Biocontrol of *Rhizoctonia solani* damping-off disease in cotton seedlings with endophytic bacteria

By

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- Plant pathogenic microorganisms are one of the serious threats causing great losses to important crop plants. Many pesticides are used but they are considered dangerous to both human health and environment.
- Microorganisms especially endophytes have been successfully used in the biological control of plant diseases caused by bacteria and fungi.
- This study intended to isolate and identify endophytic bacteria with antimicrobial activity against important phytopathogens. Then to test the most potent bacteria against phytopathogen under greenhouse conditions. Finally, identifying the produced secondary metabolites of bacteria.
- The obtained results can be summarized as following:
    1. A total of 52 isolates of bacteria were isolated from stem, root, leaf of 10 crop plants that were collected from different fields in Fayoum governorate, Egypt.
    2. The 52 isolates were screened for their antifungal activity against *Fusarium solani* as a model organism. 20 isolates were found inhibitory for the fungus.
    3. Three bacterial isolates (H40, H8, and H18) showed strong inhibition against *F.solani* and were further assayed for antimicrobial activity against 10 selected phytopathogenic bacteria and fungi.
    4. All three endophytic bacterial isolates H40, H8, and H18 showed good antimicrobial against all tested pathogens with variable extents.
    5. The three bacterial isolates were identified morphologically, biochemically and by 16S rDNA gene sequencing as *Pseudomonas aeruginosa* strain H40, *Stenotrophomonas maltophilia* strain H8 and *Bacillus subtilis* strain H18. The sequence data for the 16S rDNA of bacterial strains (H40, H18 and H8) were

deposited in the GenBank under accession no. KF407990, KF407989, KF407991, respectively.

6. From the antimicrobial activity assay, it was observed that all three bacterial strains were highly effective against *Rhizoctonia solani*. The fungus was isolated from diseased cotton seedlings infected with damping-off disease.
7. Pot experiment was carried out to assess the ability of bacteria with different treatments to control *R.solani* on cotton seedlings under greenhouse conditions. The pot experiment results showed the following:
    1. Treating cotton seedlings with endophytic bacteria highly improved seedling emergence and survival rates and reduced damping-off disease severity.
    2. It also improved seedlings fresh and dry weights, root and shoot lengths and caused growth promotion in *R.solani* infested and non-infested soils.
    3. Some other parameters were measured such as defense enzymes (peroxidase, catalase and polyphenol oxidase) and total phenolic content and all were found to increase upon treatment with bacteria in both *R.solani* infested and non-infested soils.
    4. *Stenotrophomonas maltophilia* strain H8 was the most effective strain in the pot experiment

- The GC-MS analysis of the filtrates of the three endophytic bacterial strains revealed the presence of many metabolites with antimicrobial and antioxidant activities that were thought to be directly involved in the biocontrol activity of bacteria. Those compounds include the following:
    1. *Pseudomonas aeruginosa* H40 produced many metabolites that include cuminaldhyde, estragole, gentisic acid and geldanamycin.
    2. *Stenotrophomonas maltophilia* H8 produced metabolites include phthalic acid, mono-(2-ethylhexyl) ester, 3, 4-dimethoxycinnamic acid, imidazole and ferulic acid.

3. *Bacillus subtilis* H18 produced some metabolites that include imidazole and α-Bisabolol.

- In conclusion, endophytic bacteria are promising biological control agents that produce many biologically important metabolites that could be used widely.