, D= Bio-silica NPs and E= Chem. silica NPs].

***R. solani*:**

In case of *R. solani*, all treatments exhibited significant declines in hyphal morphology, respect to control (Fig 3). Image of control group at 400_X (Fig. 3A) showed regular hyphal and extensively growth. At magnified image at 1000x, it was obtained regular hyphal (Fig 3A*). However, treatment with fungicide, Ridomil at concentration (1000 ppm) exhibited slightly irregular shape and some adsorbed hyphal (Fig. 3B). In magnified field at 1000x, it was obtained adsorbed and irregular shape (Fig 3B*). Significant deformation was noted in case of *C. nodosa* treatment (2100 ppm) (Fig. 3C), while magnified field (1000x) displayed adsorbed and deformed hyphal (Fig. 3C*). Regarding bio-silica NPsexhibited significant lysis, deformed and adsorbed hyphal morphology (Fig. 3D) (at 400x), followed by magnified field (1000x) displaying these alterations with a bright focusing (Fig. 3D*). Significant destructed and adsorbed hyphal were obtained in case of chem-silica NPs (Fig. 3E) at 400x, and as described above the bright focusing at 1000x documented these alterations (Fig. 3E*).





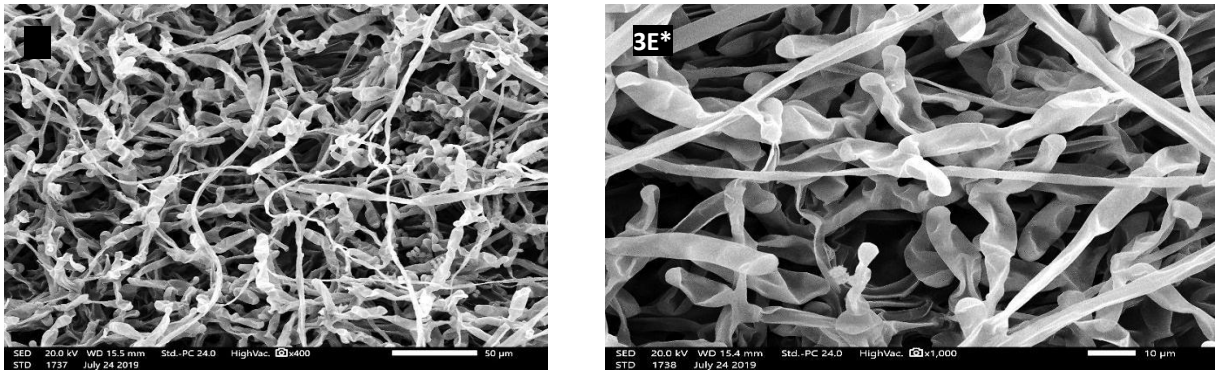


Fig. 3: Effect of different antifungal treatments in *R.solani* growth at lab conditions by using SEM visualization technique [A =Control, B= Ridomil, C= *C.nodosa*, D= Bio-silica NPs and E= Chem. silica NPs].

In magnified field at 6000 x for glides of *F. oxysporum* it was obtained the interaction of bio-silica NPswith hyphal (fig. 4A) Aggregated hyphal with conduced tissues and irregular surface containing and / or vacuolated Nps .In case of chem-silica NPs (fig 4B), image illustrates significantly detruded hyphal unsoiled with tissues resulted in disruption and losses of hyphal compound.

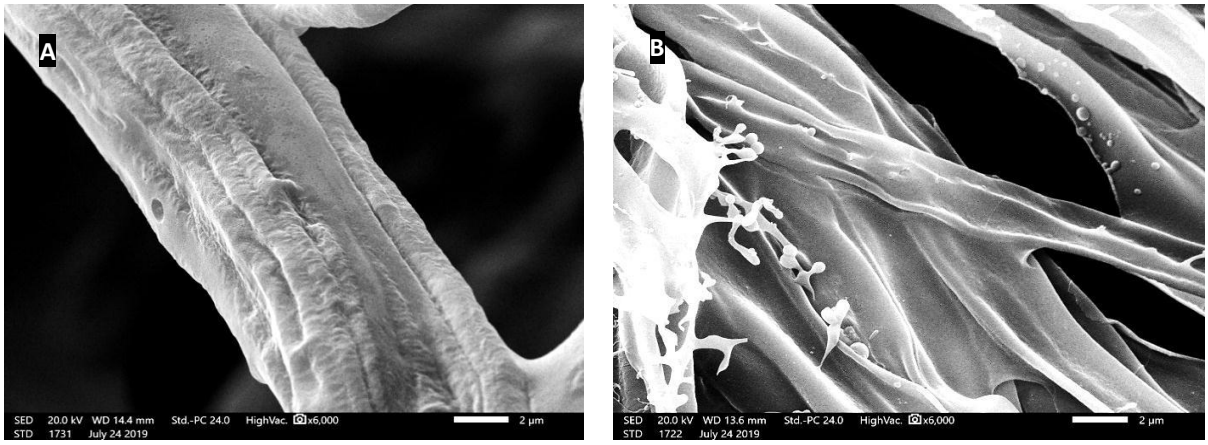


Fig. 4: SEM images illustrate the interaction of (A) bio and (B) chem. silica NPs. with *F. oxysporum* at magnified bright fields (6000 x).

In the same bright fields (6000 x) of *R. solani*, it was obtained the adsorbed hyphal with coated bio-silica NPs (fig. 5A), while some of chem-silica NPs illustrates adsorbed with irregular surface of hyphal associated with significant interaction with Nps (fig. 5B). Yehia [22] investigated the antifungal efficiency of Zinc oxide nanoparticles (Zno NPs) against both pathogenic fungal species, *F. oxysporum* and *P. expansum.* The scanning electron microscopy (SEM) revealed obvious deformation in the growing mycelia treated with Zno NPs in *F. oxysporum* might be the cause of growth inhibition key. Baka [23] indicated that, SEM and TEM used in some previous studies to show that the plant extracts were able to delay the fungal germination; the observations by SEM and TEM showed severe changes in the morphology and ultrastructure of *A.solani* when treated with the ethanol extract of the *Calotropisproceraleaves* at a concentration of 20%. Balashanmugam [24] reported that, the AgNPs exhibit higher antifungal activity when compared with the conventional antifungal drug amphotericin B against *Rsolani*, *F.oxysporum* and *Curvularia* sp. Scanning electron microscope (SEM) analysis showed distinct structural changes in the cell membranes of tested fungi AgNPs treatment. These results suggest that phytosynthesized AgNPs used as effective growth inhibitors in controlling plant diseases caused by fungi. Marinho[25]showed that the SEM of leaves extract from *Sapindus saponaria* by hydroethanolic L. due to morphological changes on *Colletotrichum gloeosporioides* and reduced the hyphae size at 50 mg/ml, while caused damage in hyphae at 100 mg/ml done mycelial rupture . Al Ali [26] inducated that the SEM the *Ziziphus sp* extract 150 ml/ml caused shrinkage on the cell wall , cellular cavities and reduced the cells number to abnormal cell on *Candida albicans.* El Shewy [21] evaluated the effect of nanoparticles forms of Tri–calcium phosphate, copper oxide and silicon dioxide nanoparticle at five concentrations to control black scurf disease in vitro and in vivo. Examination of treated R. solani with different tested nanoparticle done using Transmission Electron Microscope (TEM). Physical characteristics of tested nanoparticles exhibit that all of them are spherical in shape and varied in their sizes. In addition, all tested nanoparticles exhibited inhibitory effect of R. solani. Non-copper oxide was the most effective one (56.42 %) in suppressing the mycelial growth of R. solani at concentration 250 µl/L.

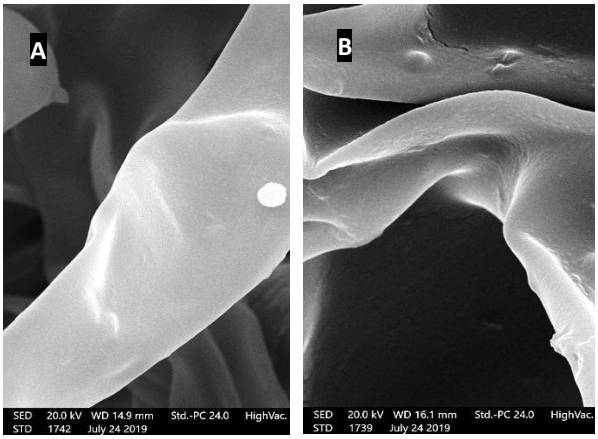


Fig. 5: SEM images illustrate the interaction of (A) bio silica NPs and (B) chem-silica NPs with *R. solani* at magnified bright fields (6000 x).

Conclusions

All tested compounds, Ridomil, *C. nodosa* extract, bio-silica NPs and chem-silica NPs were active against *F. oxysporum* and *R. solani*. *F. oxysporum* was more sensitive to all tested compounds except chem-silica NPs.

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References

[1]Gamliel-Atinsky E., Inoculum availability and conidial dispersal patterns of Fusarium mangiferae, the causal agent of mango malformation disease. Phytopathology. 99(2):160-166 (2009).

[2]Wibberg D., Rupp O., Jelonek L., Kröber M., Verwaaijen B., Blom J., Winkler A., Goesmann A., Grosch R., Pühler A. and Schlüter A., Improved genome sequence of the phytopathogenic fungus Rhizoctonia solani AG1-IB 7/3/14 as established by deep mate-pair sequencing on the MiSeq (Illumina) system". J. Biotechnol. 203: 19–21 (2015).

[3]Ullah S., Naz F., Fiaz M., Sher H., Hussain S., Ahmad S. and Khalid A.N., In vitro evaluation of commonly available fungicides against three fungal isolates. Plant Pathology & Quarantine 8(1): 67–77 (2018).

[4]Yadav A., Bhardwaj R. and Sharma R.A., Phytochemical screening and antimicrobial activity of anthraquinones isolated from different parts of Cassia nodosa. Res. J. Medicinal Plant, 7:150.157 (2013).

[5]Park H.J., Kim S.H., Kim H.J. and Choi S.H., A new composition of nanosized silica-silver for con-trol of various plant diseases. Plant Pathol J 22:295–302(2006).

[6]Taisan W. A., Bahkali A.H., Elgorban A.M. and El-Metwally M.A., Effective influence of essential oils and microelements against *Sclerotinia Sclerotiorum*. International Journal of pharmacology;10 (5): 275-281(2014).

[7]Winer B.J., Statistical Principles in Experimental Design. 2nded. New York: McGraw Hill, USA (1971).

[8]Fravel D.R., Deahl K.L. and Stommel J.R., Compatibility of the biocontrol fungus *Fusarium oxysporum* strain CS-20 with selected fungicides. Biological Control 34:165–169(2005).

[9]Njiru M. D., Integrated management of fusarium wilt of tomatoes. A Thesis Submitted In Partial Fulfillment of The Requirements for The Award of Me Degree of Master of Science Plant pathology (2013).

[10]Saad A.S.A., Kadous E.A., Tayeb E.H., Massoud M.A., Ahmed Soad M. and Abou El-Ela A.S.A., The inhibitory effect of some antioxidants and fungicides on the growth of *Alternaria solani* and *Fusarium solani* in vitro. Middle East J. Agri. Res., 3(2): 123-134 (2014).

[11]Gade A.A., Choudhari S.S. and Kapratwar K.A., Efficacy of Ridomil against Fusarium solani causing rhizome rot diseases of *Zingiber officinal* Rosc. International Journal of Advanced Scientific Research, 1(6): 25-27(2016).

[12]Banu N. and Sharada M.S., Inhibitory effects of mancozeb on growth and stimulation of resistance against fusarium wilt of brinjal. The Bioscan, 13(1): 285-290 (2018).

[13]Bowers J.H. and Locke J.C., Effect of botanical extracts on the population density of *Fusarium oxysporum* in soil and control of fusarium wilt in the greenhouse. Plant Disease, 84 (3):300-305 (2000).

[14]Mogle U. P. (2013). Effect of medicinal plant extracts against the post-harvest. Trends on life sci. 4(4), 22–27.

[15]Ibrahim O.H.M., Abdul-Hafeez E.Y. and Mahmoud A.F., Antimicrobial and Antioxidant Activities of Stem Bark Extracts of Different Ornamental Trees. Assiut J. Agric. Sci., 46 (5):19- 32 (2015).

[16]Abid M. and Hussain F., Effect of Annona squamosa ,Ocimum basilicum and Cassia fistula used as organic amendments against the soil – Borne Pathogens in . 15, 903-908 (2018).

[17]Suriyaprabha R., Karunakaran G., Kavitha K., Yuvakkumar R., Rajendran V. and Kannan V., Application of silica nanoparticles in maize to enhance fungal resistance. IET Nanobiotechnology, 8(3), 133–137(2014).

[18]Nejad M.S., Hosein G., Bonjar S., Khatami M. and Amini A., In vitro and in vivo antifungal properties of

silver nanoparticles against *Rhizoctonia solani*, a common agent of rice sheath blight disease. IET Nanobiotechnology, 11(3): 236–240(2016).

[19]Kanmani P. and Lim S.T., Synthesis and structural characterization of silver nanoparticles using bacterial exopolysaccharide and its antimicrobial activity against food and multidrug resistant pathogens. Process Biochem 48(7):1099–1106 (2013).

[20]Raghunath A. and Perumal E., Metal oxide nanoparticles as antimicrobial agents: A promise for the future. Int. J. Antimicrob. Agents, 49, 137–152 (2017)

[21]El-Shewy, Eman.S.A., Mohemed F.G., Abd-latif Faten M., Hafez E.M. and Mansour S.A, The efficacy of Copper Oxide, Tri-calcium Phosphate and Silicon Dioxide Nanoparticles in Controlling Black Scurf Disease of Potato. Annals of Agric. Sci., Moshtohor, Vol. 57(1):129 – 138 (2019).

[22] Yehia, R.S. and Ahmed O.F, In vitro study of the antifungal eficacy of zinc oxide nanoparticles against *Fusarium oxysporum* and *Penicillium expansum*. Afr J Microbiol Res 7(19):1917–1923 (2013).

[23]Baka Z. and Rashad Y., Alternative control of early blight of tomato using plant extracts from Acacia nilotica, Achillea fragrantissima and Calotropis procera. Phytopathologia Mediterranea. 55, 121–129 (2016).

[24]Balashanmugam P., Balakumaran M.D., Murugan R., Dhanapal K., and Kalaichelvan P.T., Phytogenic synthesis of silver nanoparticles , optimization and evaluation of in vitro antifungal activity against human and plant pathogens. Microbiological Res., 192: 52–64(2016).

[25]Marinho G.J.P., Klein D.E. and Luis Junior C.S., Evaluation of soapberry (Sapindus saponaria L.) leaf extract against papaya anthracnose. Summa Phytopathologica 44, 127–131(2018).

[26]Al-Ali S. and Al-Judaibi A., Biochemical and molecular effects of Phoenix dactylifera and Ziziphus spina-christi extracts on Candida albicans. J. Biosci. Med. 7, 29–43 (2019).

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