BMB Graduate Group M.S. Defense

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Ackert Hall 324A

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The role of the Putative Transcription Factor Gene *WRKY55* in Wheat against Hessian Fly Infestation

Hessian fly [*Mayetiola destructor* (Say)] is one of the most economically damaging insect pests of wheat (*Triticum aestivum* L.) in the United States and worldwide, and host plant resistance conferred by dominant *Hessian fly resistance* (*H*) genes remains the most effective and durable strategy for its management. 37 *H* genes have been identified from wheat and its wild relatives, but the effectiveness of individual *H* genes is frequently eroded over time as Hessian fly populations constantly evolve, resulting in new, more virulent biotypes. Therefore, identifying additional broadly effective *H* genes and understanding the molecular pathways downstream of recognition remain priorities for durable genetic control. *Hdic* is a dominant Hessian fly resistance gene derived from cultivated emmer wheat and introgressed into the hard red winter wheat germplasm line 'WGRC42' (PI 635054), where it confers broad-spectrum antibiosis against multiple Hessian fly biotypes. A candidate gene underlying *Hdic* resistance has been identified, and two independent gene-edited mutant lines, T175 and T179, which lose resistance to Hessian fly, have recently been generated in our lab. The downstream transcriptional regulators that translate *Hdic*-mediated recognition into antibiotic defense responses have not been established. Comparative transcriptome analysis of WGRC42 and the susceptible mutant T175 identified several putative transcription factor genes that are differentially expressed between wild-type and gene-edited mutants. Among the differentially expressed transcription factor genes is the gene *WRGC42aa047820*, which encodes the putative transcription factor gene *WRKY55*. This study tested the hypothesis that *WRKY55* functions as one of the downstream mediators for Hessian fly resistance in wheat.

One-step reverse-transcription quantitative PCR (RT-qPCR) was used to confirm the differential expression patterns of *WRKY55* in feeding-site tissue among the wheat lines WGRC42, T175, and T179 across a Hessian fly infestation time course using the iTaq Universal SYBR Green One-Step Kit (Bio-Rad Laboratories, Hercules, CA). The role of *WRKY55* in wheat resistance to Hessian fly was functionally tested using the Barley stripe mosaic virus-induced gene silencing (BSMV-VIGS) in the WGRC42 background, with Hessian fly resistance phenotypes quantified by neutral red staining of the feeding site, larval size, larval survival, and whole-plant damage.

WRKY55 is likely to act as a downstream component of *Hdic*-mediated resistance against Hessian fly infestation. This work identifies a potential mechanistic link between R-gene recognition, transcriptional regulation, and specific chemical defense reactions, which provides a direction to further test the role of the *WRKY55* gene in plant resistance and potential applications in breeding for more durable Hessian fly resistance.