Abstract:

Bacterial wilt, caused by the soil-borne phytopathogen *Ralstonia solanacearum*, is a persistent constraint on the productivity of many crops including tomato. With an aim to develop a novel sustainable strategy to control this devastating bacterium, the prospects of a seed-transmitted endophytic fungus were explored in this study. The endophytic *Penicillium* sp. PM031, isolated from the stem of a wilt-susceptible tomato cultivar was investigated in three different approaches to check its potentiality for managing this phytopathogen. Firstly, the metabolites from PM031 (ethyl acetate extract), containing ~80% of lipid molecules, showed concentration-dependent growth inhibitory activity against *R. solanacearum*. Moreover, the fungal extract caused membrane damage and negatively influenced the virulence factors necessary in early phases of bacterial infection, such as motility and biofilm formation. Secondly, the root immunity in the tomato plant was tested by employing the non-toxic concentration of PM031 extract as elicitor. Biochemical analyses revealed that the root priming evoked the levels of some defence-associated compounds and enzymes, such as total phenolics (39%), flavonoid (3.74-fold), lignin (43%), peroxidase (1.8-fold), and hydrogen peroxide. Importantly, the root priming significantly reduced the wilt symptoms in pathogen-infected plants by ~57% compared to the unprimed plants, implying that the induced disease resistance presumably occurred due to the enhanced oxidative response exerted by the primed roots. Lastly, through fungus-bacterium interaction, the scope of directly using PM031 as biocontrol agent was checked. Although *R. solanacearum*'s culture filtrate did not hamper PM031's growth; surprisingly, during direct confrontation, *R. solanacearum* overpowered PM031 as revealed by fungal growth reduction, changes in fungal membrane permeability, morphology, colonization of bacteria on fungal hyphae, changes in pH of the surrounding medium, etc. This observation led us to test a hypothesis on the signal-receptor-mediated interplay as a possible mechanism of how this bacterium senses the fungal presence. *In silico* analyses unveiled the allosteric interplay between SolR, a quorum sensing receptor of the bacterium, and ergosterol, a fungal biomarker acting as a possible signal molecule. Collectively, the findings of the present work demonstrated the prospect and reality of endophytic fungus to control *R. solanacearum*. Since inter-kingdom interactions are very dynamic and depend on multiple factors, this study opens up further scopes to develop new, rational, and sustainable ways to control bacterial wilt.

Keywords:

Antibacterial activity, endophytic fungus, fungal-bacterial interactions, fungal metabolites, host defence response, quorum sensing, root priming, wilt disease control