

PROGRESS REPORT

Investigation of Genetic Basis of 2,4-D and Dicamba Resistance in Palmer amaranth

Objective: Understand the genetic basis of 2,4-D and dicamba resistance in Palmer amaranth by the classical breeding approach.

Progress made: 2,4-D dose-response experiments confirmed that the AHR (auxinic herbicide-resistant) Palmer amaranth is 8 to 10 fold more resistant compared to two known auxinic herbicide susceptible (AHS) palmer amaranth populations (see progress report submitted January 2020). Also, the AHR Palmer amaranth was found resistant to six modes of action of herbicides, which makes this population an excellent resource to investigate what predisposes this population to evolve resistance to multiple herbicides. We have preliminary data on mechanisms of resistance to 2,4-D in this population. Our results indicate that the AHR Palmer amaranth plants are metabolizing 2,4-D faster than the AHS plants and there is no significant difference in herbicide uptake or translocation between AHR or AHS plants.

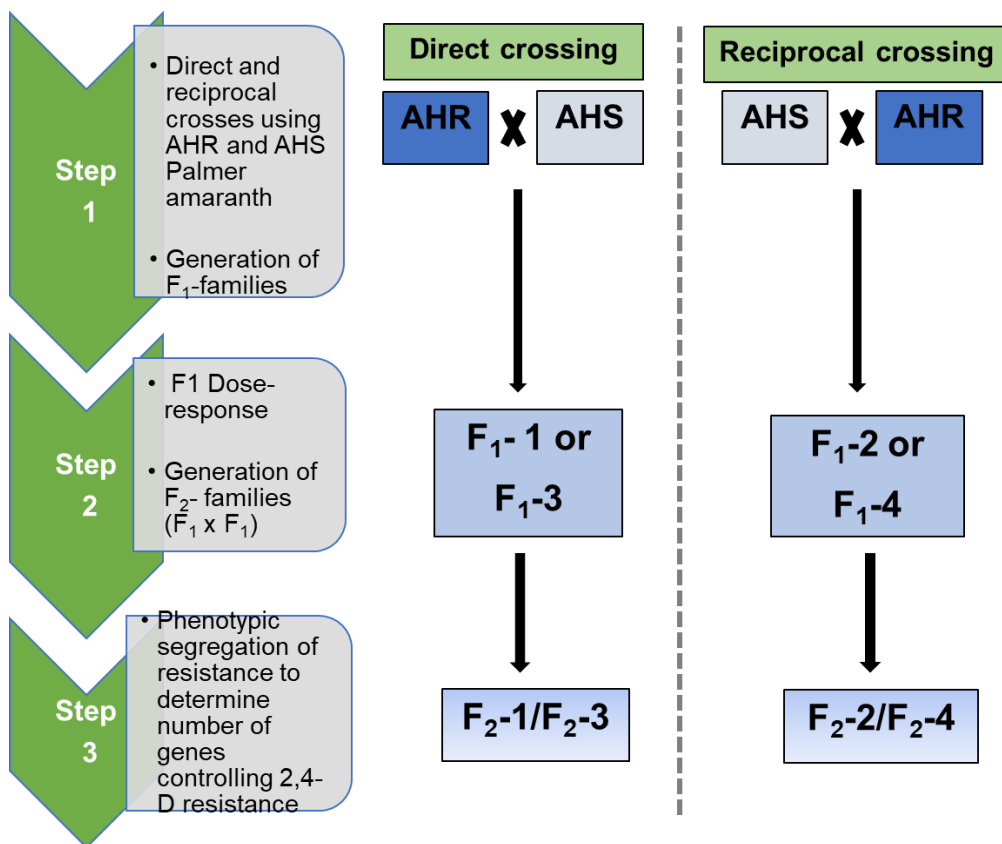
Generation of F1 progeny and determination of the response of F1 progeny to 2,4-D: F1 progeny were generated from the reciprocal crosses between AHR and AHS Palmer amaranth as follows. Male (♀) and female (♂) of AHR plants that survived 2,4-D application were used to perform direct (♀ AHR x ♂ AHS) and reciprocal crosses (♀ AHS x ♂ AHR) with susceptible AHS, to generate F1-families. In total, four families were generated from these direct (F1-1, F1-3) and reciprocal (F1-2 and F1-4) crosses.

2,4-D dose-response of F1 progenies: Two dose-response experiments were performed separately using seedlings from parental populations i.e., AHR, AHS, and F1-1 or F1-3 progenies. Ten-12 cm tall seedlings were then treated with 2,4-D at 0 (non-treated control), 140, 280, 560 (1x= field recommended rate), 1120, and 2240 g ae ha⁻¹ and six replications were maintained for each biotype and treatment combination. Treated plants were returned to the growth chambers within 30 minutes of 2,4-D treatment and were harvested at 4 weeks after treatment (WAT). During harvesting, above-ground plant biomass was collected, oven-dried, and weighed. These experiments were performed in a completely random design. The biomass data from each dose-response and for each biotype was adjusted to the non-treated control plants using the following formula: Relative shoot biomass (%) = [(Shoot biomass of treated plant/Average shoot biomass of non-treated plants)] x 100. This relative shoot biomass data was analyzed using three-parameter log-logistic regression in R-studio 3.63. From this analysis, the GR₅₀ (the amount of 2,4-D required to cause 50% biomass reduction to the plants in comparison to the control treatments) was calculated. Using GR₅₀ values, the resistance index (RI) was also calculated as a ratio of GR₅₀ of AHR or F1 to GR₅₀ of AHS.

Results: Analysis of the relative shoot biomass data from the F1-1 dose-response indicated that GR₅₀ of AHR, AHS and F1-1 were 560.3, 113.9, 501.3 g ae ha⁻¹., respectively. This suggests that the GR₅₀ of F1-1 progenies is close to the parental AHR Palmer amaranth. These results indicate

that 2,4-D resistance in F1-1 is governed by completely dominant allele(s). Contrary to that, dose-response from the F1-3 progenies showed that the GR50 of AHR, AHS, and F1-3 were 889.9, 440.2, and 166.3 g ae ha⁻¹, respectively. Since the resistance level of F1-3 is intermediate to parental AHR and AHS, it suggests that 2,4-D resistance in F1-3 progenies is governed by incompletely dominant allele(s). These experiments will be repeated two more times to confirm if 2,4-D resistance in Palmer amaranth is a completely or incompletely-dominant trait.

Generation of F2 progeny to determine the number of genes controlling 2,4-D resistance: Several F1 progenies that survived higher doses of 2,4-D (> 1120 g ae ha⁻¹) in the above dose-response study were also selected to generate pseudo-F2 progenies to determine how many genes control 2,4-D resistance in AHR Palmer amaranth. Experiments are in progress to generate F2 progenies. Figure (below) illustrates the generation of F1 and F2 progenies in Palmer amaranth to determine the inheritance of 2,4-D resistance.



Work in progress: Repetition of F1 dose-response experiments to confirm if 2,4-D resistance in AHR Palmer amaranth is a completely or incompletely dominant trait. Additionally, F2 progenies will be grown and treated with several doses of 2,4-D to determine the number of genes involved in 2,4-D resistance via phenotypic segregation.