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Fusarium graminearum is an important pathogen of wheat and barley and a producer of DON. The *TRI* gene clusters responsible for DON synthesis have two transcription factor genes, *TRI6* and *TRI10*, but their functional relationship in transcriptional regulation of *TRI* genes is not clear. For *Tri10*, its DNA binding site has not been identified and it may have non-TF functions for DON production. In addition, the regulatory relationships of pH, nitrogen metabolism, and cAMP signaling with *Tri6* and *Tri10* in DON production are not well studied. In this study, we aim to determine the functional relationship between *TRI6* and *TRI10* and characterize molecular mechanisms involved in the regulation of DON production by pH, nitrogen metabolism, and cAMP signaling. **Objective 1** is to further characterize the interaction between *TRI6* and *TRI10*. Genes co-regulated by *TRI6* and *TRI10* also will be identified and functionally characterized. In **objective 2**, we will identify the *Tri10*-binding sites and characterize its possible non-TF function in the toxosome. For **objective 3**, we will use site-directed mutagenesis approach to determine the role of conserved PacC-binding sites in the *TRI6* and *TRI10* promoters. We also will characterize the effect of AreA phosphorylation by PKA on its interaction with *Tri10* and DON production.

Overall, results from proposed experiments will be helpful to better understand the regulatory networks involved in the regulation of DON production and plant infection. Various environmental or physiological factors such pH and nitrogen sources may converge on *Tri6* and/or *Tri10* with different mechanisms to regulate DON biosynthesis. Disruption or reduction of DON production can be used as a novel approach to control FHB. Proposed study fits the research area of PBG on developing new strategies for reducing impact of FHB and mycotoxin contamination. It is a new project based on recent progresses.