### Cover Page

<table>
<thead>
<tr>
<th>Principle Investigator (PI):</th>
<th>Gideon Marais</th>
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<tbody>
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<td>701-231-8155</td>
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<td>Fiscal Year:</td>
<td>2016</td>
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<tr>
<td>USDA-ARS Agreement ID:</td>
<td>59-0200-3-006</td>
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<tr>
<td>FY16 USDA-ARS Award Amount:</td>
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<tr>
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<td>Office of Grant &amp; Contract Accounting</td>
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<td></td>
<td>NDSU Dept 3130, PO Box 6050</td>
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<td>Project/Grant Reporting Period:</td>
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<td>Reporting Period End Date:</td>
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### USWBSI Individual Project(s)

<table>
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<tr>
<th>USWBSI Research Category*</th>
<th>Project Title</th>
<th>ARS Award Amount</th>
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<tr>
<td>HWW-CP</td>
<td>Development of Winter-hardy HRWW Lines with Pyramided QTL for FHB Resistance.</td>
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**FY16 Total ARS Award Amount** $27,190

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* MGMT – FHB Management  
  FST – Food Safety & Toxicology  
  GDER – Gene Discovery & Engineering Resistance  
  PBG – Pathogen Biology & Genetics  
  EC-HQ – Executive Committee-Headquarters  
  BAR-CP – Barley Coordinated Project  
  DUR-CP – Durum Coordinated Project  
  HWW-CP – Hard Winter Wheat Coordinated Project  
  VDHR – Variety Development & Uniform Nurseries – Sub categories are below:  
    SPR – Spring Wheat Region  
    NWW – Northern Soft Winter Wheat Region  
    SWW – Southern Soft Red Winter Wheat Region
Project 1: Development of Winter-hardy HRWW Lines with Pyramided QTL for FHB Resistance.

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

The NDSU HRWW breeding program aims to develop new varieties with improved cold-hardiness, regional adaptation, yield, disease resistance and processing quality. This USWBSI project focuses specifically on accelerating progress with FHB resistance breeding. In previous funding cycles, literature-validated resistance QTL (\(Fhb1, Qfhs.ifa-5A\), two QTL on chromosome 5A of PI277012 (here called \(QTL5A-1\) and \(QTL5A-2\)), a QTL on 3A of Frontana (here called \(QTL3A\)) and \(Fhb6\)) were transferred from spring wheat. This project aims to establish \(Fhb1\) as the baseline of FHB resistance in the breeding population and since 2016 we also develop and study simple gene pyramids consisting of \(Fhb1\) plus 1-2 of the remaining QTL to identify those that would add substantively to the effect of \(Fhb1\). In this context, the following gene pyramids were/are being produced and evaluated through marker analyses and greenhouse FHB resistance phenotyping: (a) Two-gene pyramids of \(Fhb1\) and \(Qfhs.ifa-5A\) (four lines) and (ii) \(QTL5A-1\) and \(QTL5A-2\) (two lines). (b) Two- and 3-gene pyramids of \(Fhb1\) with \(Qfhs.ifa-5A, QTL5A-1\) and \(QTL5A-2\). (c) \(Fhb1\) and \(QTL3A\) pyramids. (d) Two- and 3-gene pyramids of \(Fhb1\) with \(Fhb6\) and \(Qfhs.ifa-5A\).

2. What was accomplished under these goals? Address items 1-4) below for each goal or objective.

Accomplishments with respect to the overall project goal. The primary project aim is to substantially raise the frequency \(Fhb1\) and additional, useful resistance QTL in the crossing blocks and breeding population. Towards this end, and based on experience gained thus far, we involved at least one parent with at least one of \(Fhb1, Qfhs.ifa-5A\) and \(QTL5A-1\) in approximately 90% of the 2017 breeding program crosses. However, only a limited number of near-isogenic lines with \(Fhb1\) (including Norstar-\(Fhb1\), Wesley-\(Fhb1\), Decade-\(Fhb1\), Overland-\(Fhb1\) and Jerry-\(Fhb1\)) as well as inbred lines with \(Fhb1\) have thus far been acquired. A small number of inbred lines with \(QTL5A-1, Qfhs.ifa-5A\) or \(Fhb1\) plus \(Qfhs.ifa-5A\) have also been derived and used. In the absence of dependable markers for the two 5A QTL, their integration and identification is difficult and selection is based on artificial or natural infection of field planted populations. A further problem with the introgression of the new FHB resistance is that over-dependence on this small, initial group of FHB-resistant parents in consecutive crossing blocks will erode overall genetic and phenotypic variability of the program and limit future selection for non-FHB traits such as yield, adaptation, quality and resistance to diseases other than FHB. To counter these effects, new, diverse and desirable agrotypes (mostly lacking FHB resistance) from our and other programs/nurseries are annually obtained and used in crosses with the available FHB-resistant near-isogenic and selected pure lines. Their F1 are then used as FHB-resistant parents in the next season. In this manner, structured crosses are conducted with the long-term aim to continuously combine these resistances in more complicated combinations, while simultaneously broadening overall genetic variability.

Accomplishments – Pyramids of \(Fhb1\) with 5A QTL: HRWW lines derived from CM82036 were tested with published markers; four of these appeared to have both \(Fhb1\) and \(Qfhs.ifa-5A\), and another four lines had \(Fhb1\) only, while one line showed intermediate FHB resistance in the (Form – FPR16)
field, however, did not show any of the marker polymorphisms. Two lines that were derived from the HRSW donor line RWG21 (believed to have QTL5A-1 and QTL5A-2 from PI277012) produced marker polymorphisms that were not consistent with those in the donor material. Since none of the markers proved to be useful for the selection of the PI277012 resistance QTL, it was necessary to do disease phenotyping in an attempt to confirm the resistance. The resistance carrying lines plus resistant donors and susceptible parents were included in a greenhouse trial (6 replications) to test FHB type II resistance. Based on the marker-predicted genotypes and the resistance data, three lines (12DH172, 11M221-24-1 and 14K456-K-1) are believed to have both Fhb1 and Qfhs.ifa-5A. The three lines showed resistance similar to the HRSW donor line CM82036, and significantly stronger resistance than lines with Fhb1 only. Thus, Fhb1 and Qfhs.ifa-5A QTL do have additive effect. With respect to QTL5A-1 and QTL5A-2, PI277012 showed strong resistance, which was similar to that of CM82036. However, the RWG21 (derived from PI277012) plants that were included as controls in the FHB trial showed no resistance and in fact had the most severe infection of the 15 lines tested in the trial. The two RWG21 progeny lines (11M228-19-1 and Novus-4), showed intermediate resistance. Thus, it appeared that the RWG21 germplasm line was heterogeneous and segregated at one or both of the QTL5A-1 and QTL5A-2 loci.

An attempt was then made to combine resistance QTL present in Novus-4 (QTL5A-1 and/or QTL5A-2) and 14K456-K-1 (Fhb1 and Qfhs.ifa-5A). 400 F2 from a cross of the two lines were screened with Fhb1 and Qfhs.ifa-5A markers and two sets of plants were identified, i.e. (i) 17 F2 homozygous for Fhb1 only, and (ii) 19 F2 homozygous for both Fhb1 and Qfhs.ifa-5A. Within each set of F2 families, four F3 plants per family were grown and leaves were cut on each for doing 9K SNP analyses. This was done in an attempt to find additional, mapped chromosome 5A marker loci that could aid in the interpretation of the data. The 144 F4 sub-families were then evaluated for FHB type II resistance in a replicated greenhouse trial. The data from this experiment are presently being analyzed. Preliminary indications are that QTL5A-1 from PI277012 occurs within the same chromosome region and produces a similar effect to Qfhs.ifa-5A. Like Qfhs-ifa-5A it appears to add to the Fhb1 effect and Fhb1 plus Qfhs-ifa-5A pyramids appears to be similar to Fhb1 plus QTL5A-1 pyramids. However, due to the suspected heterogeneity of the donor source, RWG21, the second PI277012 locus, QTL5A-2, has not been transferred to our winter material. A new attempt therefore needs to be made to also transfer and evaluate QTL5A-2. In addition to the above information, the experiment yielded numerous inbred lines each homozygous for two FHB QTL. We aim to plant these lines in the field in September of 2017 and to continue to evaluate them as part of the routine breeding program.

Accomplishments – Pyramids of Fhb1 with QTL3A: A near-isogenic line, Norstar-Fhb1, was crossed with the F1: Frontana (QTL3A)/Norstar and the F1 marker screened to identify dihybrid plants. Following their self-pollination, 200 F2 progeny were marker-screened to identify the Fhb1 homozygotes. The selected homozygotes were then tested with a QTL3A marker and 34 F3 families that are homozygous for both QTL were identified. Since Frontana is a HRSW, the lines also segregate for winter habit and winter types need to be identified before the two groups of homozygotes will be compared in a greenhouse trial during 2018.

Accomplishments – Pyramids of Fhb1 with Qfhs.ifa-5A, and Fhb6: F1 heterozygotes from the cross: Fhb6/Jerry//Accipiter were crossed with 12DH172 (Fhb1 and Qfhs.ifa-5A). The F1 was
marker-screened to identify trihybrid \((Fhb1, Qfhs.ifa-5A, Fhb6)\) plants. \(F_2\) have been derived and will be screened in 2018 to identify \(\pm 100\) \(Fhb1\) homozygotes. The latter homozygotes will then be marker screened in an attempt to derive selections homozygous for \(Fhb1\) only, \(Fhb1 \& Qfhs.ifa-5A; Fhb1 \& Fhb6; and Fhb1 \& Qfhs.ifa-5A \& Fhb6\). Such plants will be used for comparison of the QTL effects in a greenhouse FHB trial.

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

   The project accommodates a PhD student.

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

   The initial project results were too incomplete for publication or presentation at the National Fusarium Head Blight Forum meeting of 2016.
Training of Next Generation Scientists

Instructions: Please answer the following questions as it pertains to the FY16 award period. The term “support” below includes any level of benefit to the student, ranging from full stipend plus tuition to the situation where the student’s stipend was paid from other funds, but who learned how to rate scab in a misted nursery paid for by the USWBSI, and anything in between.

1. Did any graduate students in your research program supported by funding from your USWBSI grant earn their MS degree during the FY16 award period?  
   No

   If yes, how many?

2. Did any graduate students in your research program supported by funding from your USWBSI grant earn their Ph.D. degree during the FY16 award period?  
   No

   If yes, how many?

3. Have any post docs who worked for you during the FY16 award period and were supported by funding from your USWBSI grant taken faculty positions with universities?  
   No

   If yes, how many?

4. Have any post docs who worked for you during the FY16 award period and were supported by funding from your USWBSI grant gone on to take positions with private ag-related companies or federal agencies?  
   No

   If yes, how many?
**Release of Germplasm/Cultivars**

**Instructions:** In the table below, list all germplasm and/or cultivars released with full or partial support through the USWBSI during the FY16 award period. All columns must be completed for each listed germplasm/cultivar. Use the key below the table for Grain Class abbreviations. *Leave blank if you have nothing to report or if your grant did NOT include any VDHR-related projects.*

<table>
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<tr>
<th>Name of Germplasm/Cultivar</th>
<th>Grain Class</th>
<th>FHB Resistance (S, MS, MR, R, where R represents your most resistant check)</th>
<th>FHB Rating (0-9)</th>
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Add rows if needed.

**NOTE:** List the associated release notice or publication under the appropriate sub-section in the ‘Publications’ section of the FPR.

**Abbreviations for Grain Classes**
- Barley - BAR
- Durum - DUR
- Hard Red Winter - HRW
- Hard White Winter - HWW
- Hard Red Spring - HRS
- Soft Red Winter - SRW
- Soft White Winter - SWW
Publications, Conference Papers, and Presentations

Instructions: Refer to the FY16-FPR_Instructions for detailed instructions for listing publications/presentations about your work that resulted from all of the projects included in the FY16 grant. Only include citations for publications submitted or presentations given during your award period (7/1/16 - 6/30/17). If you did not have any publications or presentations, state ‘Nothing to Report’ directly above the Journal publications section.

NOTE: Directly below each reference/citation, you must indicate the Status (i.e. published, submitted, etc.) and whether acknowledgement of Federal support was indicated in publication/presentation. See example below for a poster presented at the FHB Forum:


Status: Abstract Published and Poster Presented

Acknowledgement of Federal Support: YES (poster), NO (abstract)

Journal publications.

None

Books or other non-periodical, one-time publications.

None

Other publications, conference papers and presentations.

None