

Comparative analysis of host specificity and pathogen interaction of *Cochliobolus heterostrophus* on maize vs. non-host species

Understanding how pathogens successfully colonize host plants while failing to infect non-host species is an important question in plant pathology. Southern leaf blight, caused by *Cochliobolus heterostrophus*, threatens maize production globally. The fungus typically penetrates leaves between epidermal cells or through stomates, then establishes hyphal networks that infiltrate deep into the leaf tissue. While well-studied in maize, its interactions with non-hosts like wheat, sorghum, and tomato remain largely unexamined. We aim to determine whether *C. heterostrophus* recognizes maize-specific signals and adjusts infection strategies accordingly. After inoculation of spores on maize, tomato, wheat and sorghum genotypes, histological analysis indicated 100% spore germination across all species, but spore attachment was highest in maize, lowest in wheat and tomato, and intermediate in sorghum at 6 hours post inoculation (hpi). While the fungus successfully penetrated all species, albeit at a lower frequency in non-host species, post-penetration differences were evident. By 24–48 hpi, extensive fungal hyphal networks were seen in maize, restricted growth to a single cell in wheat, localized browning in sorghum, and tomato displayed minimal penetration. These findings suggest systemic colonization is host-dependent, with post-penetration resistance central to non-host interactions. We further performed RNA seq analysis of the fungal transcriptome when infected on maize, wheat, and sorghum at 6, 12, and 24 hpi. At 6 hpi, gene expression patterns looked broadly similar across host (maize) and non-hosts (wheat and sorghum) though with some subtle differences already present, before diverging strongly by 12–24 hpi when host species became the major driver of differences in fungal gene expression. In contrast to wheat and sorghum, the fungus in maize at 12 and 24 hpi displayed a higher expression of specific genes that may help the fungus colonize the host, including cell wall degrading enzymes, a secreted protease, genes involved in oxidative stress defense, and genes tied to rapid growth and nitrogen metabolism among others. These candidate colonization genes were not just higher in maize overall, but also increased in expression over time within maize, while remaining low and largely unchanged in wheat and sorghum. Fungal gene expression on wheat and sorghum showed a distinct set of genes tied to general stress and defense responses. The identified set of genes expressed in a host-specific manner provide a starting point to understand how maize-derived signals may influence gene expression in *C. heterostrophus*.