Is Detoxification Contributing to PPO Inhibitor Resistance in Waterhemp (*Amaranthus tuberculatus*)?

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Introduction

- PPO inhibitor resistance (PPO-R) in waterhemp threatens the long-term utility of PPO inhibiting herbicides[1].
- A 2016 survey of PPO-R in the Midwest revealed a diversity of resistance responses from 4- to 17-fold that was unexplained by presence of R128 mutations, R allele zygosity, or frequency of resistant individuals (Figure 1).
- Non-target site resistance alleles can be a large contributor to overall resistance phenotype in the same individuals carrying target site resistance alleles[2].

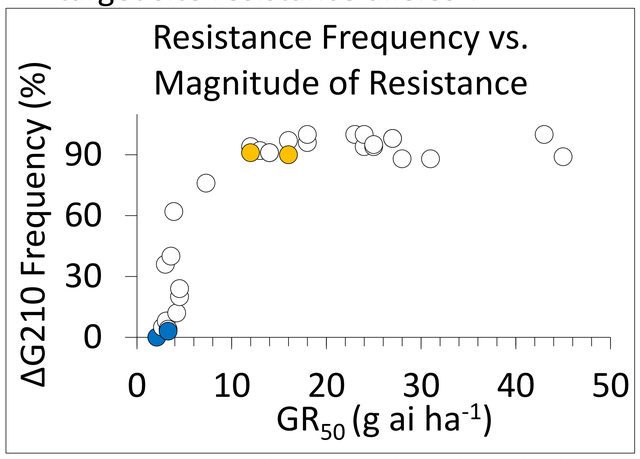

Figure 1. Resistance frequency of waterhemp populations as explained by GR_{50} . Resistance frequency accounts for a large portion of the change in GR_{50} until approximately 10 g ha^{-1} (R^2= 0.52). Blue dots indicate known S populations and yellow dots indicate known R populations.

- The compounds NBD-Cl and Malathion inhibit Glutathione S-transferases and Cytochrome P450s[3], respectively.
- These Phase I and Phase II metabolism inhibitors have been used to demonstrate detoxification (detox) based herbicide resistance.[4-7]

Hypothesis and Objective

Hypothesis: Enhanced fomesafen detoxification is contributing to the increased resistance response in two populations from the 2016 survey (IL-WAS and IN-DUB).

Objective: Determine the potential for co-occurrence of metabolic and target site resistance to PPO-inhibitors in two waterhemp populations.

Materials and Methods

- Waterhemp from six populations was germinated in flats and then transplanted into 3.8 x 25cm tubes.
  - Susceptible (IN-RAN1 and IA-340)
  - Moderately resistant (IL-BRO1 and IN-PIKE1)
  - Highly resistant (IL-WAS and IN-DUB)
- Plants were sprayed with either NBD-CL (270 g ha^{-1}), malathion (1500 g ai ha^{-1}), NBD-Cl fb malathion, or blank DMSO solvent solution.
  - NBD-Cl and DMSO solvent solution applied 2d prior to fomesafen application.
  - Malathion applied 2h prior to fomesafen application.
- Plants were sprayed with rates of 0, 20, or 60 g ai ha^{-1} of fomesafen at the 5- to 7-leaf stage.
- Waterhemp control, height, and biomass data were collected at 7 and 14 Days after fomesafen treatment.
- Data were analyzed using PROC GLIMMIX in SAS 9.4 to conduct a three-way ANOVA.
- Means separated using Tukey's HSD at α=0.05.

Results and Discussion

- Noticeable stunting was present for detox-inhibitor treatments similar to other groups[6,7] and was variable by population (Figure 2).
- At the 0 and 20 g ha^{-1} rate, NBD-Cl + Malathion increased fomesafen efficacy over all populations by 15 and 10 percentage points, respectively (Figure 3).
- At the 60 g ha^{-1} rate, NBD-Cl + Malathion increased control of IL-WAS by 20 to 26 percentage points compared to DMSO blank and NBD-Cl alone (Figure 4, Figure 5).

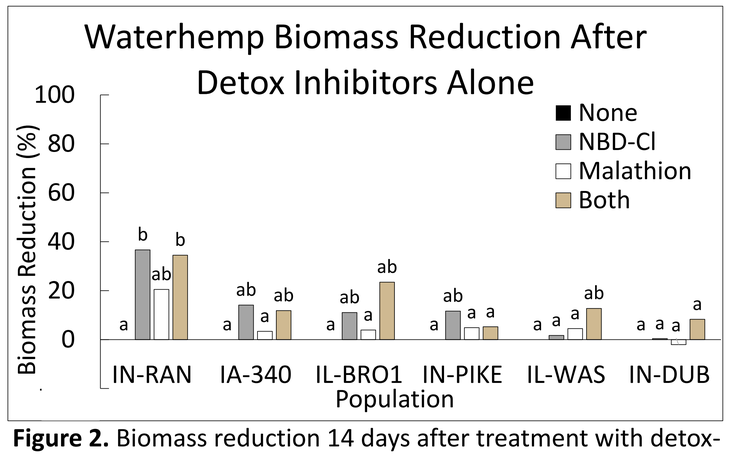

Figure 2. Biomass reduction 14 days after treatment with detox-inhibitors NBD-Cl and Malathion on waterhemp from six populations. Bars with different letters indicate significant difference between treatments.

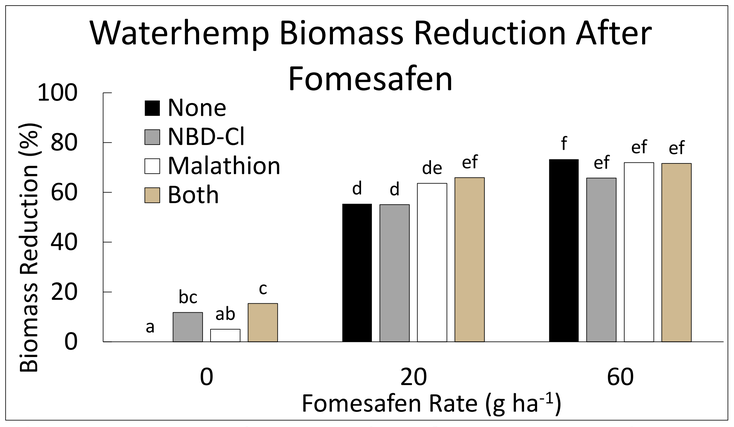

Figure 3. Biomass reduction 14 days after treatment with detox- inhibitors in combination with fomesafen. Bars with different letters indicate significant difference between treatments.

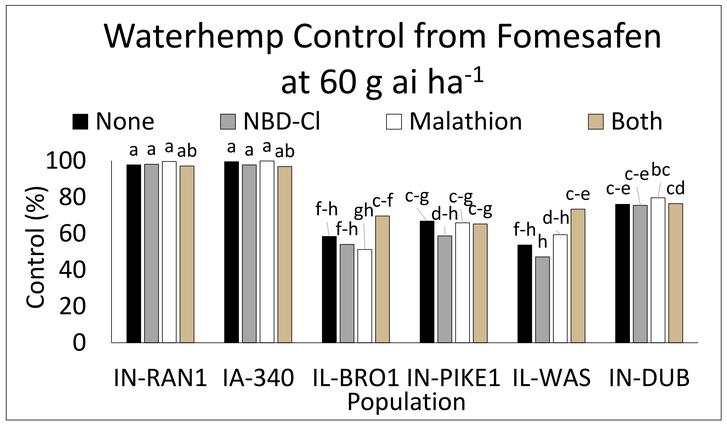

Figure 4. Waterhemp control 14 days after application of 60 g ai ha^{-1} of fomesafen and detox-inhibitor treatment. Bars with different letters indicate significant difference between treatments.

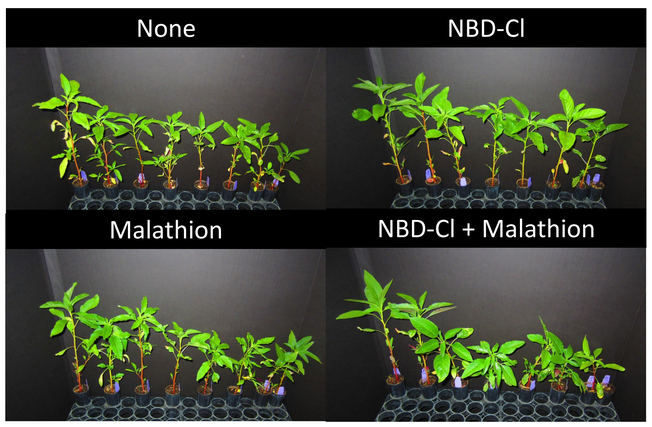

Figure 5. IL-WAS sprayed with detox-inhibitor treatments followed by 60 g ha^{-1} of fomesafen.

Conclusions

- Data were inconclusive with current methods and the hypothesis could not be fully tested.
- Evidence of detoxification based resistance should be pursued with additional methods such as LC-MS quantification of herbicide or detection of ^{14}C labeled herbicide metabolites.

Future Research

- Perform LC-MS analysis of fomesafen degradation through time in resistant populations.
- Investigate the variable response of different populations to malathion and NBD-Cl.

References

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