GENES CONTROLLING THE INFECTION PROCESS OF SEPTORIA TRITICI BLOTCH PATHOGEN OF WHEAT

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Mycosphaerella graminicola (Fuckel) J. Schröt is the causal agent of *Septoria tritici* leaf blotch, which is the major foliar wheat disease in most temperate areas. Its infection process includes: dimorphic switch from yeast-like form to filamentous growth, penetration of the germtube through stomata, colonization of mesophyll cells, transition from biotrophic phase to necrotrophic phase and finally production of fruiting bodies called pycnidia. In this study we characterized 9 genes belonging to MAP kinase (MAPK) and cAMP pathways including three MAPKs, *MgHog1, MgFus3, MgSlt2,* the regulatory and catalytic subunit encoding genes of PKA, *MgTpk2, MgBcy1*, three Gα protein encoding genes, *MgGpa1*, *MgGpa2*, *MgGpa3*, and the Gβ encoding gene *MgGpb1*. *MgHog1* mutants were osmosensitive, highly resistant to the several fungicides and were unable to switch from yeast-like to filamentous growth. *MgHog1* mutants were impaired in dimorphic switch, failed to establish infectious germ tubes and therefore were unable to penetrate wheat leaves demonstrating that the dimorphic transition is a key factor in pathogenicity of *M. graminicola.* Disruption of *MgFus3* gene prevented melanization of mycelia and formation of pycnidia *in vitro. MgFus3* mutants are non-pathogenic. The mutants of *MgFus3* were non-pathogenic, which is ascribed to impaired penetration of stomata, possibly due to inability of the mutants to recognize stomata. In *M. graminicola*, *MgSlt2* plays a role in cell wall integrity since *MgSlt2* mutants were affected in polarized growth and showed progressive autolysis during aging. They were also hypersensitive to glucanase and several fungicides and did not produce aerial mycelium or melanin on potato dextrose agar (PDA). The *MgSlt2* mutants penetrated wheat stomata regularly, but were unable to establish invasive growth and did not produce asexual fructifications and hence their virulence was severely reduced. Because *MgSlt2* is involved in cell wall integrity, *MgSlt2* mutants are probably more sensitive to hitherto unknown plant defense compounds, which might explain the compromised colonization of mesophyll tissue. Fructification of *M. graminicola* is a complex process requiring proper differentiation of the infectious. *MgTpk2* and *MgBcy1* mutants were able to germinate, penetrate and colonize mesophyll tissue, but were unable to differentiate pycnidia. Our data provide evidence that the cAMP pathway regulates filamentation through *MgTpk2* and *MgBcy1*. Disruption of *MgTpk2* impaired filamentation. In addition, the *MgTpk2* mutants became melanized faster and secreted a dark-brown pigment into yeast glucose broth medium (YGB), whereas *MgBcy1* mutants showed delayed melanization on PDA and were osmosensitive. Overall, the divergent functions of the regulatory and the catalytic subunits of PKA indicate that proper regulation of PKA activity is required for various physiological processes including differentiation, filamentation, osmoregulation and melanization. *MgGpa1* formed fluffy mycelia in liquid medium and hardly produced spores. *MgGpb1* mutants showed a nested type of growth on PDA that resulted from hampered filamentation, numerous cell fusions and increased anastomosis. Therefore, we concluded that *MgGpa1* negatively regulates filamentation, which is positively regulated by *MgGpa3* and *MgGpb1*. Pathogenicity assays revealed that *MgGpa1*, *MgGpa3* and *MgGpb1* are required for virulence of *M. graminicola* whereas *MgGpa2* is dispensable. Based on our results we conclude that *M. graminicola* is an excellent fungal pathogen model to study molecular mechanisms regulating infection process.