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Research Category: GDER

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Project Title: A Centralized Barley Transformation Facility for the FHB Community

PROJECT 1 ABSTRACT

(1 Page Limit)

The goal of this project is to develop a barley transformation facility for the *Fusarium* head blight (FHB) community. Different protocols have been developed to transform barley. Most of the publications have focused on the transformation of barley cv. Golden Promise, which is not a prevalent cultivar in the U.S. Scientists in the USDA-ARS (Aberdeen, ID) have developed the transformation method for barley cv. Conlon, a two-rowed barley released by NDSU in 1996. We have further developed the transformation protocol for Conlon, ND Genesis and Morex. In our USWBSI-approved Project ID #FY18-DI-015, entitled "CRISPR-Gene Editing Barley to Improve *Fusarium* Head Blight Resistance", we propose to disrupt Conlon barley genes that condition susceptibility to *Fusarium graminearum* (*Fg*) infection, including *HvHSK*, *Hv2OGO* and *HvEIN2* encoding homoserine kinase, 2-oxoglutarate Fe(II)-dependent oxygenase and ethylene insensitive 2 respectively. CRISPR-editing with transiently-expressing gRNA/Cas9 DNA offers the advantage of multigene-targeting and producing transgene-free, gene-edited and FHB resistant barley plants.

The UDP-glucosyltransferase (UGT), a DON-detoxifying enzyme, is present in both the FHB-resistant and susceptible barley cultivars. Therefore, it is warranted to CRISPR-edit the promoter of *HvUGT* gene to study the kinetics of *HvUGT* gene expression in different barley cultivars. Many agronomic traits have been implicated in influencing the FHB development in barley, including the hullless vs. hulled and two-rowed vs. six-rowed. The hull and row types are controlled by transcription factors encoded by *Nud* and *Vrs1* genes which can be edited by CRISPR.

Our specific objectives for this 2-year project are: (1) Collaboration with Dr. G. Muehlbauer at University of Minnesota to edit the *HvUGT* promoter in the FHB susceptible barley cv. Morex to study the *HvUGT* gene expression kinetics, (2) Collaboration with Dr. B. Steffenson at University of Minnesota to edit the *Nud* gene in two-rowed and hulled Conlon and ND Genesis barley to study the effect of hull type on the FHB development, (3) Collaboration with Dr. B. Steffenson at University of Minnesota to edit the *Vrs1* gene in the six-rowed cultivar Morex to study the effect of row type on the FHB development, and (4) Collaboration with Dr. P. Hayes at Oregon State University to develop barley anther culture for CRISPR-gene editing and barley engineering.

The developed barley transformation systems will be used within the USWBSI to edit other genes or gene components or to introduce other transgenes to improve barley FHB resistance or to study barley gene functions in several different cultivars.