Name of Organization: National Agricultural Genotyping Center (NAGC)

Project Title: Regional Patterns of Herbicide Resistance Traits in Pigweed Escapees.

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Overview

Pigweeds within the genus *Amaranthus* are some of the most economically destructive weeds in row-crop systems. The fading effectiveness of chemical control is an increasing concern, especially with many pigweed populations containing individuals with herbicide resistance (HR). These concerns have produced a flurry of research in pigweeds, which have discovered US pigweed populations contain resistance for up to nine different modes of action. The race to understand the mechanisms of HR have largely focused on two species: waterhemp and Palmer amaranth. Last year NAGC conducted a genotyping validation study using DNA extracted from North Dakota pigweeds, which uncovered genotypes associated with resistance to glyphosate and PPO-inhibiting herbicides. These preliminary results led to this year's project to characterize the genetics of HR pigweed populations that escaped herbicide applications in eastern North Dakota.

Completed work

Field Collection – Ten surveyors collected late-season pigweed escapes from 12 counties in North Dakota (Fig. 1). The severe drought conditions in central and western ND reduced pigweed sightings, resulting in collections from the eastern one-third of North Dakota. Nevertheless, 53 pigweed populations are represented in this project, which mostly consist of waterhemp (*n*=28), and various monecious pigweeds (e.g., Powell amaranth, redroot pigweed). One population of Palmer amaranth was also included. Seed heads were collected from multiple mature plants within each population.

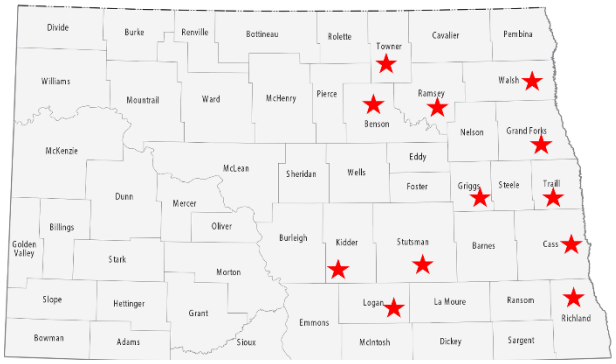

Figure 1. Red stars indicate ND counties where pigweed populations were sampled.

Greenhouse (NDSU) – Seed heads from the collected pigweeds were shipped to Dr. Ikley from September through November. The seed heads were threshed and the seeds currently reside at 4°C for stratification prior to sowing in the greenhouse. Dr. Ikley's lab is performing preliminary germination tests in preparation for the greenhouse experiment.

Genotyping (NAGC) – Currently, four high-throughput genotyping tests are ready for this project. They include two for the different target-site mechanisms for glyphosate resistance

(EPSPS-copies and EPSPS-106) and two for target-site mechanisms for PPO-inhibiting herbicide resistance (G210 and R128G). We are also independently validating two more PPO-inhibiting HR tests (R128M and G399A). In addition to the published tests, NAGC has developed a workflow to sequence HR pigweeds from the upcoming greenhouse treatments to look for other genetic changes within the PPO and ALS genes. Preliminary data to test the sequencing workflow found that North Dakota Palmer amaranth and waterhemp samples possessed genetic changes associated with ALS-inhibiting herbicide resistance.

Work to be Completed

Greenhouse (NDSU) – Pigweed seeds from each population will be sown in late December with applications of herbicides expected to start by late January. For genotyping, a single leaf will be collected from pigweeds prior to treatment at the 2 to 3 inch growth stage. Treatments will consist of seedling groups that will test two rates (1X and 3X) of glyphosate, imazamox, and fomesafen herbicides. The treatment response of seedlings from each population will be compared to susceptible controls at 21 days post-treatment.

Genotyping (NAGC) – We will continue to validate the two additional PPO genotyping tests and further optimize the sequencing workflow that will be used on the greenhouse-grown pigweeds. NAGC has been provided tissue samples from the field-collected parental plants, which can also be genotyped at the HR markers. In terms of genetic characterization, Waterhemp and Palmer amaranth dominate the HR research in other studies. Most of the novel genotyping work and test optimization left to accomplish by NAGC is within the monecious pigweed group, specifically redroot pigweed and Powell amaranth. Finding optimal sequencing primers for these species at PPO and ALS genes will help the search for target-site mechanisms that may result in herbicide resistance.