Project FY22-BA-018: Functional Validation of the Barley Fhb1 Ortholog in Susceptibility to FHB

1. What are the major goals and objectives of the research project?

- 1) To develop targeted gene knockouts in barley using CRISPR-mediated mutagenesis;
- 2) To obtain the transgene-free barley mutants with resistance to FHB.
- **2.** What was accomplished under these goals or objectives? (For each major goal/objective, address these three items below.)

What were the major activities?

- 1) Barley transformation using the amenable genotype Gold Promise;
- 2) Barley transformation using recalcitrant genotypes, such as Bowman and Conlon;
- 3) CRISPR-mediated mutagenesis in Golden Promise, Bowman, and Conlon;
- 4) Tail-PCR to identify homozygous and heterozygous CRISPR mutants;
- 5) Field testing using the homozygous mutants.

What were the significant results?

- 1) We developed a stable protocol for barley transformation in Golden Promise and Bowman.
- 2) We improved barley transformation efficiency in recalcitrant genotypes.
- 3) The barley ortholog of *FHB1* (*HvHRC*) in Golden Promise and Bowman was knocked out.
- 4) Homozygous mutant plants were obtained.
- 5) Knocking *HvHRC* in Bowman enhanced FHB resistance based on the 2023 field data.

List key outcomes or other achievements.

- 1) We successfully transformed recalcitrant barley genotypes.
- 2) We set up an efficient CRISPR protocol in barley.
- 3) Loss-of-function mutation of *HvHRC* enhanced FHB resistance based on the 2023 field data.

3. What opportunities for training and professional development has the project provided?

This project provided a Postdoc Researcher and an undergraduate student with training on barley transformation and gene editing.

4. How have the results been disseminated to communities of interest?

We presented our preliminary results at the 2023 National FHB Forum.

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5. What do you plan to do during the next reporting period to accomplish the goals and objectives?

Field test and greenhouse tests with more replicates will be conducted to confirm the preliminary data that *HvHRC* conditions FHB susceptibility.