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Project Title: Control of FHB by Targeting Pathogen Effector and Host Protein Interactions

PROJECT 1 ABSTRACT (1 Page Limit)

The proposed research addresses both of the PBG research priorities for FY18-19: 1. Characterize plantfungal interactions to identify important genes, proteins or small molecules that may be used to develop FHB resistance or to reduce DON contamination in barley and wheat; and 2. Gain new understanding of initial fungal infection that may be utilized to boost FHB resistance.

The primary goal of this project is to aid in the development of FHB resistance and DON reduction by characterizing *Fusarium graminearum* effectors that interact with plant molecules to initiate infection and suppress plant defense responses. Effectors play an important role in the pathogenesis of many bacterial and fungal pathogens. As a hemibiotrophic pathogen, *F. graminearum* likely secretes effectors to suppress plant defenses during initial infection and induce cell death in later stages. We have selected thirty-seven putative effectors via bioinformatic analyses of genome sequences from sixty strains representing three different *F. graminearum* populations found in the U.S. (NA1, NA2 and NA3).

Our objectives are:

- 1. Determine the expression profile of selected candidate effectors during pathogenesis. RT-qPCR will be performed for gene expression studies.
- 2. Characterize the effect of candidate genes on FHB pathogenesis via mutagenesis experiments. Mutants will be created for selected effectors by split marker or *Agrobacterium*-mediated mutagenesis. The impact of mutations on pathogenesis will be evaluated on wheat and barley heads by dip inoculation (for type I resistance) and on wheat heads by point inoculation (for type II resistance). DON production of mutants will be determined using GC/MS and compared with controls.
- 3. Characterize effector-interaction partners. Once critical effectors associated with FHB pathogenesis are identified, effector localization in planta will be verified by GFP fusion and microscopic examination. Host protein targets of critical effectors will be pursued by co-immunoprecipitation assays.

The major outputs from the proposed research will be the identification of key effectors used by all, or individual pathogen populations to initiate infection and suppress plant immunity, and the identification and characterization of host protein targets of these effectors. These effectors and their interaction partners will be excellent targets for traditional breeding, RNAi or CRISPR/Cas9 genome editing that will enable USWBSI-funded scientists and others to develop transgenic plants that can block FHB pathogenesis and reduce mycotoxin contamination of grain.