

Evaluation the efficiency ginger plant extracts and their compatible with some bacterial species for controlling eggplant root rot disease caused by rhizoctonia solani

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Abstract

This study was carried out to evaluated the efficiency of Ginger plant extracts and their compatibility with four bacterial species as agents for controlling eggplant plants infected by root rot disease caused by *R. solani* under laboratory and greenhouse conditions. Results of the effect of Ginger cold water extract on radial growth of *R. solani* under laboratory conditions showed that treatments revealed positive effect in reducing fungal growth of pathogen and the effect depend on extract concentration, the effect was increased when extract concentration was elevated, the inhibition percentage of the ginger cold extract ranged between 26.06% for the concentration of 100 mg/ml and 61.46% for the concentration treatment of 400 mg/ml. The results showed that the concentration of 400 mg/ml completely inhibited the growth of the pathogenic fungus to 0 mm with an inhibition percentage of 100%. The results revealed that the alcoholic extract of ginger showed that the growth of the pathogenic fungus was completely inhibited at a concentration of 20 mg per ml. The findings showed that *B. subtilis* treatment revealed significant excellence in growth inhibition as compared with other bacterial strains treatment which recorded 69.76 % as compared to 31.29 and 56.61% for *Streptococcus* sp, and *P. fluorescence* treatments. Data showed that the bacterial suspension of *B subtilis* significant superior in increment number of germinated seeds and germination percentage which recorded 86.70 % and 8.67 seed / plant as compared to 7.33 seed / plant and 73.30 % of control treatment. The results revealed that the combination of *B. subtilis* bacteria and the alcoholic extract of ginger plant gave the highest significant values in increasing plant growth and disease index parameters.

Keywords: Ginger plant extract, *Rhizoctonai solani*, Bacterial strains, Biocontrol agents

Introduction

Eggplant plant is considers as one of the important agricultural crops so it rich by multi vitamins such as vitamin C , B1 and B6 , and minerals like Fe , Ca , K , MG and antioxidants , and because it contains a low of calories and lipids which lead to regard this crop has useful healthy advantages for human and their healthy (Naeem and Ugur , 2019) .This crop is exposed to many pathogenic fungi including root rot disease that causing by *Rhizoconia solani* which lead to weak plant growth and eventually lead to decreased its productivities (Kaniyassery et al. , 2023 , Kumar et al . , 2024).The looks of researchers and competent in plant disease were tended to find a modalities and methods that considers safety and unpolluted to environment and to introduced healthy food from poisonous chemical pesticides and may be cancerous to human and other living organisms (Pathak et al. , 2022) , and one from these methods were utilizing plant extracts in controlling pathogenic fungi including *Rhizoconia solani* causing root rot disease (Behiry et al.,. 2022, Hefish et al., 2023,

Hlokwe et al., 2020) . Many of scientific references were pointed to the role of Zinger plant extract (*Zingiber officinale*) and extract in growth inhibition of some plant pathogenic fungi (Kalhor et al., 2022) , in previous study Sunder and Hasan (2014) were studied that inhibitory effect of some plant extracts against *F. oxysporum* and they found that radial growth of pathogen significant inhibited most the tested plant extracts except sunflower plant extract , the inhibition percentage were increased with the increasing in extracts concentration , and concluded that Ginger plant extract may be utilized as effective factor against this pathogen , whereas (Cherkapally et. al. , 2017) were tested the antifungal activity of nine plant extracts against *F. oxysporum* , *R. solani* and *Macrophomina phaseolina* in laboratory experiments, they found that Garlic extract glove was completely inhibited the growth of *M. phaseolina* in all tested concentrations. Some of scientific references pointed to role of many of bacterial species in controlling plant pathogenic fungi of eggplant plant, in previous study (Karimi et. al. , 2012, Fatima et al., 2025) which evaluated six of bacterial isolates of *Pseudomonas*

aerogenosa and *P. patida* and six isolates of *Bacillus subtilis* as biocontrol agents against *F. oxysporum* under laboratory and greenhouse conditions and they found that some of these isolates has high ability in protection plant seeds or soil that previous treated with bacteria as compared with control treatment which significant increased most tested growth parameters (plant height , dry weight and soft weight of plant) . Mehdi and jomaah (2020) studied and evaluated the efficiency of *B. subtilis* in controlling eggplant root rot disease and concluded that the use of the bacteria led to a reduction in disease severity and improved plant growth parameters. Waheed et. al. (2014) was evaluated of six strains of *Azotobacter Bacillus*, and *Pseudomonas* bacteria in decreasing eggplant plant infected with *R. solani* under laboratory and greenhouse conditions and they found that *B. subtilis* were significantly excellence in reducing plant infection with this fungus as compared to other bacterial strains. So, this study was conducted to evaluated of water and alcoholic extracts of Ginger and their compatibilities with some bacterial species in controlling root rot disease caused by *R. solani* and plant wilt caused by *F. oxysporum* under laboratory and greenhouse conditions.

Materials and Methods

Culturing and reactivating of bacterial species on culture media

Four bacterial species (*Pseudomonas florescence*, *Bacillus subtilis*, *Azospirillum sp.* and *Streptomyces sp.*) were cultured and activated on nutrient agar and saved under 4 °C until used.

Culturing and activating of eggplant pathogenic fungi

In this study we were utilized one severe isolate of *R. solani* culturing on potato dextrose agar medium which the isolates were reactivated and purified and then saved at 4 °C until used .

Preparing of cold and hot water extract of Ginger plants:

Firstly we were took 50 gm of dry powder (Ginger rhizomes) and placed in electric blender with 1 liter of distilled water , then we were mix for 10 minutes

and we filtered the mixture through satirized filter paper by aiding vacuum instrument , then we were saved the water extract of each plant until used , and with the same method we were utilized hot water instead of cold water to prepared hot extract of Ginger so we were soaked dry powder (Ginger rhizomes) for 1 hour and we were supervene the same steps to prepare hot water extract of the two Ginger plants .

Preparing of alcoholic extract of Ginger plant

We were obtained Ginger plant rhizomes from the domestic market, we firstly aerobic dried the rhizomes and crushed by electric grinder and then we weighted 25 gm of powder and added to 100 ml of ethanol alcohol at room temperature overnight, then we were filtered the mixture by using Whatman No. 1 and then we were dried with electric oven at 40 °C for 24 hours (Grace et. al., 2017; Jam et al., 2025).

Study the effect of antagonistic activity of bacterial strains against *R. solani* under laboratory conditions:

We were used four bacterial strains (*Streptococcus sp.* , *P. fluorescence* , *Azospirillum sp.* and *B. subtilis*) to study the effect of bacterial suspension of these bacteria on radial growth and percentage inhibition of the tested pathogens (*R. solani*) , we cultured the four strains on Nutrient broth medium at 28 °C for 10 days and then we were abstracted the mixture by using vacuum instrument with Millipore bacterial filter paper , then we added 0.1 ml of supernatant to petri dish containing PDA and inoculated each petri with one loop (5 mm diameter) of one of the two tested pathogens , then we were daily monitored the pathogen growth and recorded the radial growth rate of pathogen until the growth pathogen reached to the edge of petri dish in control treatment .

Study the effect of cold, hot and alcoholic Ginger extract on growth of *R. solani*

Five concentrations of cold ant hot ginger extract (0.0 , 100 , 200 , 300 ,400 and 500) mg/ ml were chosen to study the effect of these concentration on radial growth of *R. solani* under laboratory conditions , firstly we were added 1 ml of the tested plant extract concentration to a petri dish containing 20 ml of PDA , then we were inoculated the center of these petri

dishes with one disc (5mm diameter) of the pathogen (*R. solani*) and then incubated the petri dishes at 25 °C and we were daily monitored the radial growth of the pathogen to recorded its radial growth until the growth of the control treatment were reached to the edge of petri dish .

Effect of bacterial suspension extracts on germination and vital of eggplant seeds under laboratory conditions

Conical flask (1 liter volume) containing nutrient agar was chosen to prepared to grown the four bacterial strains (*Streptococcus sp.* , *P. fluorescence* , *Azospirillum sp.* , *Bacillus subtilis*) , inoculated with one disc of tested bacterium (10 mm diameter) then incubated at 25 °C for 10 days , filtered the bacterial suspension through Millipore filter (0.45 µ) then we were added 10 ml of the bacterial suspension of one the four bacterial strains to a petri dish containing sterilize filter paper and then added 10 seeds of eggplant plant , incubate the petri dishes at 25 °C and watered the dishes daily to record the germinate seeds and percentage of seeds germination .

Study the compatibility of *B. subtilis* and ginger water extract and their effect on some growth and disease parameters of *R. solani* under greenhouse conditions

One isolate of plant growth promoting bacteria selected from laboratory tests results that showed the best result in suppression of eggplant pathogen (*R. solani*) and we used the water extract of ginger plant for soaking eggplant seeds for 15 minutes then dried seeds with filter paper no. 1, plastic pots containing sterilizing soil (5 Kg) then we were cultured the seeds according to the following treatments:

1. Positive control treatment (without pathogen).
2. Negative control treatment (with pathogen).
3. Ginger extract treatment.
4. *B. subtilis* treatment.
5. Ginger extract+ *B. subtilis* treatment.
6. Ginger extract + pathogen.
7. *B. subtilis* + pathogen.
8. *B. subtilis* + ginger extract + pathogen.

Results and discussions

Results of the effect of cold extract of ginger plant on radial growth of *R. solani* under laboratory conditions showed that treatments significantly reduced radial growth of pathogen , The results showed that all concentrations of ginger plant for the cold extract did not lead to complete inhibition of the growth of the pathogenic fungus, as the results showed that the highest inhibition percentage reached 61.46% at the concentration of 400 mg/ml, and achieved a diameter growth rate of 36.33 mm compared to 94.33 mm for the control treatment (Table 1). However, the inhibition percentage of the ginger cold extract ranged between 26.06% for the concentration of 100 mg/ml and 61.46% for the concentration treatment of 400 mg/ml. The reason for this condition may be that the concentration of the active ingredient in the cold ginger extract was low or not concentrated, which led to a decrease in the percentage of inhibition of the growth of the pathogenic fungus.

Table 1: Effect of cold extract of ginger plant on radial growth of *R. solani* under laboratory conditions

Concentration mg / ml	Radial growth rate (mm)	Inhibition (%)
0.0 mg / ml	94.33	0.0
100 mg / ml	69.67	26.06
200 mg / ml	62.33	33.78
300 mg / ml	43.67	53.75
400 mg / ml	36.33	61.46
LSD (P≤0.05)	4.46	4.78

The results of table 2 pointed that the treatment (400 mg / ml) significant excellence on other treatments in reducing growth rate of pathogen (*R. solani*) which decreased the growth to zero mm with inhibition percentage (100 %). The results of Table 2 showed that the effect of the hot ginger extract on the growth of the pathogenic fungus was a direct relationship, whereby the higher the concentration of the hot extract, the higher the percentage of inhibition significantly compared to the control treatment. The results indicated that the concentration of 400 mg/ml completely inhibited the growth of the pathogenic fungus to 0 mm with an inhibition percentage of 100%. The reason for this is that heating the ginger extract led to an increase in the concentration of the active ingredient which led to the complete inhibition of fungal growth.

Table 2: Effect of hot extract of ginger plant on radial growth of *R. solani* under laboratory conditions

Concentration mg / ml	Radial growth rate of <i>R. solani</i> (mm)	Inhibition (%)
0.0 mg / ml	91.0	0.0
100 mg / ml	62.0	31.68
200 mg / ml	50.67	44.31
300 mg / ml	39.33	56.76
400 mg / ml	0.0	100.0
LSD ($P \leq 0.05$)	2.37	2.36

Results of the effect of ginger ethanol extract on radial growth rate of under laboratory conditions showed that treatments varied in their abilities in inhibition of growth rate of the pathogen (*R. solani*), the effect was depending on extract concentration and type of tested bacteria, (20 mg / ml) concentration inhibited the growth of *R. solani* completely (table 3). The results of table 3 revealed that the effect of the alcoholic extract of ginger increased directly with the increase in the concentration of the alcoholic extract. At a concentration of 5 mg per ml, the diameter growth rate of the pathogenic fungus was 52.0 mm with an inhibition rate of 44.67%. Then it increased to 80.0% at a concentration of 15 mg per ml, while the growth of the pathogenic fungus was completely inhibited at a concentration of 20 mg per ml. These results showed that extracting the active ingredient of ginger using alcohol led to a reduction in the concentration required to inhibit the growth of the fungus *R. solani* from 400 mg per ml in the hot aqueous extract to 20 mg per ml in the alcoholic extract of ginger. This gives great importance to the alcoholic extraction method of ginger in controlling the growth of the pathogenic fungus under laboratory conditions and gives encouraging results in controlling eggplant root rot disease (table 3).

Table 3: Effect of alcoholic extract of ginger plant on growth of *R. solani* under laboratory conditions

Treatment	<i>R. solani</i>	
	Radial growth rate (mm)	% Inhibition
0.0 mg / ml	93.67	-
5 mg / ml	52.0	44.67
10 mg / ml	35.67	62.0
15 mg / ml	19.0	80.0
20 mg / ml	0.0	100
LSD ≥ 0.05	4.45	4.67

The findings of the antagonistic activity of the tested

four bacterial strains showed that bacterial strains varied in their responsive in growth inhibition of the pathogen (*R. solani*), *B. subtilis* treatment revealed significant excellence in growth inhibition as compared with other bacterial strains treatment which recorded 69.76 % as compared to 31.29 and 56.61% for *Streptococcus sp.*, and *P. fluorescence* treatments respectively (table 4). These results obtained from Table 4 revealed that *B. subtilis* significantly outperformed the rest of the tested bacterial species through the high inhibition rate which reached 69.76%, which led to its selection in greenhouse experiments to study the effect of these bacteria in conjunction with the alcoholic ginger extract.

Table 4: Antagonistic activity of bacterial strains against *R. solani* under laboratory conditions

Treatment	<i>R. solani</i>	
	Radial growth rate (mm)	% Inhibition
Pathogen only	60.67	-
Pathogen+ Strep.	41.67	31.29
Pathogen+ P.f	26.33	56.61
Pathogen+Azosp.	36.0	40.64
Pathogen+B. s	18.33	69.76
LSD ≥ 0.05	2.49	3.75

Table 5 : Effect of bacterial suspension on germination and vital of eggplant under laboratory conditions

Treatment	Number of germinated seeds / plants	Inhibition %
Pathogen only	7.33	73.30
Pathogen+ Strep.	7.67	76.70
Pathogen+ P.f	8.0	80.0
Pathogen+A zosp.	7.33	73.30
Pathogen+B . s	8.67	86.70
LSD ≥ 0.05	0.81	8.06

Data of table 5 showed that four bacterial strains varied in their abilities in increment the germination and vital of eggplant plants, the bacterial suspension of *B. subtilis* significant superior in increment number of germinated seeds and germination percentage which recorded 86.70 % and 8.67 seed / plant as compared to 7.33 seed / plant and 73.30 % of control

treatment respectively. The results of Table 5 showed that *B. subtilis* gave the highest significant value for the average number of germinated seeds per plant (8.67%) seeds per plant, which encouraged us to use these bacteria in greenhouse experiments

Also, the results of table 6 showed that treatments significant improved of the tested growth parameters (shoot length, root length, number of leaves / plants, soft and dry weight of plant as compared to negative and positive control treatment. The results of Table 6 revealed that the combination of *B. subtilis* bacteria and the alcoholic extract of ginger plant gave the highest significant values in increasing plant growth parameters (shoot length of , root length , number of leaves per plant, fresh and dry weight per plant) as they recorded 69.0 cm, 36.67 cm, 33.33 leaves per plant, 354.87 g per plant, 132.63 g per plant, respectively, compared to the control treatment which recorded 46.33 cm, 34.33 cm, 22.33 leaves per plant, 212.43 gm per plant , 76.93 gm per plant respectively . The results also indicated that the compatibility between *B. subtilis* and ginger extract led to a significant reduction in the pathogenicity index of *R. solani* from 66.0 in the treatment of the

pathogen alone to 18.0 in the treatment of *B. subtilis* and the plant extract and pathogen, while the treatment of *B. subtilis* with the pathogen recorded 34.33 and the treatment of the plant extract with the pathogen recorded 40.67 (Table 6) These results are consistent with the results of the study by Mahdi and Jumaah (2020), which concluded that the plants treated with *B. subtilis* had a significantly lower incidence and severity of root rot disease in eggplant plants. The results also showed a significant increase in the fresh and dry weight of the shoot and root system, in addition to an increase in plant weight, number of leaves per plant, and leaf area compared to untreated plants. Al-Ajeely and Matloob (2025) found that *P. fluorescence* significantly inhibited the growth of the pathogenic fungus *R. solani* in laboratory experiments by 100%. They also revealed that the combination of the bacteria and chitosan, together or alone, significantly reduced the incidence and severity of eggplant root rot and significantly improved growth parameters. Matloob and Al-Baldawy (2020) showed that the compatibility between *T. harzianum* and *A. chroococcum* showed a high antagonistic capacity against the fungus *R. solani*, which causes eggplant root rot disease.

Table 6: Compatibility between *B. subtilis* and ginger extract and their effect on some growth and disease parameters artificially infected with *R. solani* under greenhouse conditions

Treatment	Shoot length cm	Root length cm	No. leaves / plant	Soft weight of plant gm/ plant	Dry weight of plant gm/ plant	Disease index
Control	46.33	34.33	22.33	212.43	76.93	0.01
Pathogen	18.33	13.67	4.67	87.37	26.83	66.0
Plant extract	55.67	34.0	25.33	325.53	124.60	-
<i>B. subtilis</i>	64.33	37.67	30.67	375.30	145.30	-
Plant ext.+B.s	77.67	40.67	37.67	425.40	172.20	-
Plant ext.+P	51.0	29.67	21.33	284.03	102.0	40.67
B.s +P	61.33	31.67	28.67	294.93	105.40	34.33
B.s +Pl. ext.+P	69.0	36.67	33.33	354.87	132.63	18.0
LSD ≥ 0.05	4.10	5.28	3.02	6.12	3.35	6.51

B. s =*B. subtilis*, Pl. ext.= Plant extract, P=Pathogen.

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