

Impact of Elevated Temperature on Spot Blotch Development in Wheat and Induction of Resistance by Plant Growth Promoting Rhizobacteria

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Abstract : Plants are constantly interacting with various abiotic and biotic stresses. In changing climate scenario plants are continuously modifying physiological processes to adapt to changing environmental conditions which profoundly affect plant-pathogen interactions. Spot blotch in wheat is a fast-rising disease in the warmer plains of South Asia where the rise in minimum average temperature over most of the year already affecting wheat production. Hence, the study was undertaken to explore the role of elevated temperature in spot blotch disease development and modulation of antioxidative responses by plant growth promoting rhizobacteria (PGPR) for biocontrol of spot blotch at high temperature. Elevated temperature significantly increases the susceptibility of wheat plants to spot blotch causing pathogen *Bipolaris sorokiniana*. Two PGPR *Bacillus safensis* (W10) and *Ochrobactrum pseudogrignonense* (IP8) isolated from wheat (*Triticum aestivum* L.) and blady grass (*Imperata cylindrical* L.) rhizosphere respectively, showing in vitro antagonistic activity against *Bipolaris sorokiniana* were tested for growth promotion and induction of resistance against spot blotch in wheat. GC-MS analysis showed that *Bacillus safensis* (W10) and *Ochrobactrum pseudogrignonense* (IP8) produced antifungal and antimicrobial compounds in culture. Seed priming with these two bacteria significantly increase growth, modulate antioxidative signaling and induce resistance and eventually reduce disease incidence in wheat plants at optimum as well as elevated temperature which was further confirmed by indirect immunofluorescence assay using polyclonal antibody raised against *Bipolaris sorokiniana*. Application of the PGPR led to enhancement in activities of plant defense enzymes- phenylalanine ammonia lyase, peroxidase, chitinase and β -1,3 glucanase in infected leaves. Immunolocalization of chitinase and β -1,3 glucanase in PGPR primed and pathogen inoculated leaf tissue was further confirmed by transmission electron microscopy using PAb of chitinase, β -1,3 glucanase and gold labelled conjugates. Activity of ascorbate-glutathione redox cycle related enzymes such as ascorbate peroxidase, superoxide dismutase and glutathione reductase along with antioxidants such as carotenoids, glutathione and ascorbate and osmolytes like proline and glycine betain accumulation were also increased during disease development in PGPR primed plant in comparison to unprimed plants at high temperature. Real-time PCR analysis revealed enhanced expression of defense genes- chalcone synthase and phenyl alanineammonia lyase. Over expression of heat shock proteins like HSP 70, small HSP 26.3 and heat shock factor HsfA3 in PGPR primed plants effectively protect plants against spot blotch infection at elevated temperature as compared with control plants. Our results revealed dynamic biochemical cross talk between elevated temperature and spot blotch disease development and furthermore highlight PGPR mediated array of antioxidative and molecular alterations responsible for induction of resistance against spot blotch disease at elevated temperature which seems to be associated with up-regulation of defense genes, heat shock proteins and heat shock factors, less ROS production, membrane damage, increased expression of redox enzymes and accumulation of osmolytes and antioxidants.

Keywords : antioxidative enzymes, defense enzymes, elevated temperature, heat shock proteins, PGPR, Real-Time PCR, spot blotch, wheat

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