FY22 USDA-ARS/USWBSI Project ID: FY22-GD-003

## **Project Abstract**

Project Title:	A Barley Genetic Engineering Facility for FHB Research Community	
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The overall goals of this project are to establish a barley genetic engineering facility to provide a no-cost transformation service for the Fusarium head blight (FHB) research community; continue to develop and apply CRISPR-gene editing technology to discover genes involved in FHB susceptibility; and engineer FHB resistance in barley cultivars grown in the U.S. The barley genetic engineering facility will provide a counterpart to the centralized wheat transformation center headed by Dr. H. Trick. Currently, no such centralized facility exists to serve the needs of USWBSI barley researchers. The proposed barley genetic engineering facility will provide consultation services to help researchers design and construct transgenes and CRISPR-editing vectors; deliver constructs into immature embryo-derived calli *via* biolistic or *Agrobacterium* transformation; regenerate transformed/edited plants; screen putative transgenic T<sub>0</sub> plants and deliver seeds up to 10 T<sub>1</sub> plants to the users. Using funding provided by USWBSI since 2018, we have developed our own CRISPR-gene editing platform for barley genetic engineering. We have mostly used integrating CRISPR vectors delivered into cells of immature embryo explants to produce a significant number of transgenic barley plants in the U.S.-grown cultivars, Conlon, Genesis and Morex. This experience will ensure the success of this proposed barley genetic engineering facility for the FHB research community.

Using our CRISPR-editing platform, we have constructed both transient and integrating CRISPR-vectors to edit the barley genes *HvEIN2* (encoding ethylene insensitive 2), *HvHSK* (encoding homoserine kinase) and *Hv2OGO* (encoding 2-oxoglutarate Fe (II)-dependent oxygenase), each representing a potential FHB susceptibility gene as are their Arabidopsis counterparts. While having produced several lines of Conlon barley with *HvHSK*- and *Hv2OGO*-edited, gene-editing barley cv. Conlon, Genesis and Morex is not efficient. Therefore, we propose to continue our efforts to improve CRISPR-gene editing technology and use it to discover genes involved in FHB susceptibility and to engineer FHB resistance in barley cultivars grown in the U.S. Experience gained from this research effort will enhance our expertise and help us develop the capacity of the proposed barley genetic engineering facility to produce transgenic barley plants for barley researchers in the USWBSI community.

Our specific objectives and outcomes for this project are: 1. Establish a dedicated barley genetic engineering facility for the FHB research community, 2. Develop and make use of anther/microspore transformation and regeneration protocols for barley gene editing, 3. Continue our efforts to produce gene-edited Genesis and Morex plants to discover genes involved in barley susceptibility or resistance in FHB.