

# Transcriptional regulation of iron homeostasis of the hemibiotrophic phytopathogen *Colletotrichum graminicola*

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Iron is an essential microelement for all organisms. Due to its low solubility combined with the potential to produce damaging highly reactive oxygen species, a tight regulation of iron uptake and storage is essential for all living cells.

Pathogenic fungi employ several strategies for iron uptake from the host tissue: (i) reductive iron assimilation (RIA), (ii) siderophore-mediated Fe<sup>3+</sup> acquisition (SIA), (iii) heme uptake, and (iv) low affinity iron uptake. As free heme is rare in maize - the host plant of *Colletotrichum graminicola* - this hemibiotrophic fungus mainly applies RIA and SIA. Saprophytic hyphae growth under iron starvation leads to an up-regulation of both RIA and SIA pathways. During the biotrophic stage of infection RIA is highly active, while SIA is specifically suppressed. The subsequent necrotrophic stage is characterized by a reversal in the iron uptake specificity. Maize leaves pretreated with the *C. graminicola* siderophore Coprogen respond with an increased defense reaction including respiratory burst when these leaves were infected later on. In contrast, Coprogen alone did not induce a defense response. This reveals that *C. graminicola* specifically represses the SIA pathway, possibly to evade plant recognition. During the necrotrophic phase such hiding is no longer required.

This strategy resembles the specific repression of the synthesis of  $\beta$ -1,3-Glucane, a pathogen associated molecular pattern (PAMP), during biotrophy.

In other fungal species the tight regulation of the SIA und RIA pathways occurs on transcriptional level mediated by two transcription factors SreA and HapX, respectively. However, the so far studied *Aspergillus* spp. were either necrotrophs or saprophytes. Here we report on the identification of *sreA* and *hapX* homologs from the hemibiotrophic fungus *C. graminicola* that were denominated as *CgSRE1* and *CgHAP10*. We showed that both genes are iron-dependent regulated on transcriptional level in saprophytic hyphae. Targeted deletions of these loci led to delayed growth in response to iron availability. Remarkably, the  $\Delta$ *sre1* strain showed altered hyphal morphology resembling cell wall-deficient mutants. Cell wall deficiency, therefore, could be responsible for the reduced virulence of  $\Delta$ *Cgsre1*.

Detailed functional characterization of the putative transcription factors *CgSre1* and *CgHap10* during biotrophic and necrotrophic stages will gain further knowledge of iron acquisition and regulation of iron homeostasis in fungal virulence and provide valuable data to develop novel plant protection strategies.