

# **WAR AGAINST WEEDS: SILVER BULLETS ARE FOR WEREWOLVES**

*(AKA COVER CROPS, HERBICIDE FATE, AND RESISTANT WEEDS)*

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**K-STATE**  
Research and Extension

2023 NDSU  
Advanced Crop Advisors  
Workshop

**NDSU** | WEED SCIENCE

# How cover crops suppress weeds

- Alter moisture, temperature during weed seed germination
- Outcompete emerging weeds for light, water, and nutrients
- Release allelochemicals that inhibit weed seed germination



# Take aways

- Stop grazing while optimum CC growing conditions are still present (adequate moisture with 15-20°C).
- Grazing 4.2 AU ha<sup>-1</sup> until CC biomass reaches 135 kg ha<sup>-1</sup> does not reduce winter annual weed suppression.
- Farmers should be cautious when grazing CC and attempting to control summer annual weeds. Stocking density at 0.5 AU ha<sup>-1</sup> with CC biomass less than 1060 kg ha<sup>-1</sup> reduces weed suppression.



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# Planting Green Trial

Planted Rye in September 2020, 2021

## Factorial Treatment Structure

- Rye management
  - No rye, Rye terminated 14 DPP, Rye terminated at planting (plant green)
- PRE (Fierce EZ @ 6 fl oz/A)
  - Pre at planting, no PRE
- Planting Date
  - “Standard planting” (~Mid May), “late planting” (14 days later)

# Planting Green Trial

Rye management, PRE?, Planting Date	POST Treatment
No rye, No PRE, Standard Planting	Enlist One (2 pt) + Liberty (2 pt) + Warrant (3 pt) @ 4" waterhemp  POST timing differed based on treatment
No rye, PRE, Standard Planting	
No rye, No PRE, Late Planting	
No rye, PRE, Late Planting	
Term rye 14 DPP, No PRE, Standard Planting	
Term rye 14 DPP, PRE, Standard Planting	
Term rye 14 DPP, No PRE, Late Planting	
Term rye 14 DPP, PRE, Late Planting	
Plant green, No PRE, Standard Planting	
Plant green, PRE, Standard Planting	
Plant green, No PRE, Late Planting	
Plant green, PRE, Late Planting	

# Planting Green Trial



- Data Recorded:
  - Days from plant to 4” waterhemp (Enlist One + Liberty + Warrant POST)
  - Waterhemp control and yield at harvest
  
- Some sites also collected soil samples...
  - more on that later...

# Planting Green Trial

## Waterhemp Control at Harvest\*

- 2021 – No factor was significant
  - 91 to 99 percent control across all management systems
- 2022 – Rye, PRE independently significant
- 14 DPP termination = Planting Green > No rye
  - 98 = 96 > 84 percent control
- PRE > No PRE
  - 98 > 88 percent control

\*POST of Enlist One (2 pt) + Liberty (2 pt) + Warrant (3 pt) @ 4" waterhemp



# Planting Green Trial – 2021

## Days from Planting to 4" Waterhemp

Rye management, PRE?, Planting Date	Days from Planting to POST
No rye, No PRE, May 19	26
No rye, PRE, May 19	36 (10 more days)
No rye, No PRE, June 1	23
No rye, PRE, June 1	29 (6 more days)
Term rye 14 DPP, No PRE, May 19	29
Term rye 14 DPP, PRE, May 19	42 (13 more days)
Term rye 14 DPP, No PRE, June 1	23
Term rye 14 DPP, PRE, June 1	36 (13 more days)
Plant green, No PRE, May 19	29
Plant green, PRE, May 19	42 (13 more days)
Plant green, No PRE, June 1	29
Plant green, PRE, June 1	36 (7 more days)

# Days from Planting to 4" Waterhemp

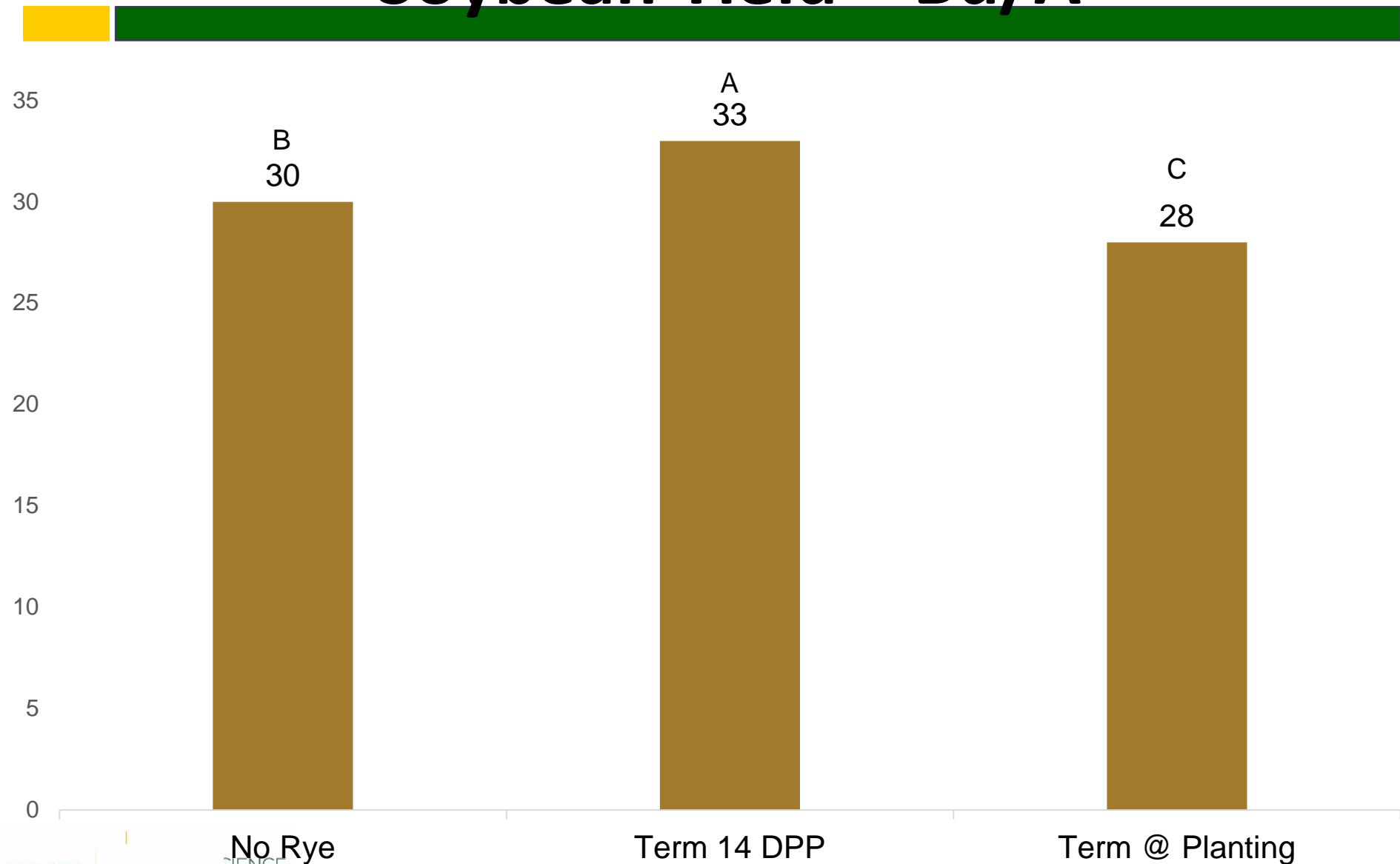
## Two Year Average



- No rye
  - 6.25 days more days with PRE
- Terminate rye 14 DPP
  - 14.5 days more days with PRE
- Plant green
  - 12.75 days more days with PRE

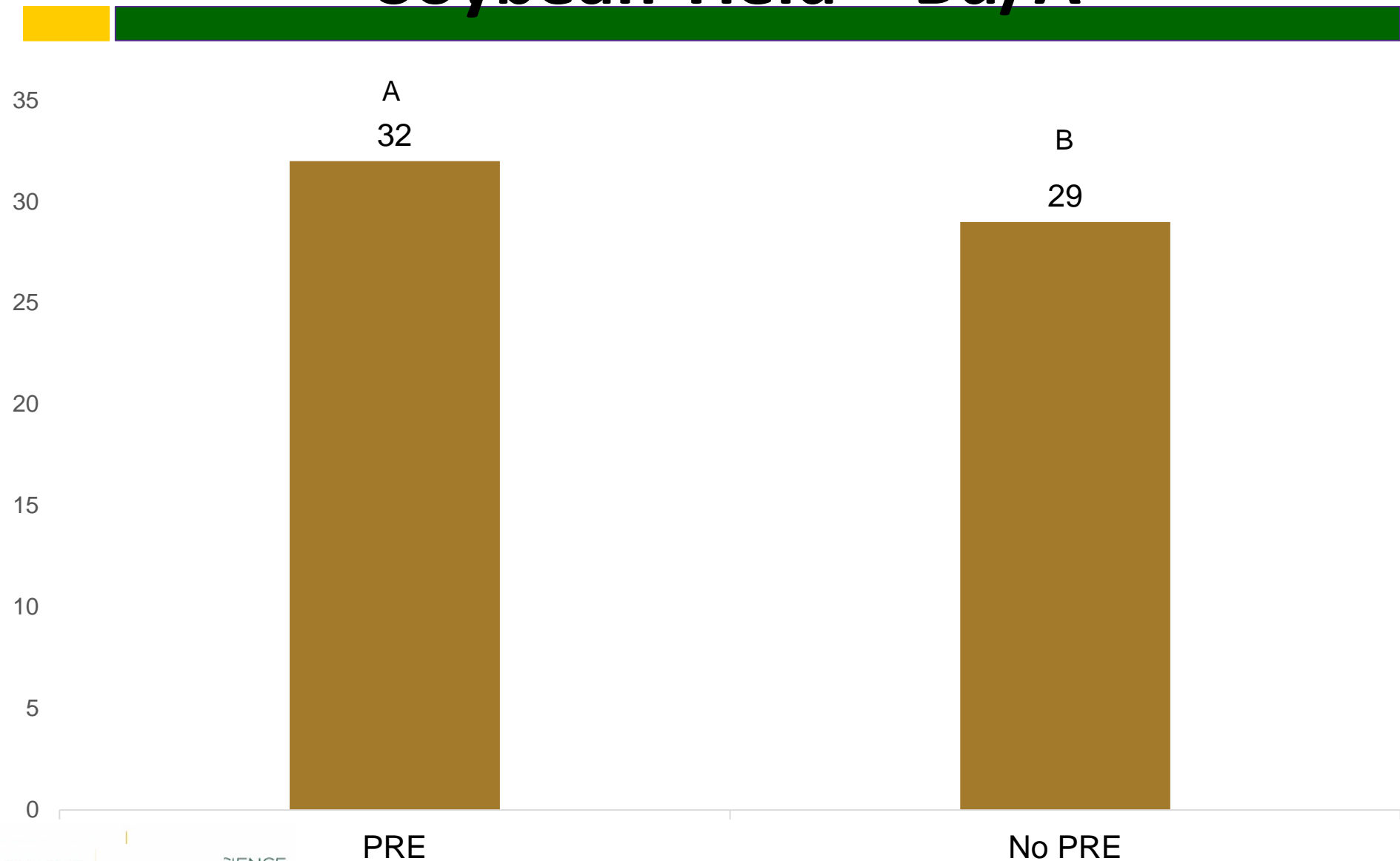
# Planting Green Trial – 2021

## Soybean Yield – Bu/A



# Planting Green Trial – 2021

## Soybean Yield – Bu/A



# Do cover crops 'tie up' herbicides?

- Plant mulch may bind 7 to 10% of metolachlor residue present after application
- No in-field data on other herbicides

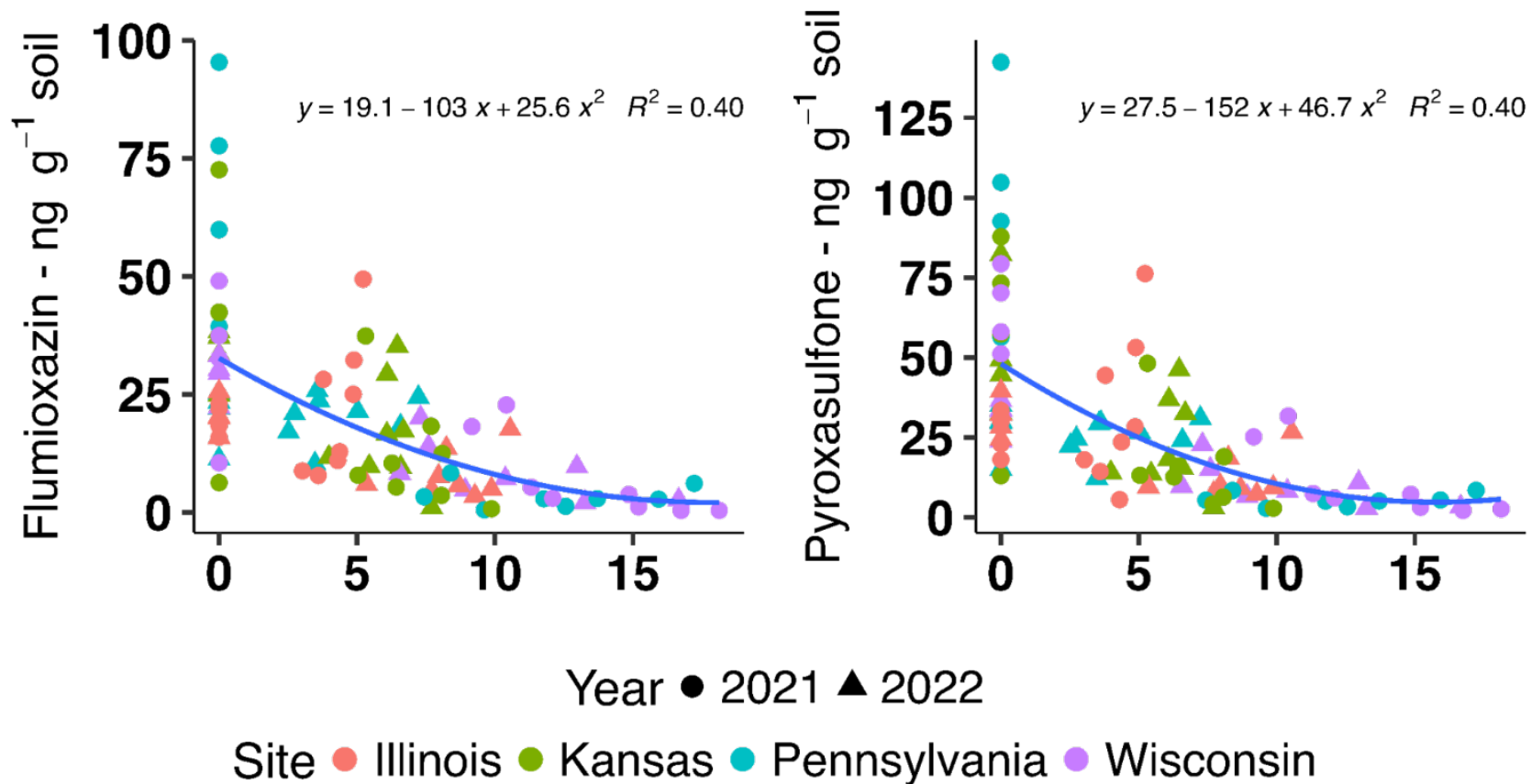


# How do cover crops affect herbicide fate?

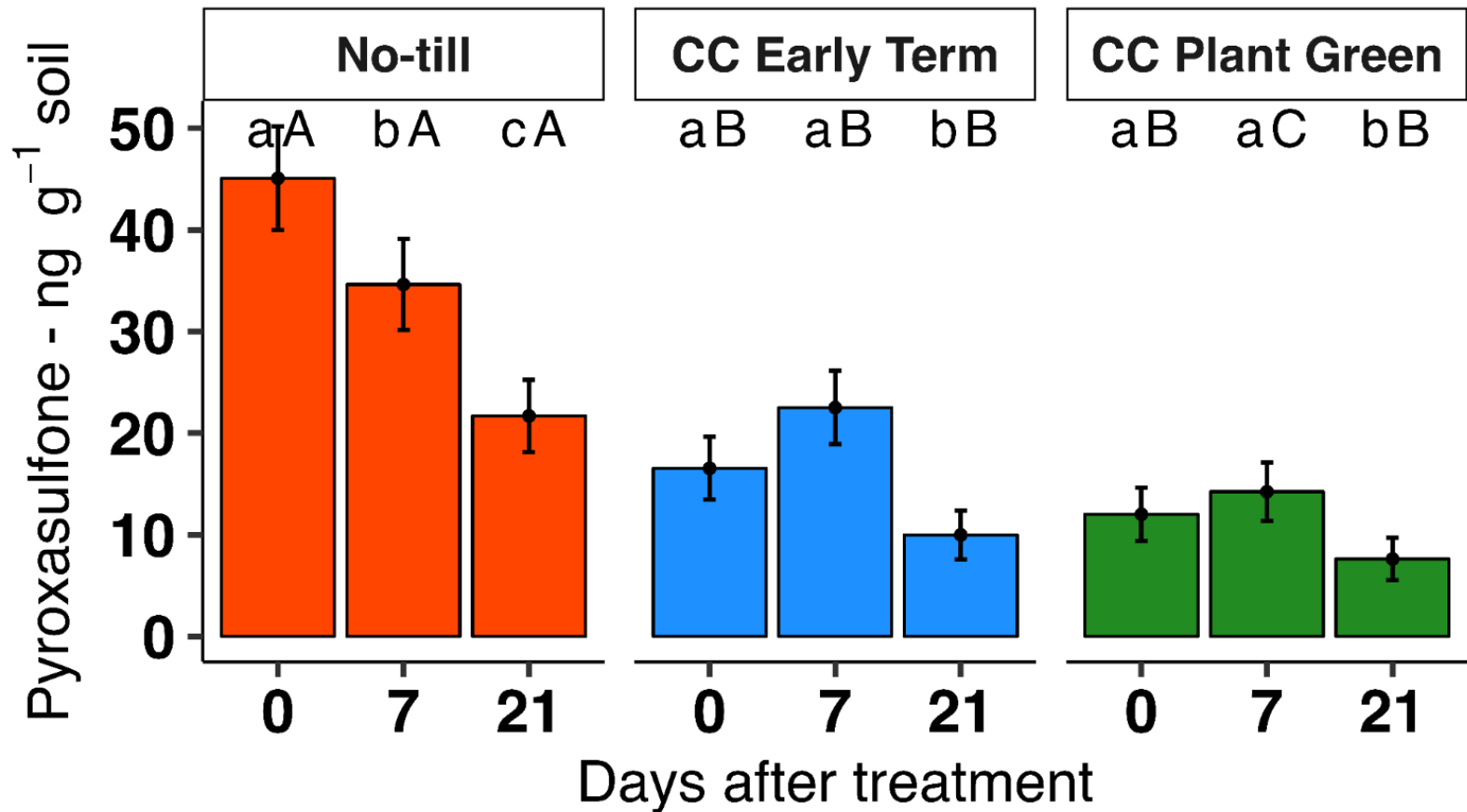
- Soil sampled from
  - no-till without PRE (check)
  - no-till with PRE (no-till)
  - CC terminated ~12 days before soybean planting with PRE (CC early term)
  - CC terminated at soybean planting with PRE (CC plant green)
- Sampled 0, 7, and 21 days after treatment



# Herbicide dissipation

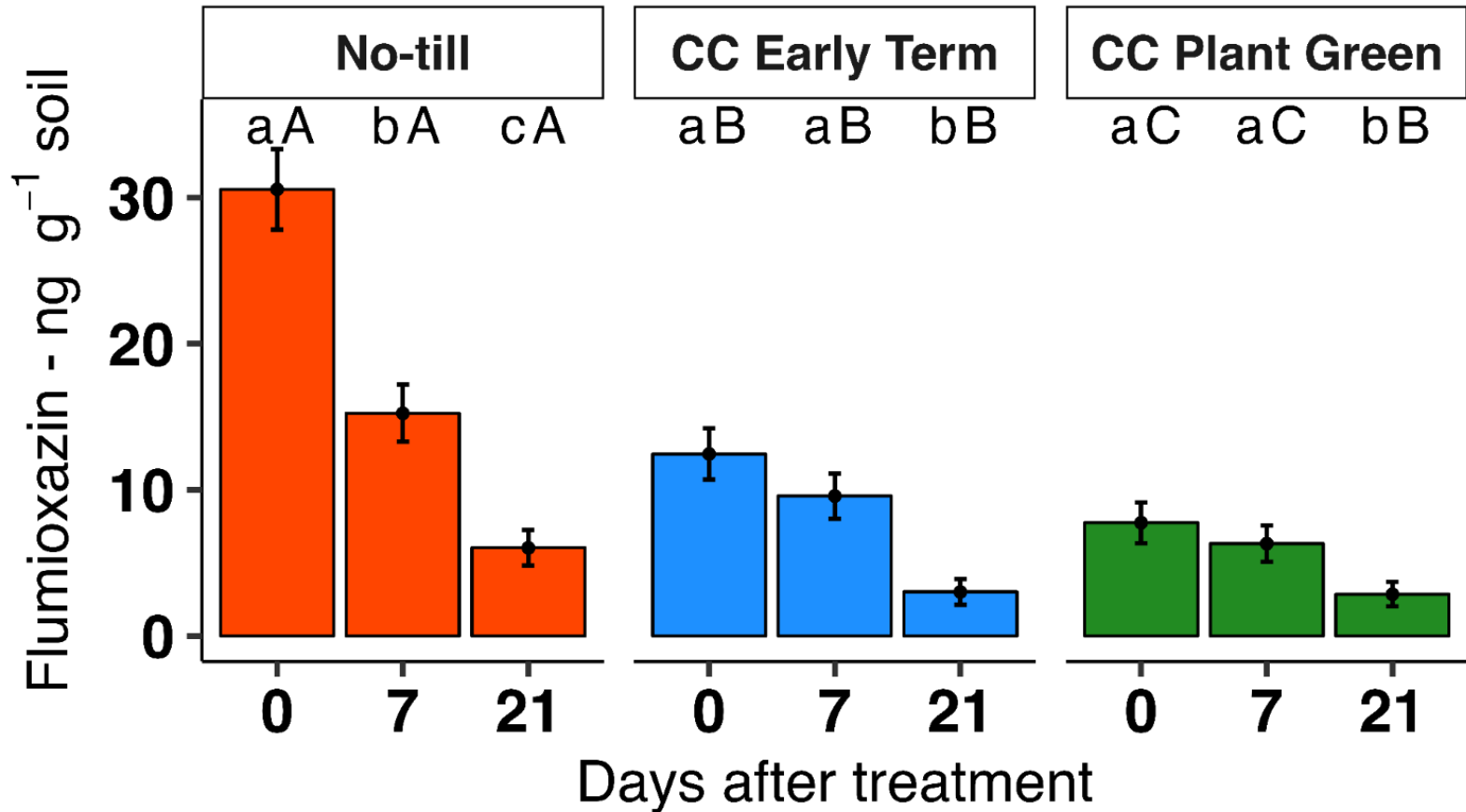


# Cereal rye reduced pyroxasulfone deposition





# Cereal rye reduced flumioxazin deposition



# Take aways

- CC biomass affected the fate of flumioxazin and pyroxasulfone applied PRE
  - Waterhemp control was not reduced
- Delaying CC termination until PRE greater impact on the fate of flumioxazin than pyroxasulfone.
  - Greater difference between no-till vs CC than early term vs plant green
  - Confounding factors include CC biomass accumulation, daily precipitation, temperature, and soil characteristics

# Do cover crops affect pigweed seeds?



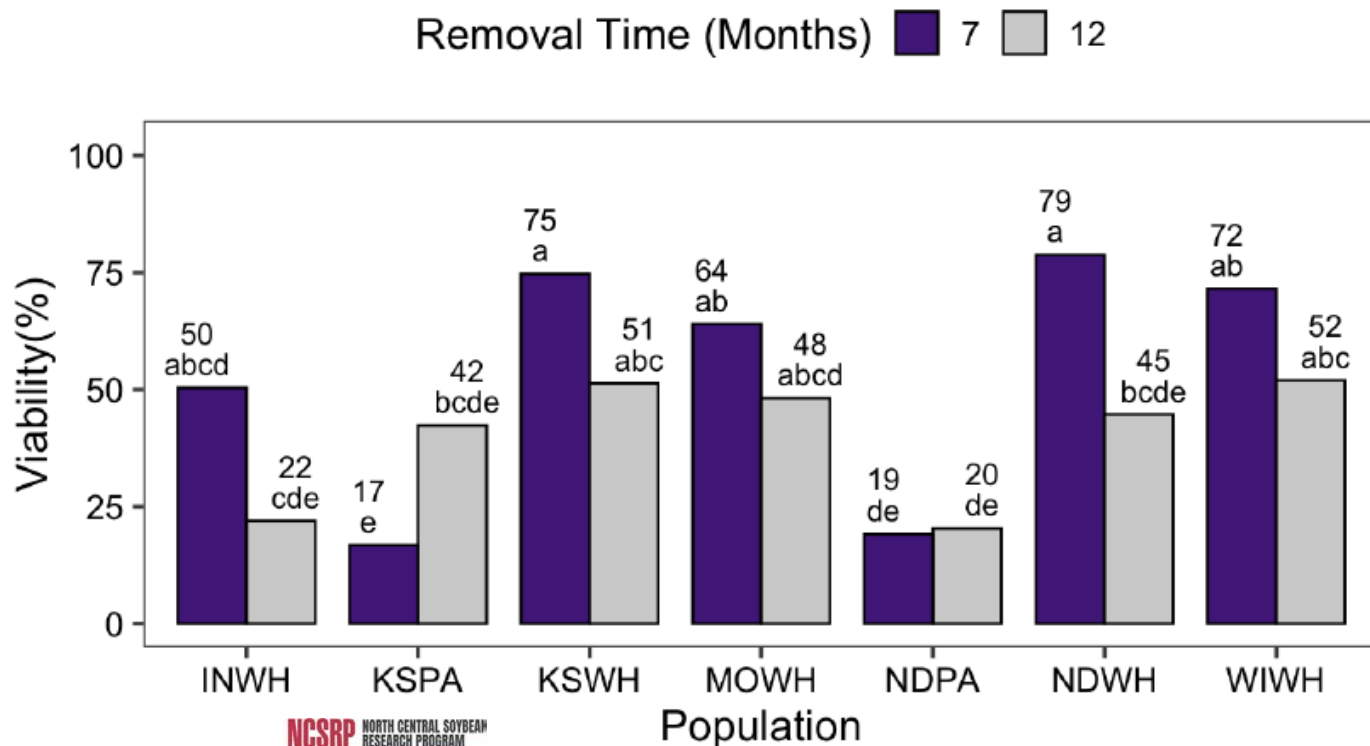
# Seed burial

- 50 seeds placed in fine (120 mm) wire mesh packets
- Packets were buried fall 2021
  - Each site:
    - Local population
    - KS waterhemp (KSWH)
    - KS Palmer amaranth (KSPA)
  - Rossville, KS included all populations



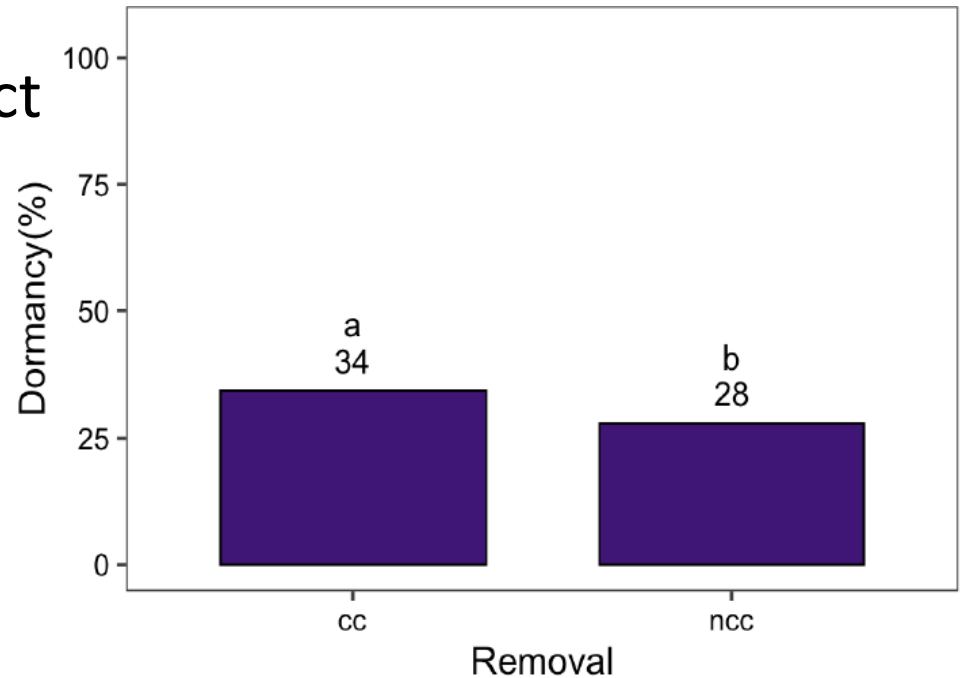
# All Populations at Kansas

- Only North Dakota waterhemp viability was less after twelve months compared to seven months



# KS Palmer amaranth and waterhemp

- Viability
  - No cover crop effect
- Dormancy
  - Greater in seeds buried in cover crops than in no cover crop treatments



Pooled across all locations

# Take aways

- Cereal rye changes the weed seedbank
  - Increased dormancy of KS pigweeds
    - This may mean a prolonged germination period
  - Further research is needed



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## **Related episodes:**

### **Integrated Weed Management**

**S1, E5 - Feb 19, 2021**

### **Cover Crops for Weed Management**

**S1, E8-9 - Mar 16 & 23, 2021**

### **Better Early than Later**

**S2, E5 - Oct 13, 2021**

### **Planting Green**

**S3, E6 - Feb 23, 2022**



# Weeds in Low pH Soil

- Buchanan et al (1975)
  - Redroot pigweed, wild mustard biomass reduced in low pH (4.7) compared to 6.3.
  - Crabgrass unaffected by soil pH
- Weaver and Hamill (1985)
  - Green foxtail biomass increased at pH 4.8 compared to 7.3
  - Powell amaranth biomass lower at pH 4.8 compared to 6.0 or 7.3
- Chauhan and Johnson (2009)
  - Junglerice germination not affected by pH (4 to 9)

# Weeds in Luc

(1975)  
 ed, wild mustard  
 3.  
 ected by soil pH

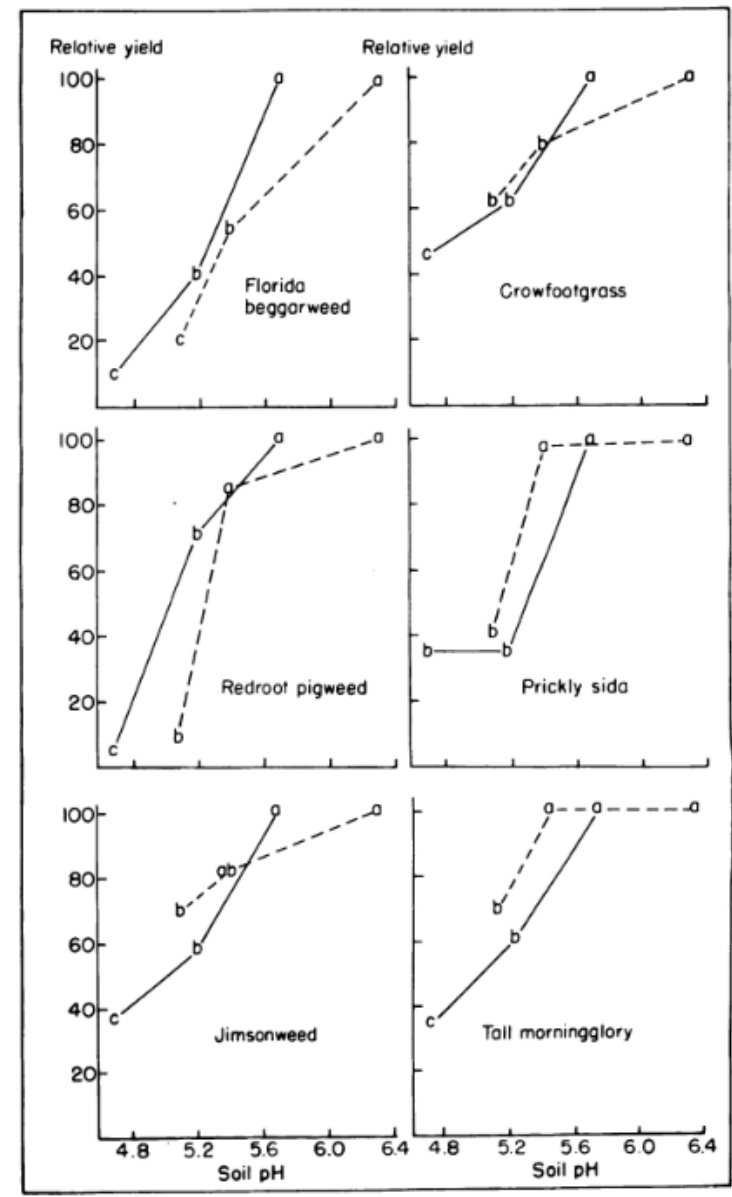
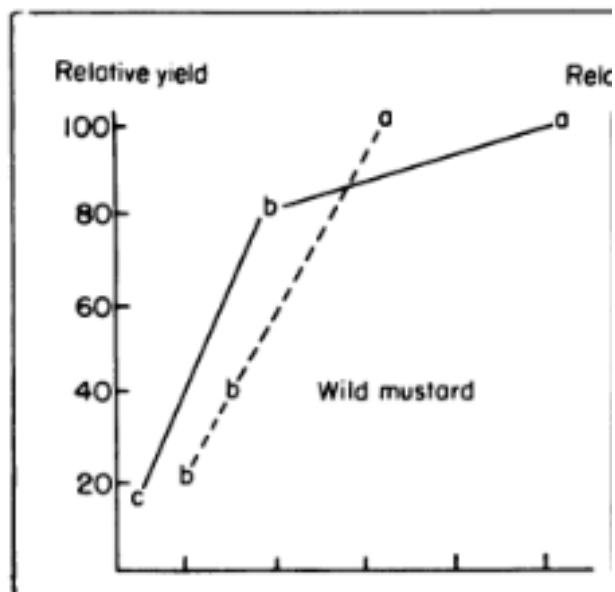
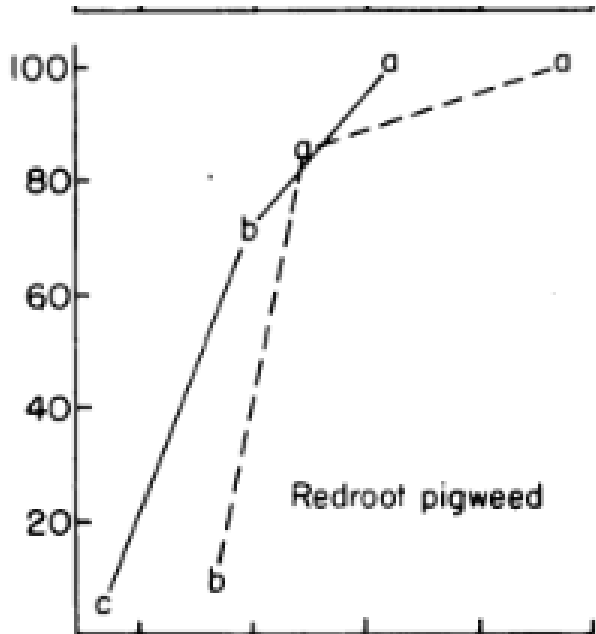


Figure 1. Relative growth of Florida beggarweed, crowfootgrass, redroot pigweed, jimsonweed, tall morningglory, and prickly sida in Lucedale sandy loam soil (solid line) and in Hartsells fine sandy loam soil (broken line) at different levels of soil pH. Any two means for a soil not marked with the same letter are significantly different at the 0.05 level of probability.

# Kochia in Low pH Soil?

## Everitt et al (1983)

- Kochia germination not affected by:
- Soil pH (2-11)
- Light
- Salts (NaCl, CaCl<sub>2</sub>, MgCl<sub>2</sub>, KCl, Na<sub>2</sub>SO<sub>4</sub>, MgSO<sub>4</sub>)
- Temperature (5 to 25 C)
  
- Evetts and Burnside (1972)
- No effect pH 2-8
  - 35 to 45% reduction at pH 10

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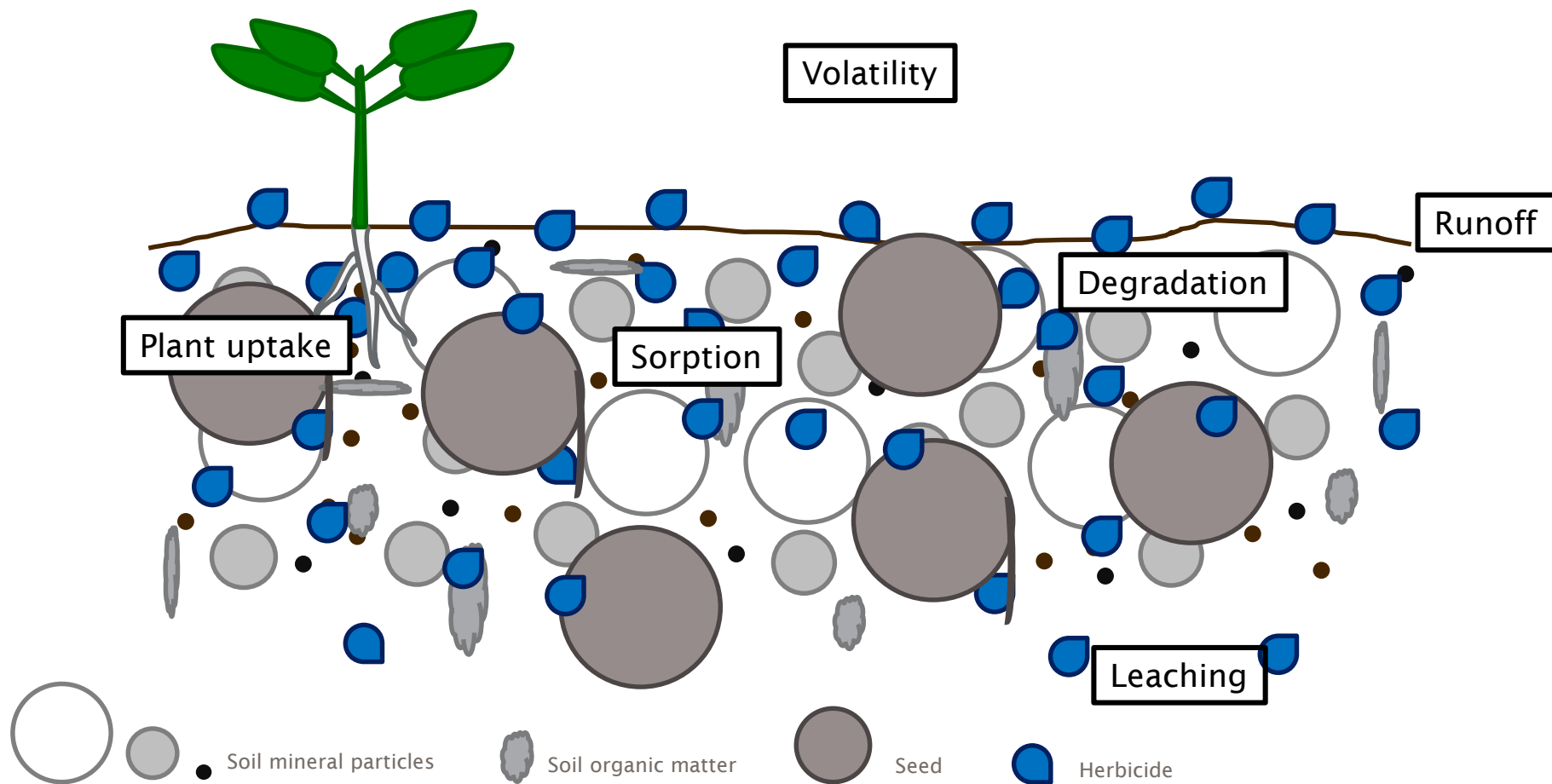
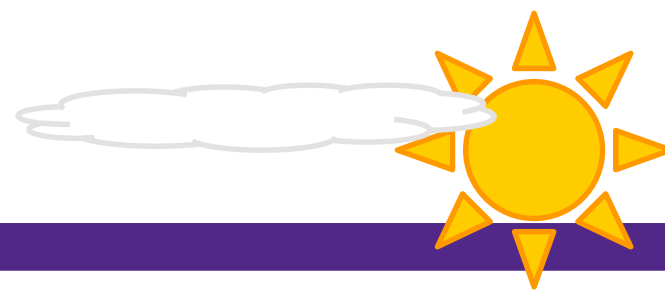


# General Rules for Herbicide Breakdown

(Pages 100-104 in ND Weed Guide)

1. Many herbicides are broken down in soil by microbial decomposition. In addition, **SUs and triazines are broken down by chemical reactions like acid hydrolysis.**
2. Herbicide molecules must be free from binding to soil particles or organic matter for soil microorganisms to degrade.
3. Most herbicide molecules are more tightly adsorbed to soil particles in dry soils than moist soils.
4. **Chemical degradation of herbicides in soil is affected by soil pH. Acid hydrolysis nearly ceases at soil pH above 6.8.**

# Herbicide fate



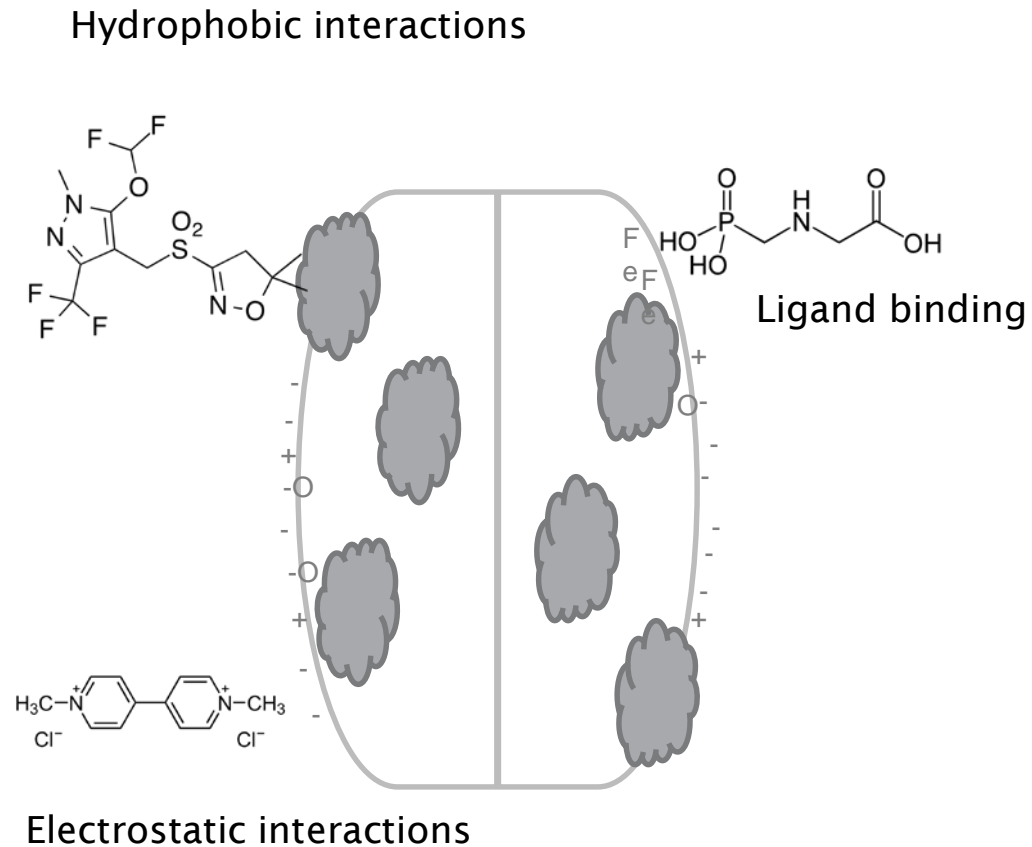
# Degradation



- Photodegradation
  - Chemical reactions caused by light
- Chemical
  - Chemical reactions not directly involving living organisms
    - Soil pH
- Microbial
  - Caused by algae, fungi, actinomycetes and bacteria

# Microbial degradation

- Rate of microbial degradation influenced by:
  - Previous applications
  - Microbial activity
  - Sorption





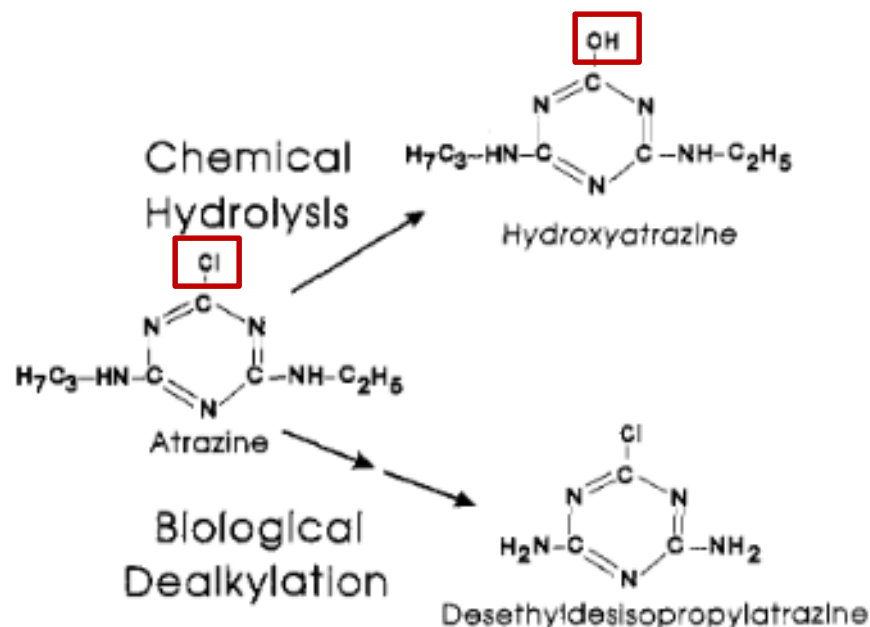
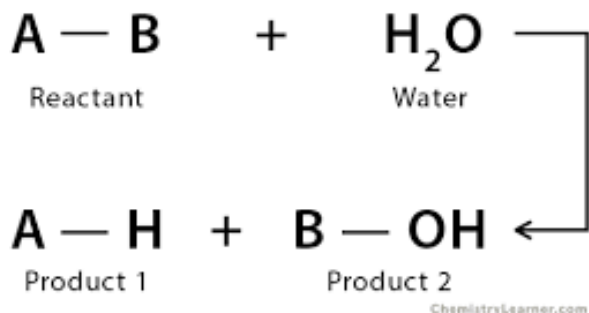
# Fate of some herbicides

Herbicide	Grp	Example	Water solubility g/mL	Koc mL/g	Dt50 days	Primary degradation
chlorsulfuron	2	Glean	40	36	36	hydrolysis
imazethapyr	2	Pursuit	60-90	52	51	microbial
atrazine	5	Aatrex	60	100	29	microbial, acid hydrolysis
metribuzin	5	Dimetric	30-60	48	19	microbial
fomesafen	14	Reflex	100	100	86	photodegradation, anaerobic microbes
sulfentrazone	14	Authority	120-300 (increases with pH)	43	500	microbial
mesotrione	27	Callisto	15-21	100	5	microbial

# Low Soil pH and Atrazine/SU

- 3. Broken down primarily by acid hydrolysis. Microbial degradation is very slow.
- 4. Non-microbial hydrolysis for most residual SU herbicides ceases at soil pH above 6.8.

## Hydrolysis



# Oat Biomass After Atrazine

## Hiltbold and Buchanan 1977

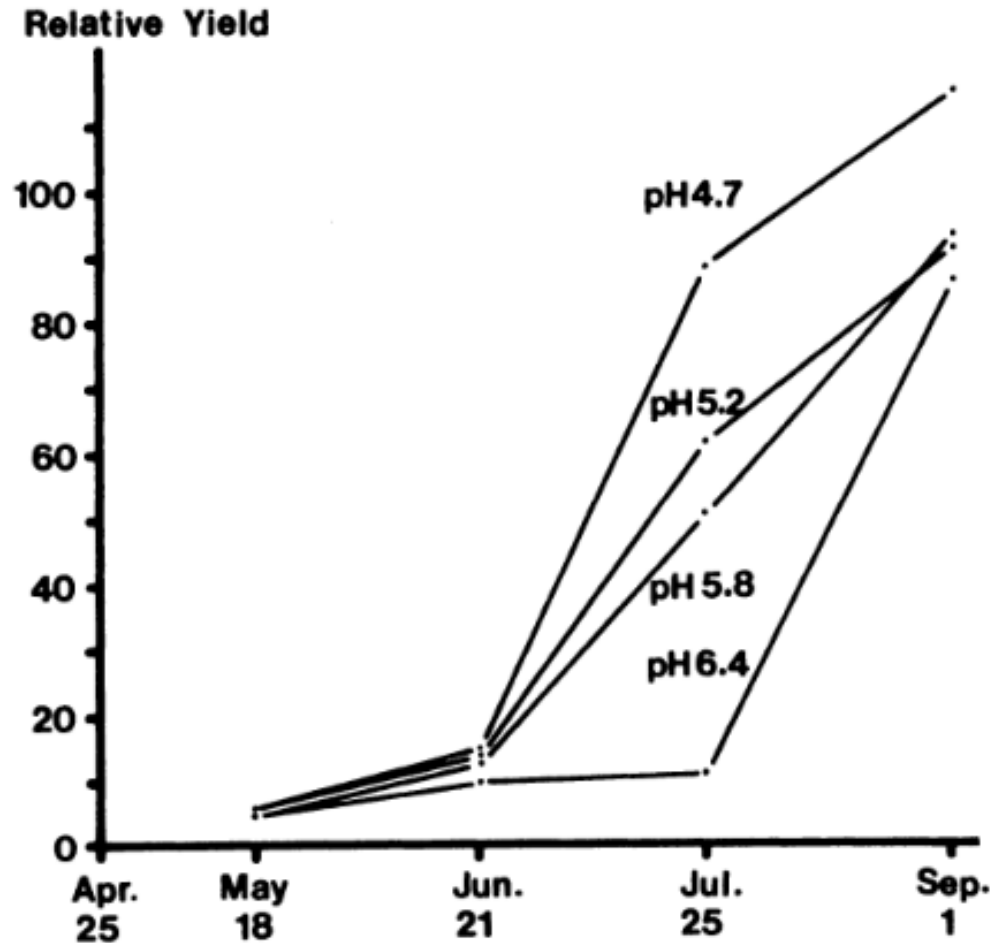
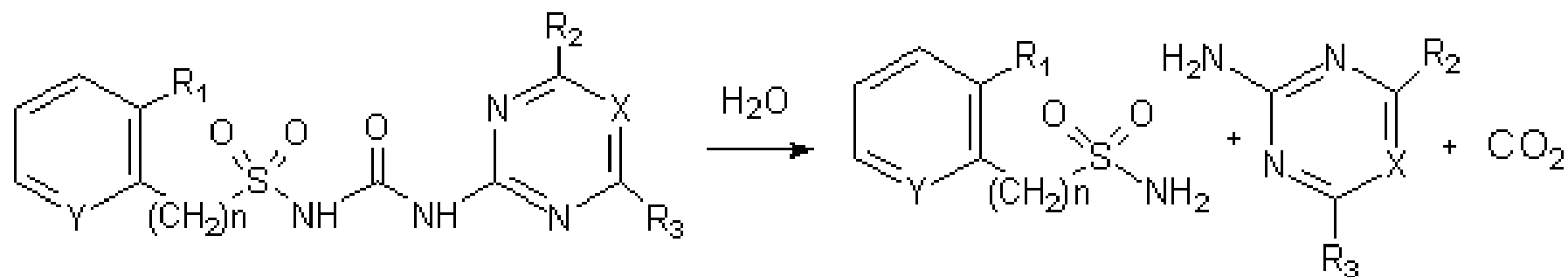


Figure 1. Relative yield of oats (% of control) in soil from four of the total 20 plots of Hartsells fsl soil ranging in pH from 4.7 to 6.4 after application of 2.24 kg/ha of atrazine on April 25, 1972.

# SU Hydrolysis



**Cleavage 1**

# Chlorsulfuron in Low pH

## Fredrickson and Shea (1986)

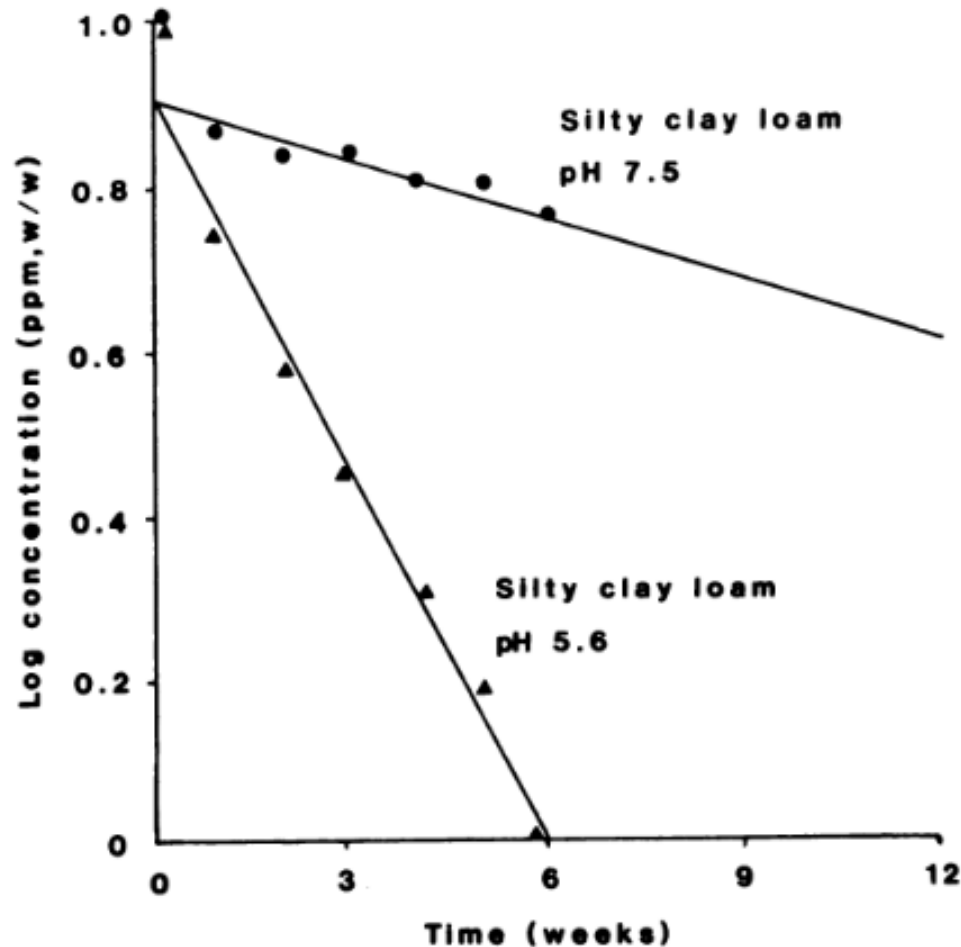
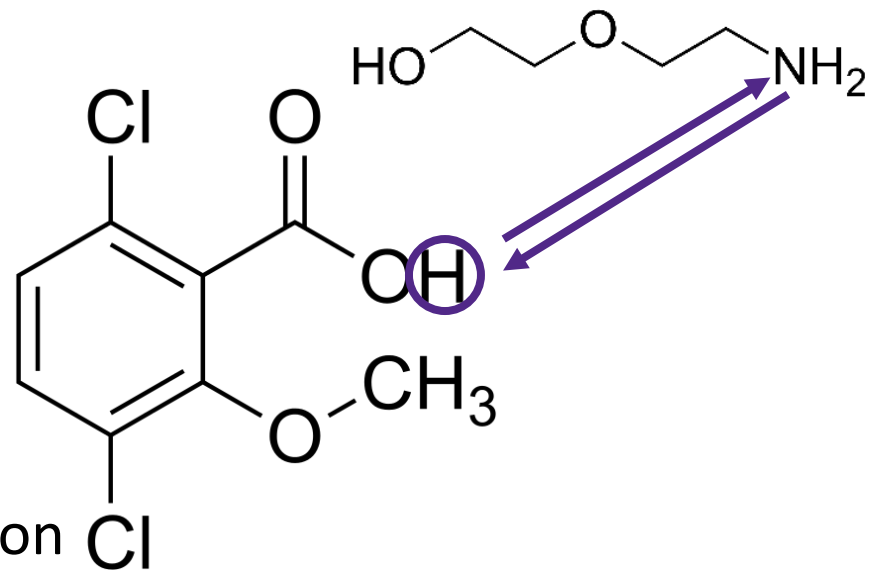


Figure 2. Log plot of chlorsulfuron degradation in a silty clay loam at pH 5.6 and adjusted to pH 7.5. Regression equations are  $\hat{Y} = -0.16X + 0.94$  for pH 5.6 ( $r^2 = 0.99$ ), and  $\hat{Y} = -0.03X + 0.95$  for pH 7.5 ( $r^2 = 0.87$ ).

# Why pH matters

- Weak acid herbicides associate/dissociate as function of pH
  - pKa varies for each molecule
    - pH 5 for dicamba

- This influences
  - Absorption
  - Translocation
  - Binding at active site
  - Behavior in spray solution
  - Volatility



# Low Soil pH and Herbicides

- When soil pH is higher than pKa of weak acid herbicides:
  - Herbicides exist in anionic form (negative charge)
  - Repelled by soil (negative charge)
  - More available in soil solution
  - Ex – Sulfentrazone pKa = 6.56

# Low Soil pH and Herbicides

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  - Herbicides exist in anionic form (negative charge)
  - Repelled by soil (negative charge)
  - More available
  - Ex – Sulfenyl

SPARTAN CHARGE Herbicide Use Rate Table (Safflower) Post Plant Pre-emergence Applications					
Broadcast Rate	SPARTAN CHARGE Herbicide oz/A (lb ai/A)				
	Soil Texture				
% Organic Matter	Coarse	Medium pH 7.0-7.5	Medium pH lower than 7.0	Fine pH 7.0-7.5	Fine pH lower than 7.0
1.5 - 3.0 %	Do not use	2.5 (0.07)	3.0 (0.08)	3.5 (0.09)	4.0 (0.11)
3.0+ %	Do not use	3.5 (0.09)	4.0 (0.11)	4.5 (0.12)	5.0 (0.14)

Refer to the SPARTAN CHARGE herbicide section 3 label for information on soil types under the COARSE, MEDIUM, and FINE categories.



# Low Soil pH and Herbicides

- Availability in soil solution necessary for herbicide degradation AND plant uptake

## Sulfentrazone

- MORE available (less adsorbed) at low pH

## Atrazine

- Less persistent at low pH

## Imidazilinones

- More persistent at low pH
- Increased adsorption at lower pH (<6)
- More herbicide in soil solution at high pH

## Sulfonylureas

- Less persistent at low pH



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## Related episodes:

**Bioassay and Herbicide Degradation**

**S2, E2 - Sept 22, 2021**

**You've Got to Read the Labels**

**S3, E11 - Mar 30, 2022**

**Fall Applied Herbicides & Soil pH**

**S4, E4 - Sept 28, 2022**

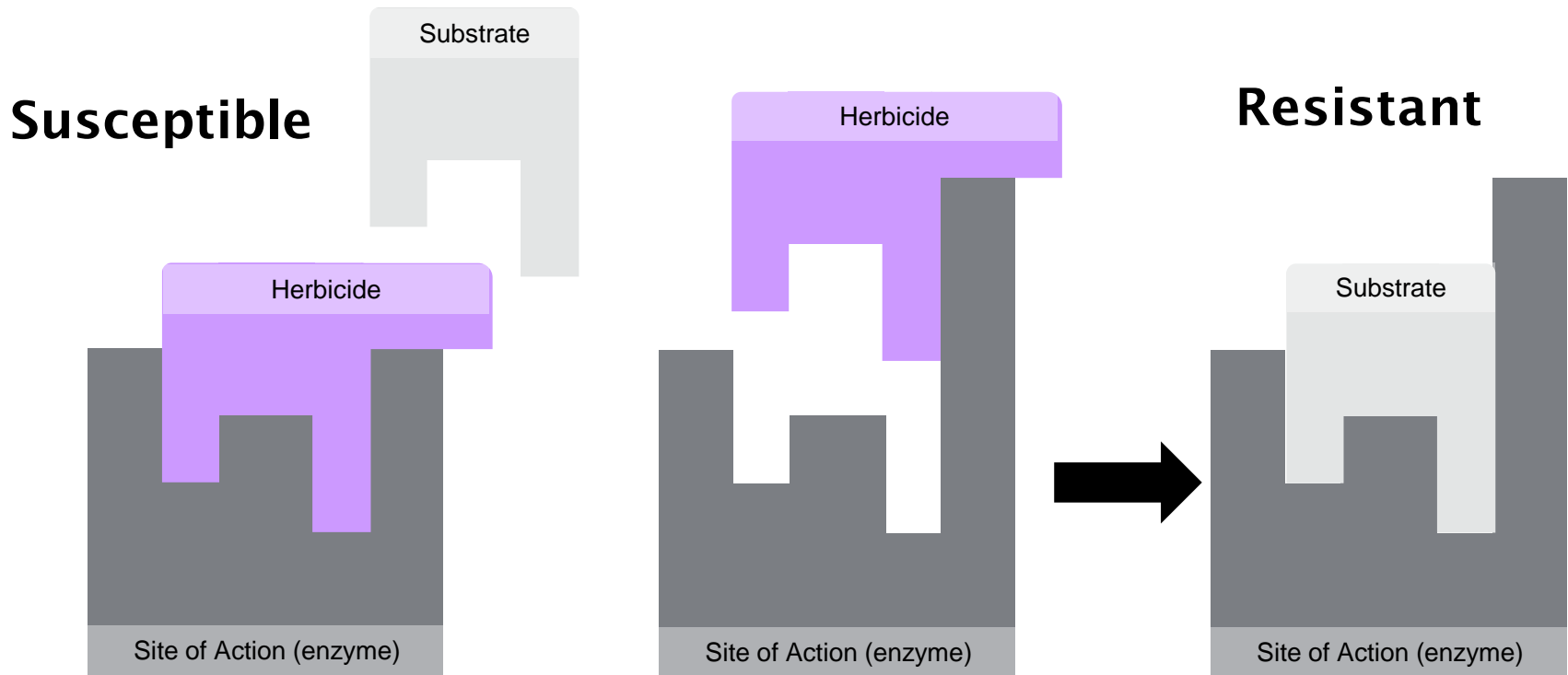
# Causes of resistance



## Target-site mutation

- One gene
- Develops faster

# Altered Site of Action



# Causes of resistance



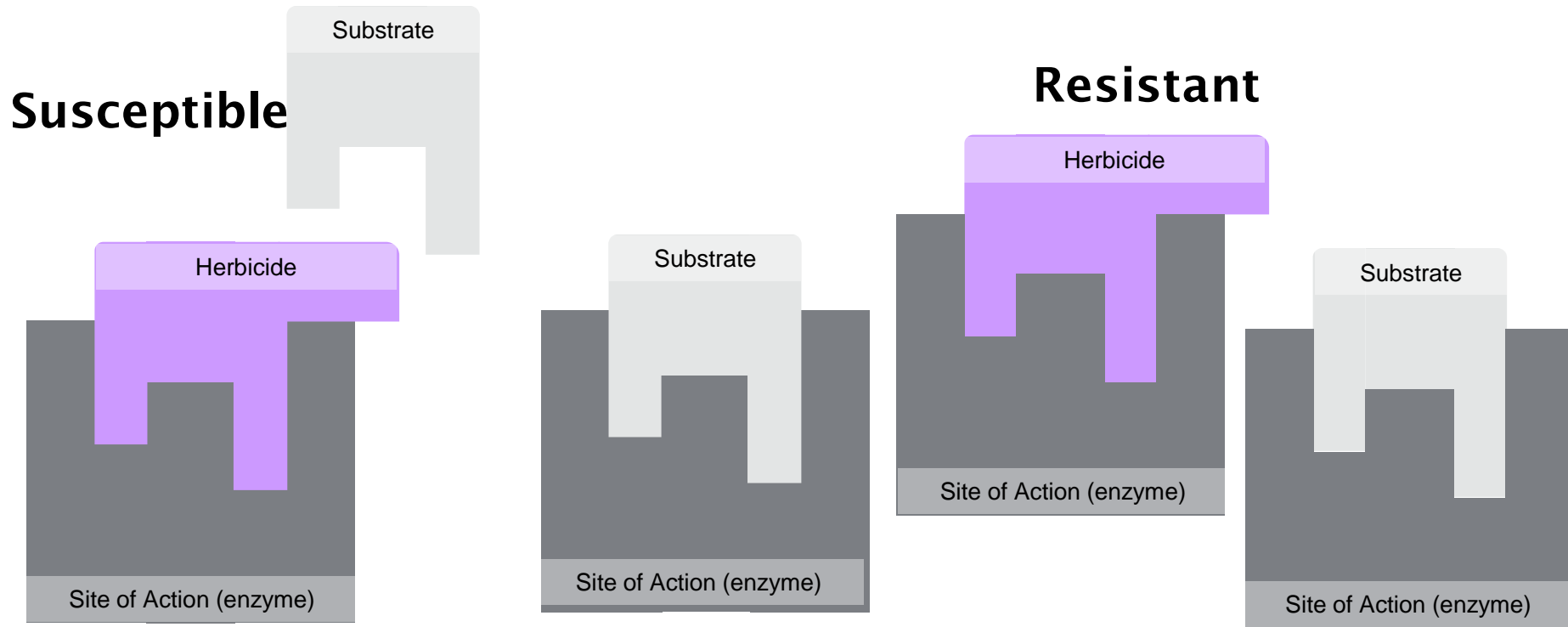
## Target-site mutation

- One gene
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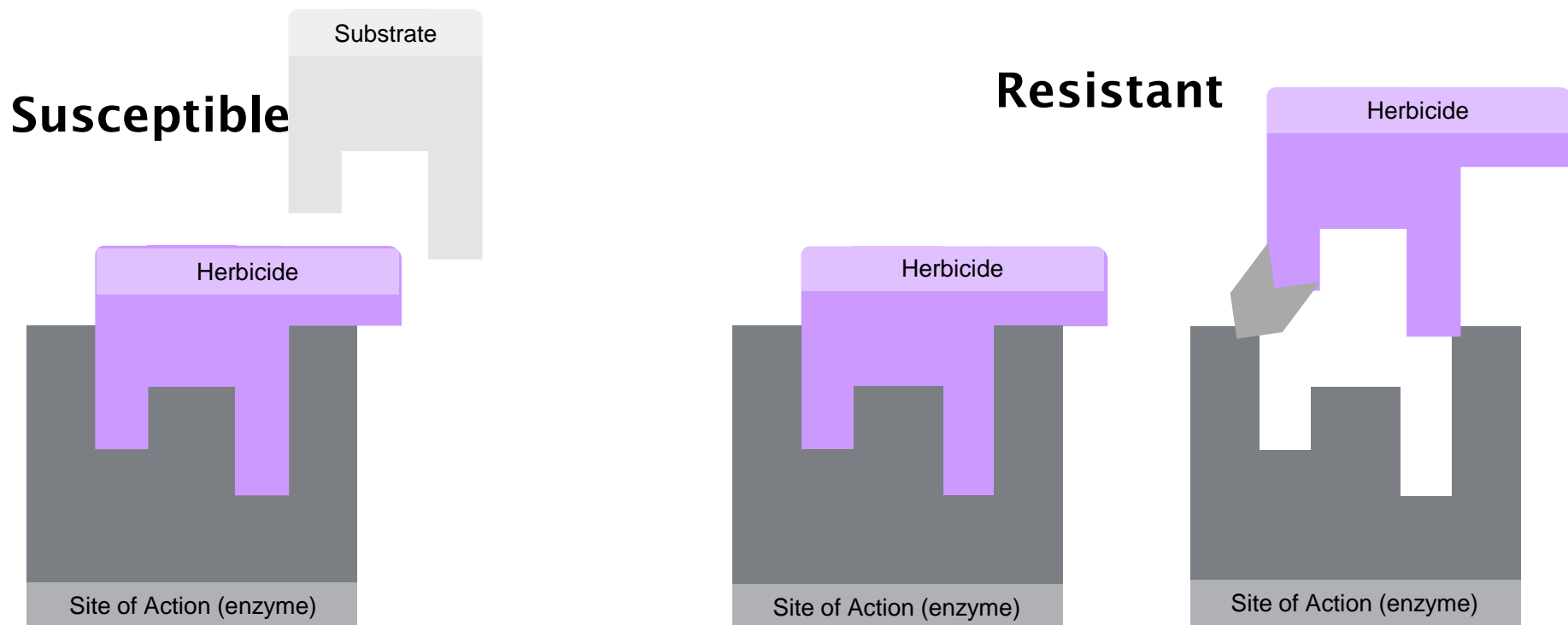
## Nontarget-site mutation

- > 1 gene
- Develops slower
  - Begins with low degree of resistance
  - Cross resistance more likely

# Increased Protein Expression

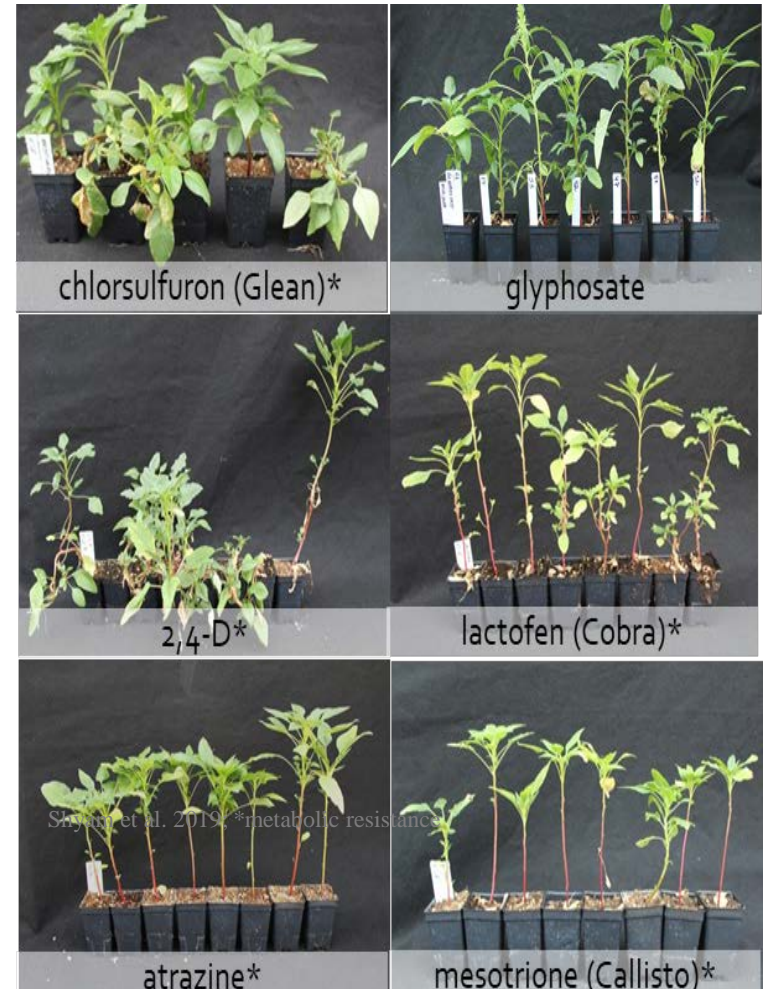


# Enhanced herbicide metabolism



# Metabolic resistance

- Herbicide converted to inactive forms before plant is killed
  - Cytochrome P450s and glutathione S-transferases
  - We must rethink assumptions regarding herbicide resistance
    - A single resistance mechanism can cause resistance to multiple herbicide groups, reduces effectiveness of mixing and rotating herbicides

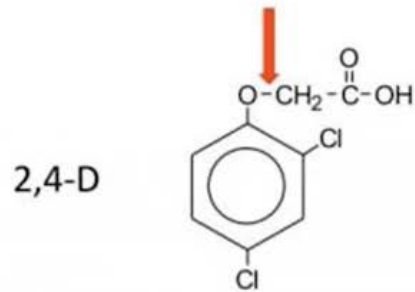




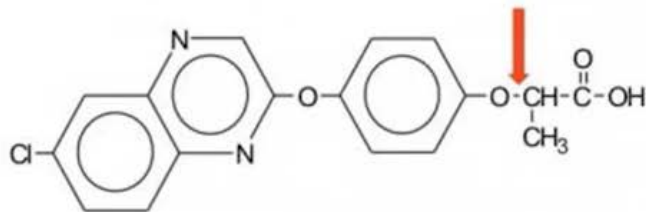
# Metabolic resistance webinar

Pat Tranel – University of Illinois

## Commercial example of cross-resistance



Enlist  
2,4-D and “fop” resistance



quizalofop

# Corn EPOST – 2022

Herbicide (Rate/A) POST @ V2 corn (~14 days after planting)	Palmer Control 28 days after POST
Acuron GT (3.75 pt)	<b>35 C</b>
Acuron GT + Aatrex (3.75 pt + 1 pt)	<b>75 A</b>
Resicore + Powermax (1.25 qt + 26.6 fl oz)	<b>50 BC</b>
Resicore + Aatrex + Powermax (1.25 qt + 1 pt + 26.6 fl oz)	<b>75 A</b>
Realm Q + Aatrex + Durango (4 oz + 1 pt + 24 fl oz)	<b>40 C</b>
Capreno + Harness + Powermax + Aatrex (3 fl oz + 2 pt + 1 qt + 1 pt)	<b>88 A</b>
Anthem Maxx + Callisto + Aatrex + Weathermax ( 4 fl oz + 3 fl oz + 1 pt + 22 fl oz)	<b>78 A</b>
Harness + Impact + Aatrex (1.75 pt + 1 fl oz + 1 pt)	<b>80 A</b>
Harness + Sinate + Aatrex (1.75 pt + 28 fl oz + 1 pt)	<b>89 A</b>
Armezon PRO + Aatrex + Powermax (18 fl oz + 1 pt + 1 qt)	<b>70 AB</b>
Status + Outlook + Aatrex + Powermax (5 oz + 1 pt + 1 pt + 1 qt)	<b>81 A</b>





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## **Related episodes:**

**Herbicide Resistance Update**

**S1, E4 - Feb 2, 2021**

**WSSA Winners from Illinois**

**S3, E7 - Mar 2, 2022**

**Herbicide Resistance Update**

**S3, E13 - Apr 13, 2022**

**Herbicide Resistant Ryegrass**

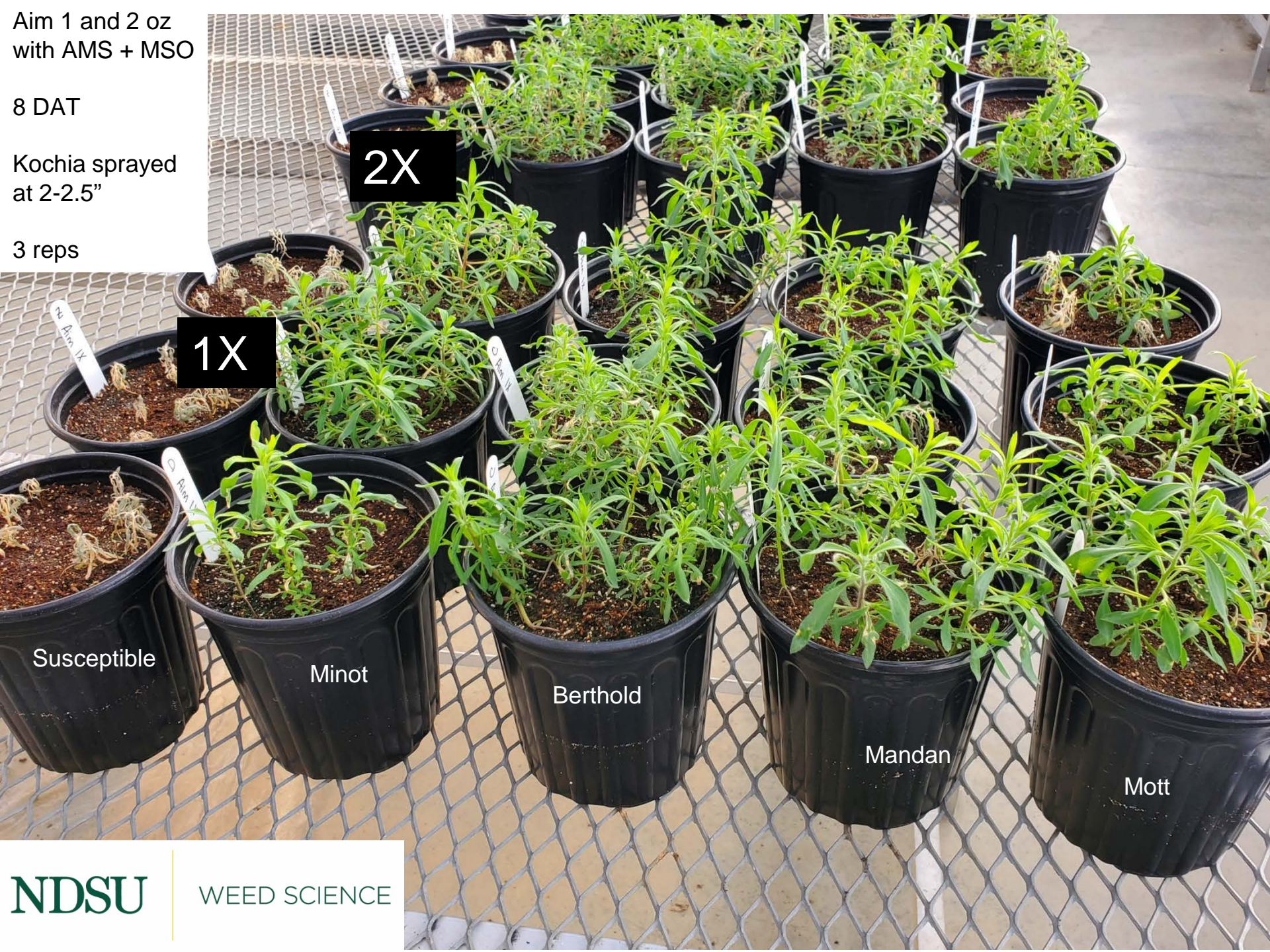
**S5, E2 - Jan 25, 2023**

Aim 1 and 2 oz  
with AMS + MSO

8 DAT

Kochia sprayed  
at 2-2.5"

3 reps



2X

1X

Susceptible

Minot

Berthold

Mandan

Mott

Aim 1 and 2 oz  
with AMS + MSO

13 DAT

Kochia sprayed  
at 2-2.5"

3 reps



## Kochia control with Aim 1 oz to 16 oz



Photo: 6 DAT

# PPO-inhibitor (Group 14) Resistance



## Review

Received: 21 July 2017

Revised: 5 September 2017

Accepted article published: 1 October 2017

Published online in Wiley Online Library: 22 November 2017

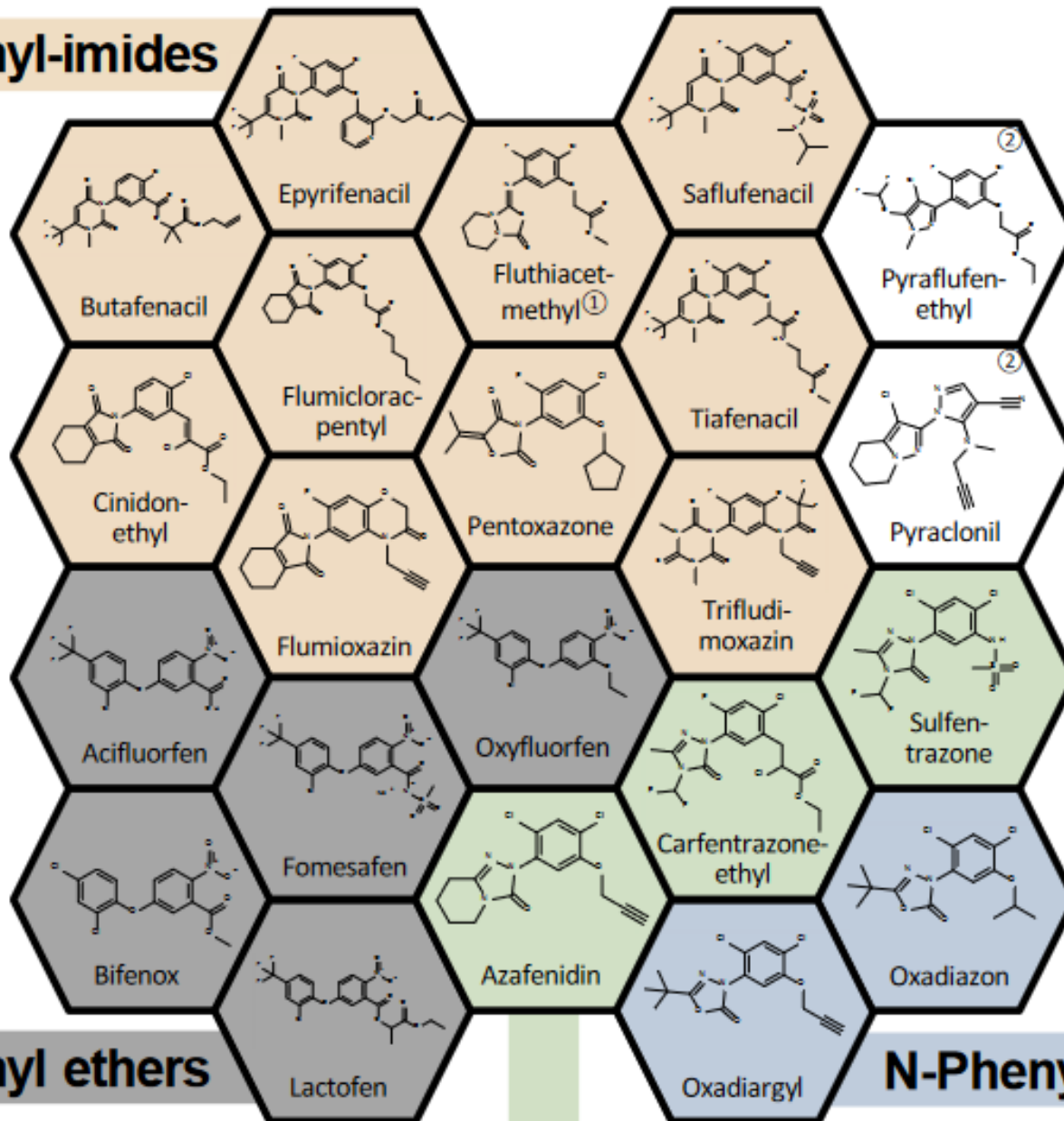
(wileyonlinelibrary.com) DOI 10.1002/ps.4744

## Origins and structure of chloroplastic and mitochondrial plant protoporphyrinogen oxidases: implications for the evolution of herbicide resistance

Franck E Dayan,<sup>a\*</sup>  Abigail Barker<sup>a</sup>  and Patrick J Tranel<sup>b</sup> 



## N-Phenyl-imides

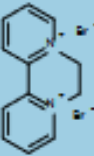


## Diphenyl ethers

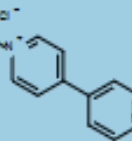
## N-Phenyl-triazolinones

## N-Phenyl-oxadiazolones

22



Diquat



Paraquat

# PPO-resistant waterhemp - POST

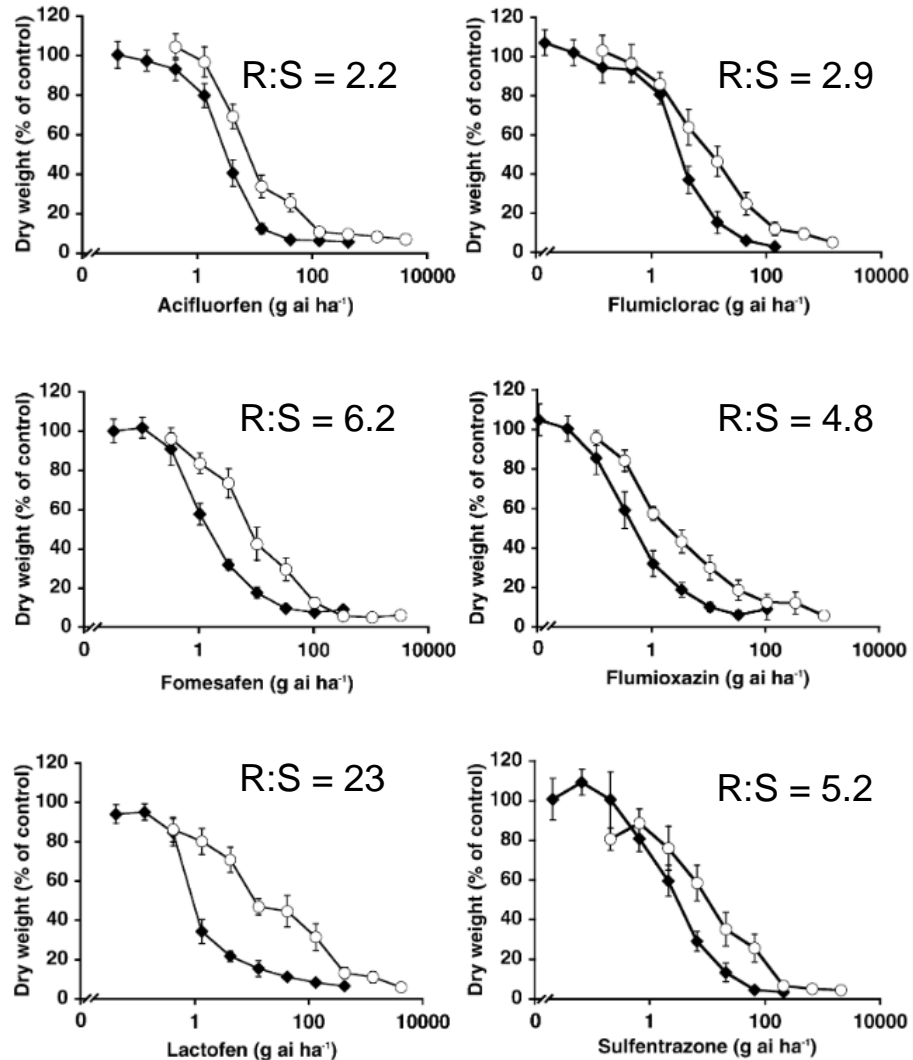


FIGURE 1. Protoporphyrinogen oxidase inhibitor dose-response curves of the Adams County herbicide-resistant (ACR = ○) and Wayne County herbicide-susceptible (WCS = ◆) waterhemp populations after postemergence applications of acifluorfen, flumiclorac, flumioxazin, fomesafen, lactofen, or sulfentrazone. Vertical bars represent ± SEM (n = 12).

# PPO-resistant waterhemp - PRE

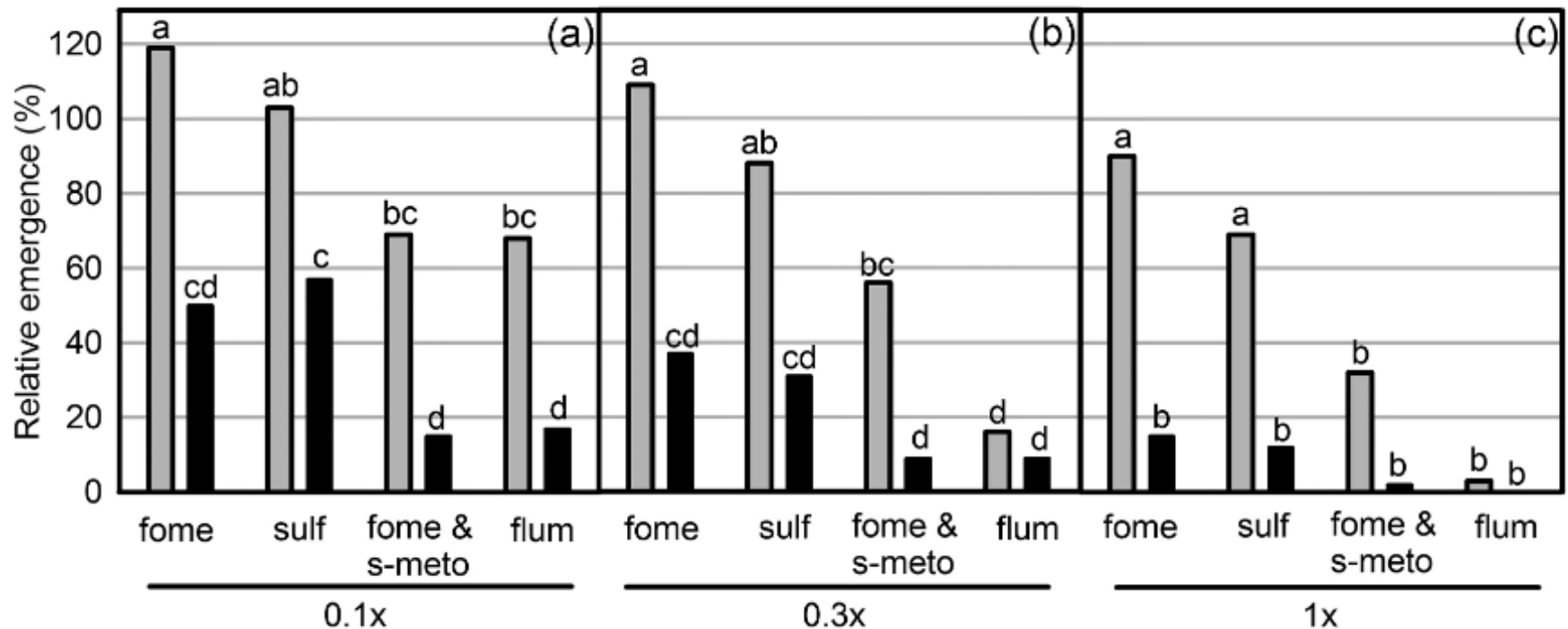
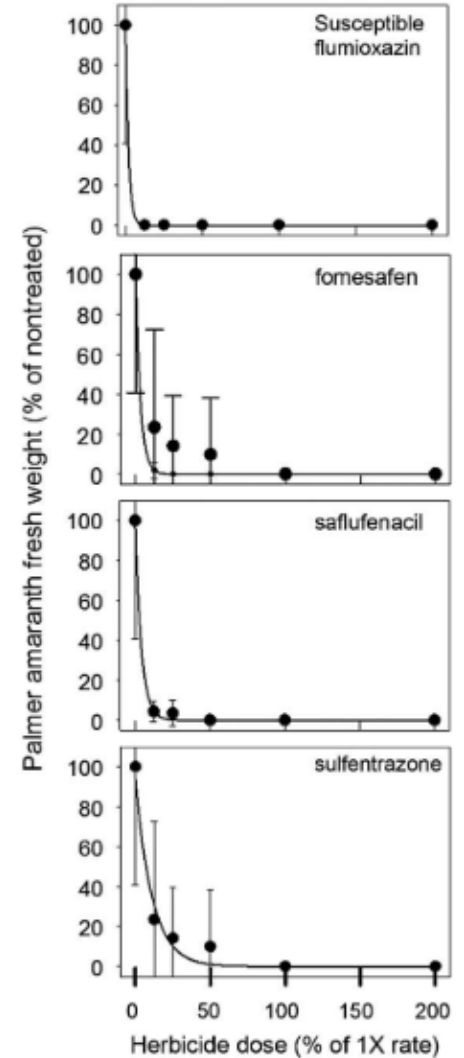
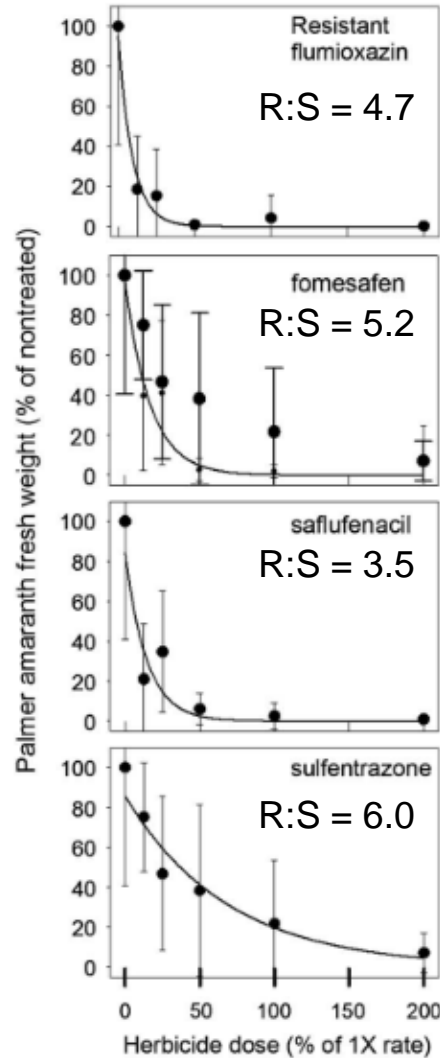


Figure 1. Relative cumulative waterhemp emergence of protoporphyrinogen oxidase inhibitor-resistant (PPO-R) (grey) and susceptible (PPO-S) (black) biotypes 10 d after treatment from four soil-residual herbicides applied at (a) 0.1×, (b) 0.3×, and (c) 1× labeled rates. The 1× labeled rates were as follows: fomesafen at 420 g ai ha<sup>-1</sup>, sulfentrazone at 280 g ai ha<sup>-1</sup>, flumioxazin at 107 g ai ha<sup>-1</sup>, and fomesafen at 420 g ai ha<sup>-1</sup> + s-metolachlor at 1,910 g ai ha<sup>-1</sup>. Means were separated using Tukey's honestly significant difference ( $\alpha = 0.05$ ). Abbreviations: fome, fomesafen; sulf, sulfentrazone; s-meto, s-metolachlor; and flum, flumioxazin.

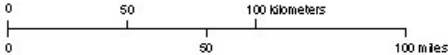
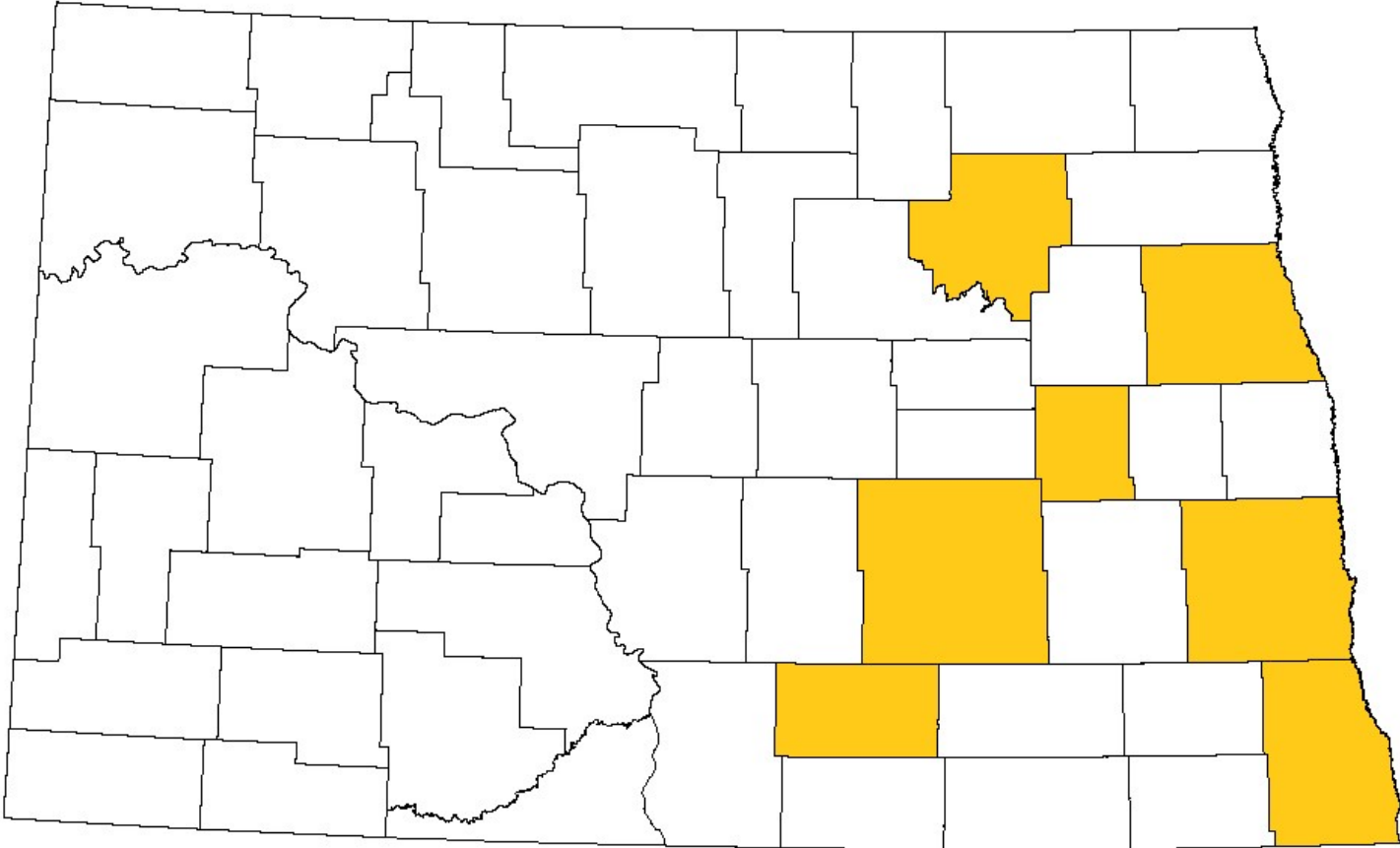
# PPO-resistant Palmer amaranth (PRE)



# Group 14-Resistant Waterhemp



NORTH DAKOTA



# PPO-Inhibitor Resistance

## *Mechanisms of Resistance*

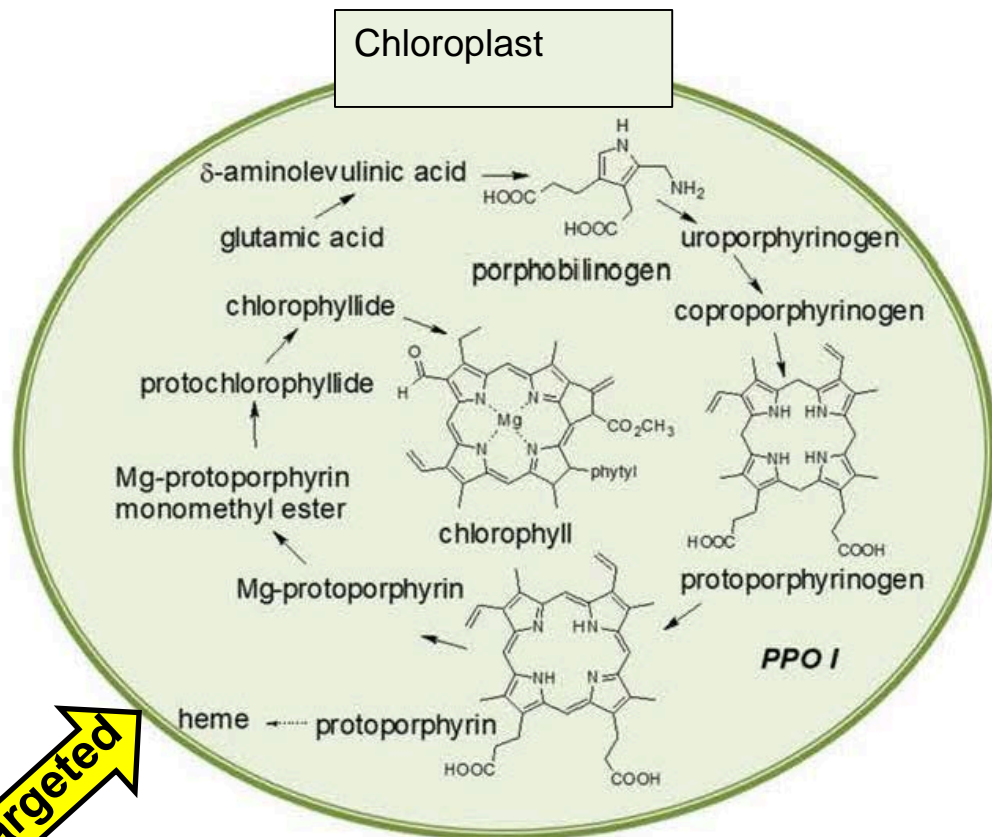
### Target Site Resistance: *PPX2* gene (PPO2 isozyme)

- Deletion of glycine residue at 210th position
  - $\Delta$ Gly210
  - 50% increase in PPO2 active site “pocket”
  - 100- to 500-fold reduction in sensitivity to diphenylether herbicides
- Substitution at Arg98 position
  - Arg98Gly, Arg98Met, or Arg98Leu

# PPO Inhibitor

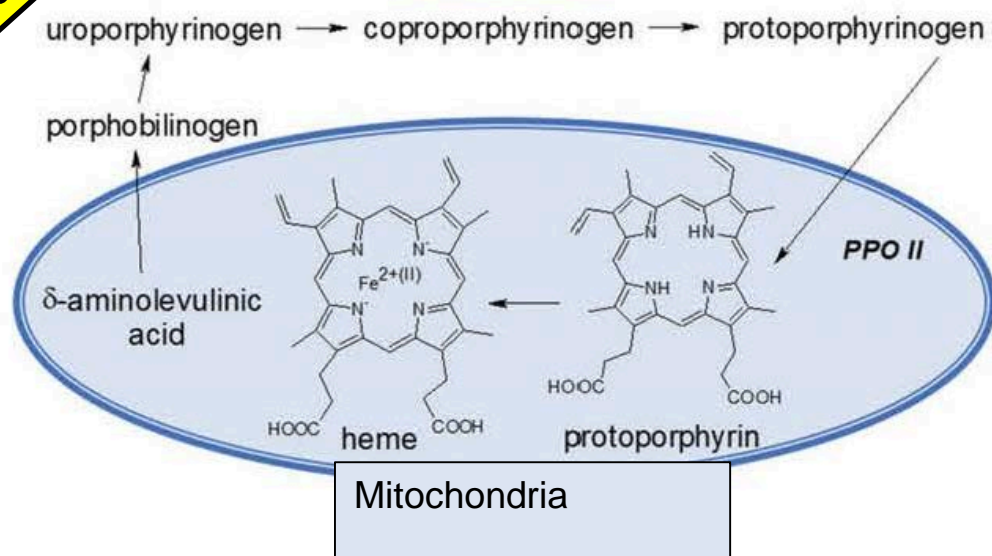
## Mechanism of Action

PPXI (PPX1) gene encodes PPO I (PPO1) isoform in the chloroplast



Can be dual targeted

PPXII (PPX2) gene encodes PPO II (PPO2) isoform in the mitochondria



# PPO-inhibitor (Group 14) Resistance

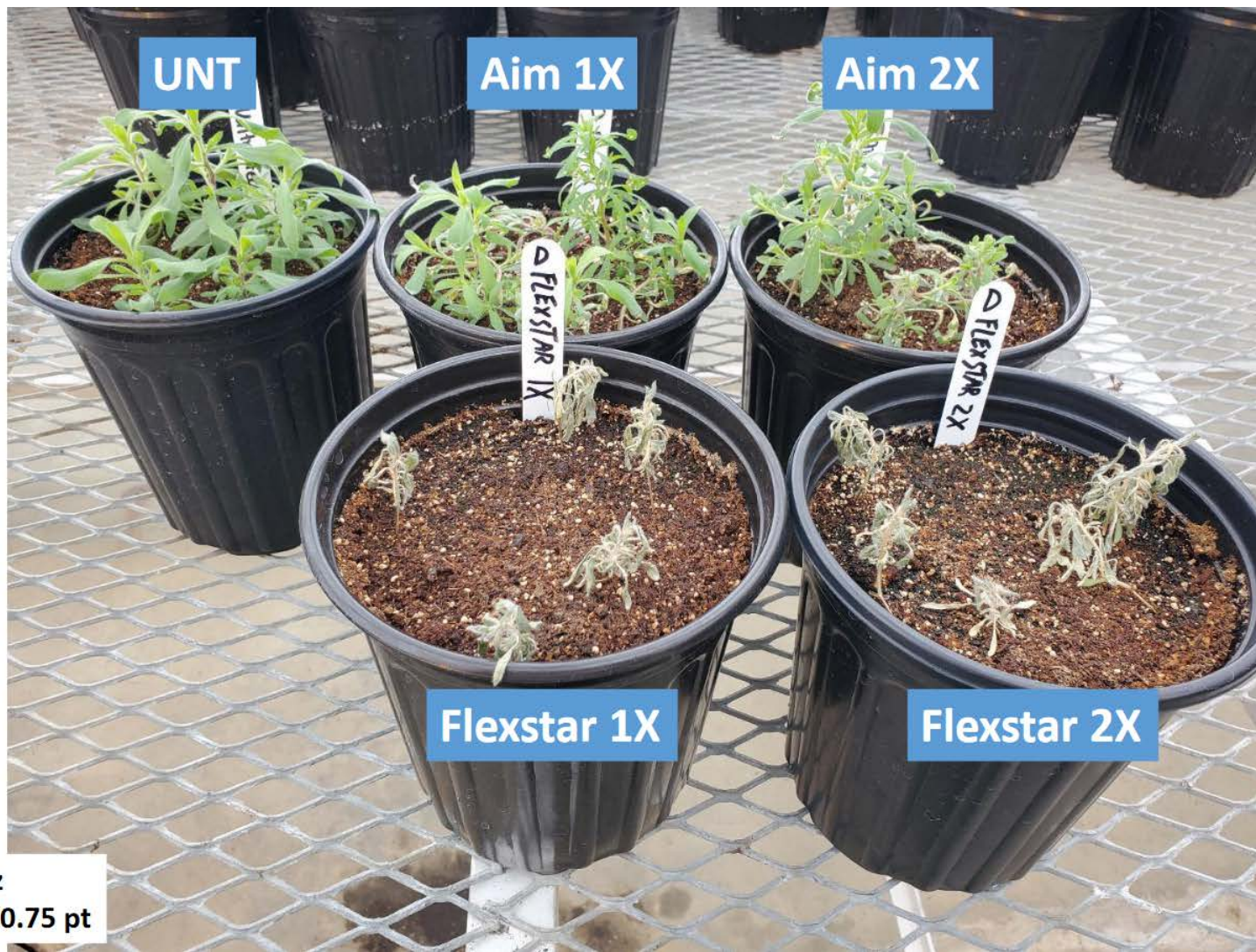
## 2.2 Plant protoporphyrinogen oxidases

There are two distinct isoforms of PPO in plants, as described above. Both are derived from a common HemY ancestor. Despite the common ancestry and identical functionality, the two isoforms carry <30% identity at the protein level. Both carry <30% identity to mammalian PPO proteins as well, although PPO1 and PPO2 carry approximately 70% identity to their counterparts in other plants.<sup>49</sup> The two isoforms separate into two branches on a phylogenetic tree generated from a homolog search for the tobacco (*Nicotiana tabacum*) PPO1 and PPO2 protein sequences (Fig. 4). The isoforms are separated clearly by phylum. Both monocots and eudicots are separated into the same branch of the tree for each isoform. The ability to mutate and gain resistance to PPO-inhibiting herbicides has been identified at two different regions. The blue squares in Fig. 4 indicate the only organisms that do not have the Arg98 in the active site. All plant species investigated maintained the arginine in both PPO1 and PPO2. All conserved residues of the active site, including the Arg98, are shown in Table 2. The tandem repeat facilitating the Gly210 deletion is only found in PPO2 in some plant species, identified by red triangles in Fig. 4. As mentioned in Section 1.5, *A. tuberculatus* and *A. palmeri* have evolved resistance to PPO-inhibiting herbicides via this deletion. Our sequence analysis identified *Kochia scoparia* as another problematic weed possessing this tandem repeat.<sup>50,51</sup> As the soybean market expands north and west in the USA, it will be interesting to see if *K. scoparia* evolves resistance to PPO inhibitors via this same deletion mutation. Other short tandem repeats in this region could also lead to the deletion of the same codon encoding other amino acid residues (green triangles in Fig. 4), but there is no evidence that this has occurred in resistant biotypes.



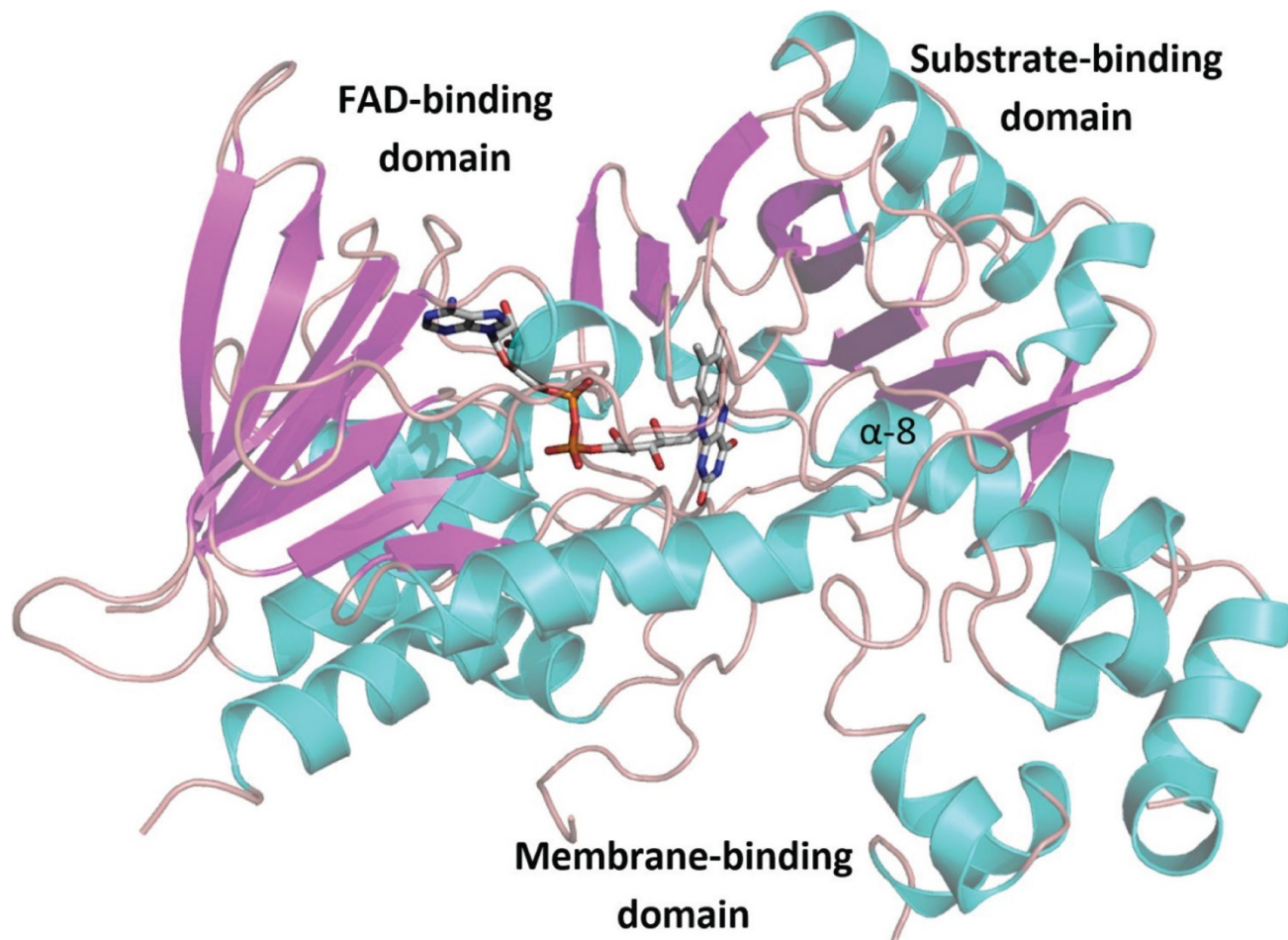
# HOWEVER

Minot

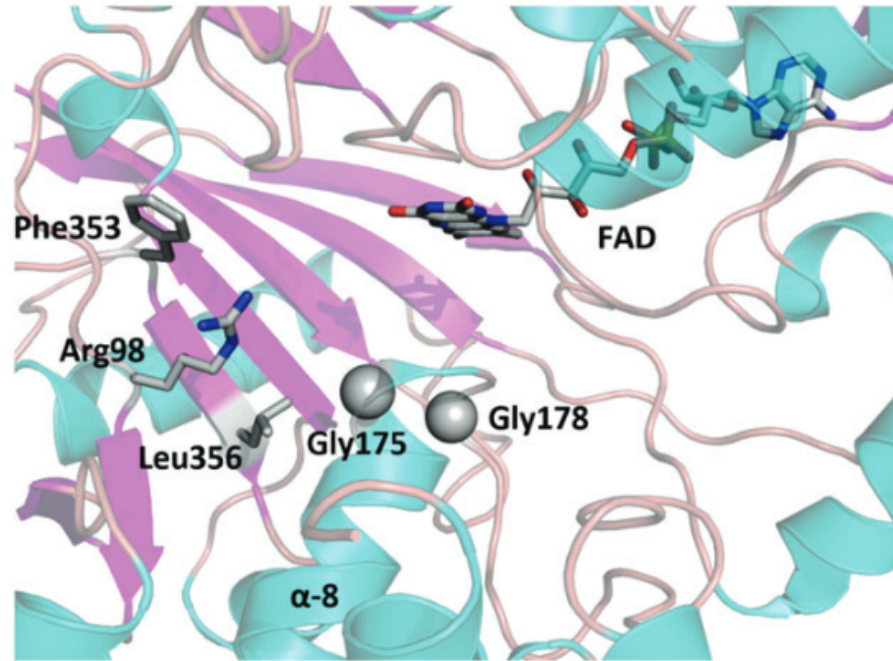


Aim 1X = 1 oz  
Flexstar 1X = 0.75 pt

# PPO-inhibitor (Group 14) Resistance



# PPO-inhibitor (Group 14) Resistance



**Figure 7.** Catalytic domain of *Nicotiana tabacum* mitochondrial PPO showing FAD forming the roof of the pocket and Gly175 at the terminal end of the  $\alpha$ -8 helix serving as a pivot for the tetrapyrrole ring at the bottom of the pocket. Gly178 indicates the site of a codon deletion leading to herbicide resistance on the mitochondrial PPO. Arg98 is involved in the positioning of the tetrapyrrole ring via ionic interactions of the carboxylic side chain of the substrate. Arg98 to Leu, Met or Gly substitutions on the mitochondrial enzyme are associated with PPO-herbicide resistance. The highly conserved Phe353 and Leu356 residues interact with the D-ring of the substrate and are thought to be involved in the binding of the herbicide.

# PPO-Inhibitor Resistance

## *Mechanisms of Resistance*

### Non-Target Site Resistance: Metabolism

- Wild oat
  - Resistance to sulfentrazone with no previous exposure
  - Enhanced cytochrome P450 activity associated with resistance to ACCase inhibitors
  
- Waterhemp
  - Resistance to HPPD inhibitors via metabolism
  - Selectivity within PPO inhibitors
    - Resistant to saflufenacil
    - Sensitive to lactofen
  
- Crop tolerance in soybean related to P450 activity for both sulfentrazone and saflufencil



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Received: 2 May 2021 | Accepted: 17 August 2021

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## ORIGINAL ARTICLE



WILEY

## Maternal water stress reduces sensitivity of offspring to herbicides in *Amaranthus palmeri* (Palmer amaranth)

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### Funding information

UC Davis New Research Initiatives and Interdisciplinary Research Grant; USDA-NIFA Grant, Grant/Award Number: CA-D-PLS-2511-H

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### Abstract

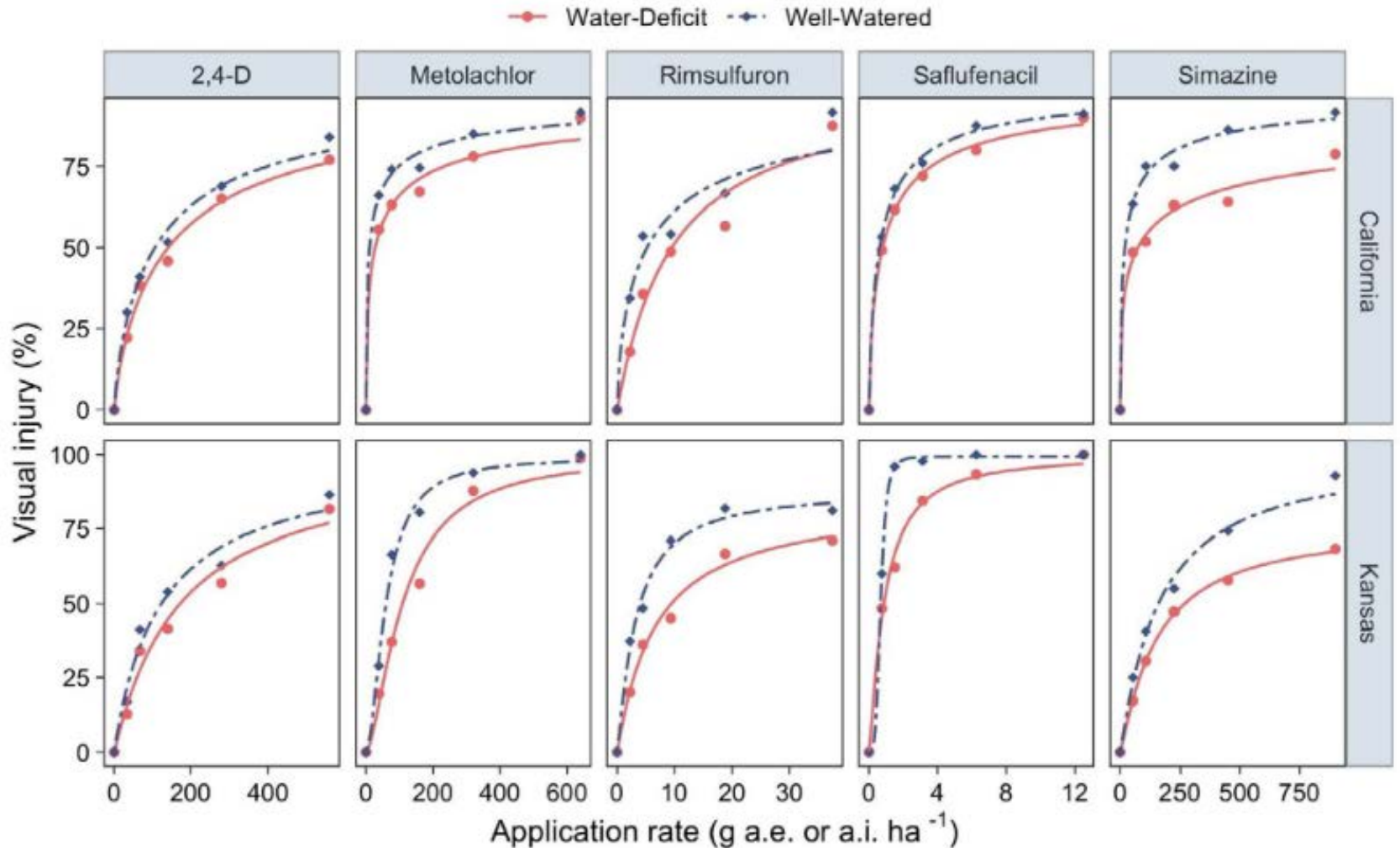
The immediate impact of prevailing environmental conditions on sensitivity of weeds to herbicides is well documented but little is known about the effects of parental environments on the responses of progeny to herbicides. It has been suggested that parental plants subjected to abiotic stress may result in progeny with increased tolerance to stresses. We tested this hypothesis by growing two populations of Palmer amaranth (*Amaranthus palmeri*) from California and Kansas under well-watered (WW) and water-deficit (25% of WW treatment) conditions to obtain F1 seeds. We then tested the responses of F1 seedlings to various doses of five herbicides with different modes of action. Dose-response analysis of visual plant injury, aboveground biomass, and chlorophyll content showed that in 16 out of 30 comparisons, sensitivity to herbicides in *A. palmeri* progeny originated from a maternally water stressed environment was significantly lower than those obtained from WW plants. Only in one case the progeny from stressed plants was significantly more sensitive to herbicide (saflufenacil) than that from non-stressed plants, and no differences were observed between the two types of progeny in all other (13) comparisons. The reduced sensitivity in progeny from stressed plants was consistent in terms of the evaluated response variables in population from California with the application of S-metolachlor, rimsulfuron, and simazine; and in population from Kansas with the application of S-metolachlor and rimsulfuron. Our study suggests that *A. palmeri* plants experiencing drought during the season may produce progeny that might be more difficult to control because of an increase in herbicide tolerance endowed through transgenerational effects of water stress.

### KEYWORDS

does response, drought, herbicide tolerance, Parental environmental effects, weed control

# Maternal water stress reduces sensitivity of offspring to herbicides in *Amaranthus palmeri* (Palmer amaranth)

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# Do Other Stresses Impact Herbicide Sensitivity?

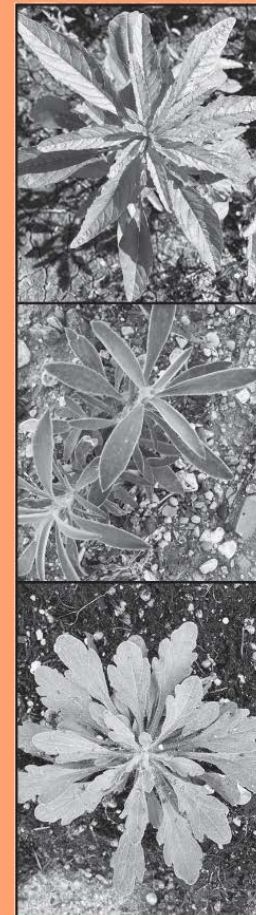
- Drought led to larger seed, quicker germination, and germination in drier conditions (Matzrafi et al (2020))
- Low dose of herbicides (Dyer 2018)
- Herbicide drift (Vieira et al 2020)
- Plants surviving desiccation??

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W253-23

This Publication Supersedes All Previous Issues

2023

## North Dakota Weed Control Guide

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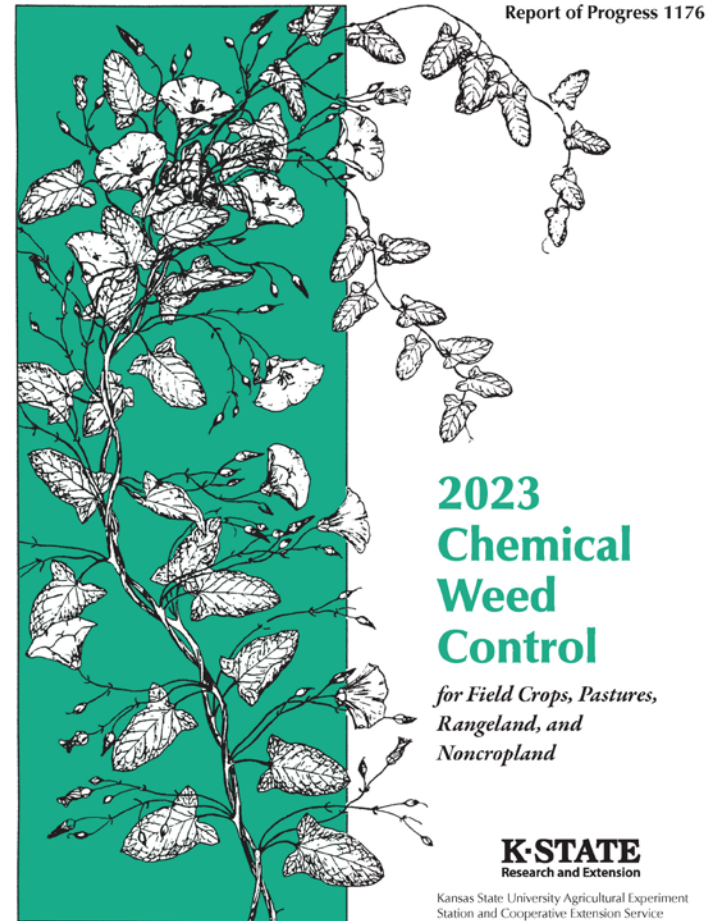
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