## Project 2: Assessment of Fungicide Sensitivity in Field Populations of Fusarium Causing FHB

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

Fungicides are commonly used as part of an integrated management plan to reduce Fusarium head blight caused predominantly by the fungus, *F. graminearum*. In 2019, Syngenta released Miravis<sup>®</sup> Ace, a premix fungicide that contains an active ingredient in the "second generation" succinate dehydrogenase fungicide class (adepidyn (pydiflumetofen); SDHI, FRAC group 7) and a triazole, for use in suppressing FHB. This is the first fungicide containing an active ingredient other than a DMI labelled for use in suppressing FHB; however, additional second-generation SDHI active ingredients have been used for several years to suppress numerous fungal diseases in wheat and cropping systems such as corn and soybeans. SDHI resistance has been observed in other pathosystems, including diseases in wheat and barley; however, no studies have examined populations of *F. graminearum*. The goal of this project was to generate baseline sensitivities for pydiflumetofen in *F. graminearum* populations across wheat and barley production regions in the US. Objectives include:

- 1) Establish centralized testing locations and protocols for fungicide sensitivity testing for Fusarium isolates as part of the USWBSI
- 2) Develop baseline sensitivity and associated virulence of current and historic isolates of *Fusarium* to SDHI and DMI fungicides collected from FHB symptomatic wheat in US wheat production areas
- 3) Place unique and/or valuable isolates into a national storage facility to facilitate collaboration between MGMT and PBG RACs
- **2.** What was accomplished under these goals or objectives? (For each major goal/objective, address these three items below.)

#### What were the major activities?

<u>Objective 1</u>

In project year 1 (2022-2023), Dr. Martin Chilvers and I established centralized testing at each of our respective locations (Figure 6). Video calls and virtual meetings have been continued to coordinate email correspondence for sample request, isolate collections, and approaches for fungicide sensitivity screening. Samples from extension specialists across the US have been solicited via email and through awareness at presentations during the spring 2023 NCERA184 meeting.

#### FY23-YR2 USDA-ARS/USWBSI Performance Progress Report PI: Koehler, Alyssa | Agreement #: 59-0206-2-111

- Samples from Midwest and Pacific Northwest:
  - Dr. Martin <u>Chilvers</u>, Michigan State University
  - chilvers@msu.edu

# • Samples from Southern and East Coast Regions:

- Dr. Alyssa Koehler, University of Delaware
- akoehler@udel.edu



Figure 6: Centralized testing locations at the University of Delaware (blue) and Michigan State (green)

#### Objective 2

Base line EC50 values from 2019-2020 have been established. From the original 16 states, mycelial growth EC50 values averaged by state and year(s) ranged from 0.16 to 0.56  $\mu$ g/mL, cumulatively averaging 0.37  $\mu$ g/mL and 0.31  $\mu$ g/mL in 2020 and 2021, respectively. The average EC50 value of historic isolates was 0.27  $\mu$ g/mL. These values have been presented in a manuscript that will be submitted fall 2024. We now continue to solicit isolates and improve screening methods for more high throughput screening by moving towards a discriminatory dose assay rather than full EC50 panel.

#### Objective 3

Dr. Chilvers and I have been in touch with USDA colleagues and there have been multiple zoom meetings to connect across projects and determine how to best approach isolate storage. Due to space limitation, we will need to prioritize valuable isolates and we will work across the MGMT and PBG RACS to characterize isolates and make these decisions regarding long term storage if/when isolates with reduced sensitivity are identified.

#### What were the significant results?

In 2022, our testing location received 279 isolates from six states (KS, KY, LA, MD, NC, PA). In 2023, 40 representative isolates were screened using a new YBA media. From this set EC50 values ranged from 0.04 µg/mL to 3.3 µg/mL. Since this range was wider than what had previously been observed, we began a series of experiments to further compare YBA and PDA media. The main advantage of YBA is that *Fusarium* growth is more symmetric, but there are tradeoffs in visualization as the lighter hyphae benefit from backlighting that is not needed when rating bright pink plants (Figure 8). Data was best analyzed by four parameter EC50 models and YBA plates saw a spread across more concentrations, which provided greater accuracy in fitting the model (Figure 9). PDA values trended higher, which indicated that the original 2023 YBA screen may have had isolates with higher EC50 values rather than being an artifact of media selection. Based on our continued testing, we will proceed with YBA as the primary screening media for future trial years. Media type did not differ at a level great enough to change the discriminatory dose selection, but we plan to run one more set of isolates in a full

EC50 panel before setting the first discriminatory dose. Our goal is to move towards discriminatory dose screening for more high throughput screening allowing us to test greater isolate numbers. 2023 was a very low scab year across the US. In total, we received 123 isolates including OH (n=28), DE (n=12), KY (n=26), NY (n=57 from 2022). In 2024 we plan to screen all remaining 2022-2024 isolates by establishing a YBA discriminatory dose.



Figure 7: Range of EC50 values in 2022 isolates from six states



*Fusarium graminearum* isolate growing on Yeast <u>Bacto</u> Agar (top) and Potato Dextrose Agar (bottom).

Figure 8: Comparison of growth on YBA versus PDA



Figure 9: Comparison of inhibition on YBA and PDA media types

#### FY23-YR2 USDA-ARS/USWBSI Performance Progress Report PI: Koehler, Alyssa | Agreement #: 59-0206-2-111



Figure 10: Distribution of EC50 values in YBA and PDA media types

#### List key outcomes or other achievements.

Two centralized testing locations are in operation and we have continued to improve the protocols for fungicide sensitivity testing. Our next step will be to select a discriminatory dose screening value to enable screening of more isolates in an efficient high-throughput manner.

Baseline efficacy within wheat and malting barley isolates have begun to be established across multiple states and in comparison to historic isolates. Two publications will be submitted in 2024 to provide a foundation for continued work.

Conversations have been initiated to discover the process and resources available to secure high value isolates in national storage facilities and to work across MGMT and PGB projects to maximize utility of isolate collections.

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

This project is currently supporting research technician gaining new skills in fungicide efficacy screening. Our lab has collaborated with the post doc in Dr. Chilvers lab to standardize EC50 data analysis and modeling. All previous EC50 screening values were updated to standardize model fitting protocols and analysis methods across labs during this period.

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

Updates on fungicide sensitivity were disseminated through articles in the University of Delaware's Weekly Crop Update, which reaches over 700 growers, consultants, and stakeholders and provides a platform to discuss disease concerns and other production issues. An overview of this project was presented at a Scabinar Event in March 2024. Updates on project results were also highlighted at Delaware Ag Week held in January 2024 and at the NCERA184 meeting in February 2024. Once manuscripts are published, research summaries will be prepared for publication with the Crop Protection Network to further broaden the audience receiving project results.

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

We will continue to solicit and collect isolates from across our testing region. The past two seasons have been quite low FHB pressure, but it seems that spring 2024 pressure was elevated in many states, so we anticipate higher sample numbers to be submitted in 2024. We aim to run another full EC50 panel and then make a selection for discriminatory dose assays to expand the number of isolates screened.