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Ciencia, producción y sociedad: hacia un manejo sustentable

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PPO-Inhibiting Herbicide Resistance: What We Have Learned in Almost 10 Years of Research



**Dr. Aimone Porri, Head of Weed Resistance Laboratories,
BASF Agricultural Solutions Deutschland GmbH**

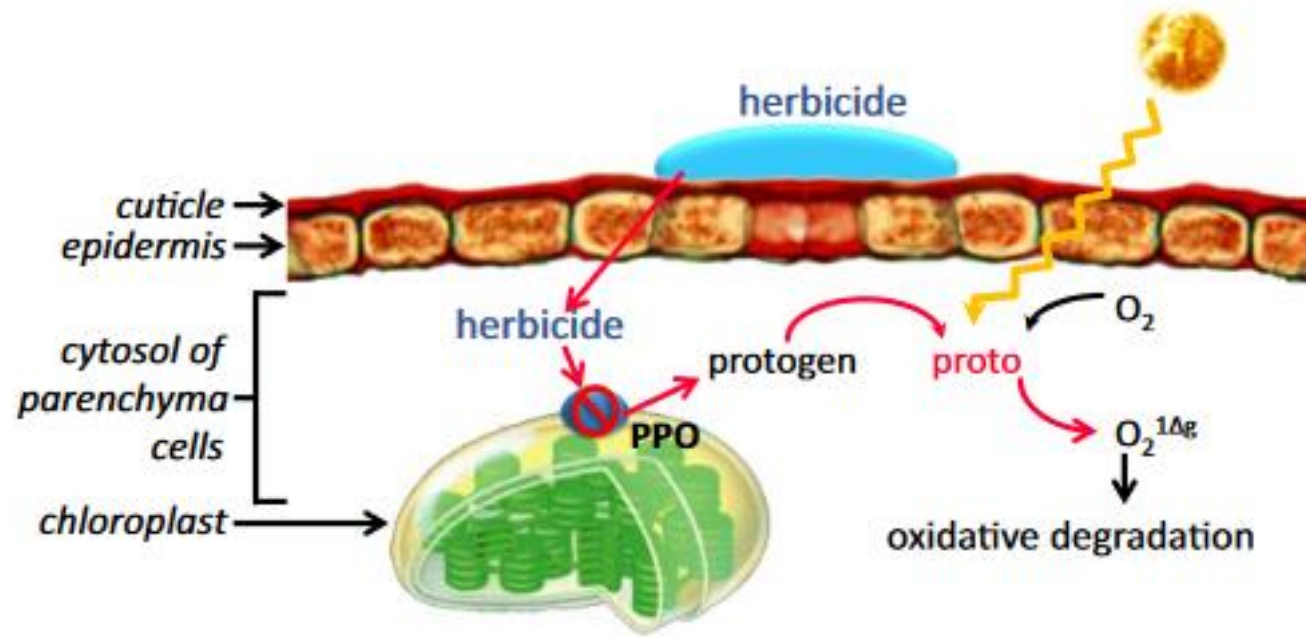
Keep up to date on my latest research on herbicide resistance [Dr Aimone Porri - Google Scholar](#)

Presentation outline

- ❖ Overview of PPO-inhibitor resistance
- ❖ Target-site mutations that have evolved
- ❖ Why the PPO isoform matters
- ❖ New PPO inhibitors to combat resistance
- ❖ What nearly 10 years of PPO-resistance research have taught us



PPO-inhibiting herbicides: an important mode of action since the 1960s



- ❖ PPO inhibitors block protoporphyrinogen oxidase, causing protoporphyrin IX to accumulate. In the presence of light and oxygen, it generates reactive oxygen species that destroy cell membranes, leading to rapid tissue desiccation and necrosis.

Recommended

Discovery, mode of action, resistance mechanisms, and plan of action for sustainable use of Group 14 herbicides

Abigail L. Barker¹, John Pawlak², Stephen O. Duke³, Roland Beffa⁴, Patrick J. Tranel⁵, Joe Wuerffel⁶, Bryan Young⁷, Aimone Porri⁸, Rex Liebl⁹, Raphael Aponte⁹, Douglas Findley⁹, Michael Betz¹⁰, Jens Lerchl⁸, Stanley Culpepper¹¹, Kevin Bradley¹² and Franck E. Dayan¹



Overview of PPO-inhibitor resistance, how has it evolved?

Table 2. Number of unique cases of resistance to protoporphyrinogen oxidase (PPO) inhibitors, and whether the case involves a single resistance or multiple resistance profile.^a

Weed species	Number of unique cases	Resistance to PPO-inhibiting herbicides	
		PPO inhibitors only	Multiple resistance
<i>Amaranthus</i> spp.	23		
<i>Amaranthus hybridus</i>	2	2	
<i>Amaranthus palmeri</i>	4	0	4
<i>Amaranthus retroflexus</i>	3	1	2
<i>Amaranthus tuberculatus</i>	14	2	12
<i>Ambrosia artemisiifolia</i>	6	0	6

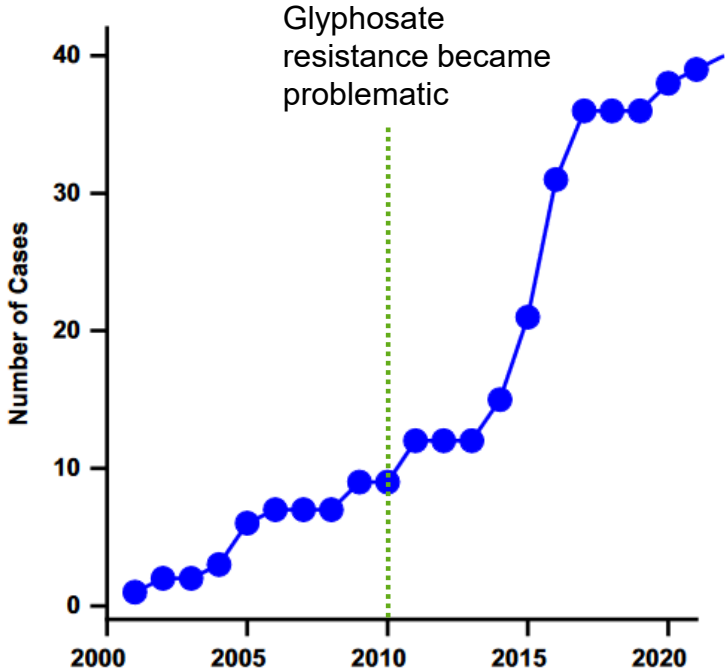
^aData from the International Herbicide-Resistant Weed Database (Heap 2023).



B. Scoparia (2023)

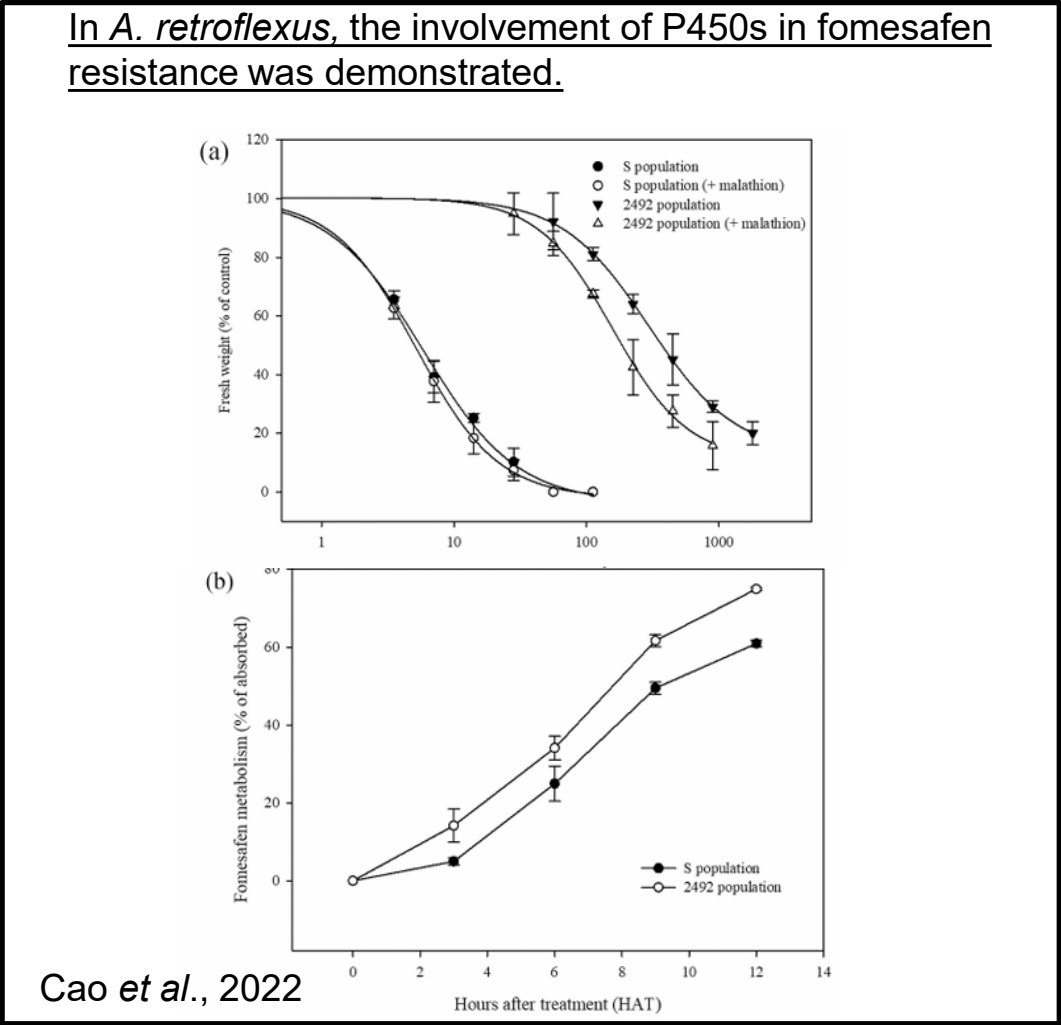
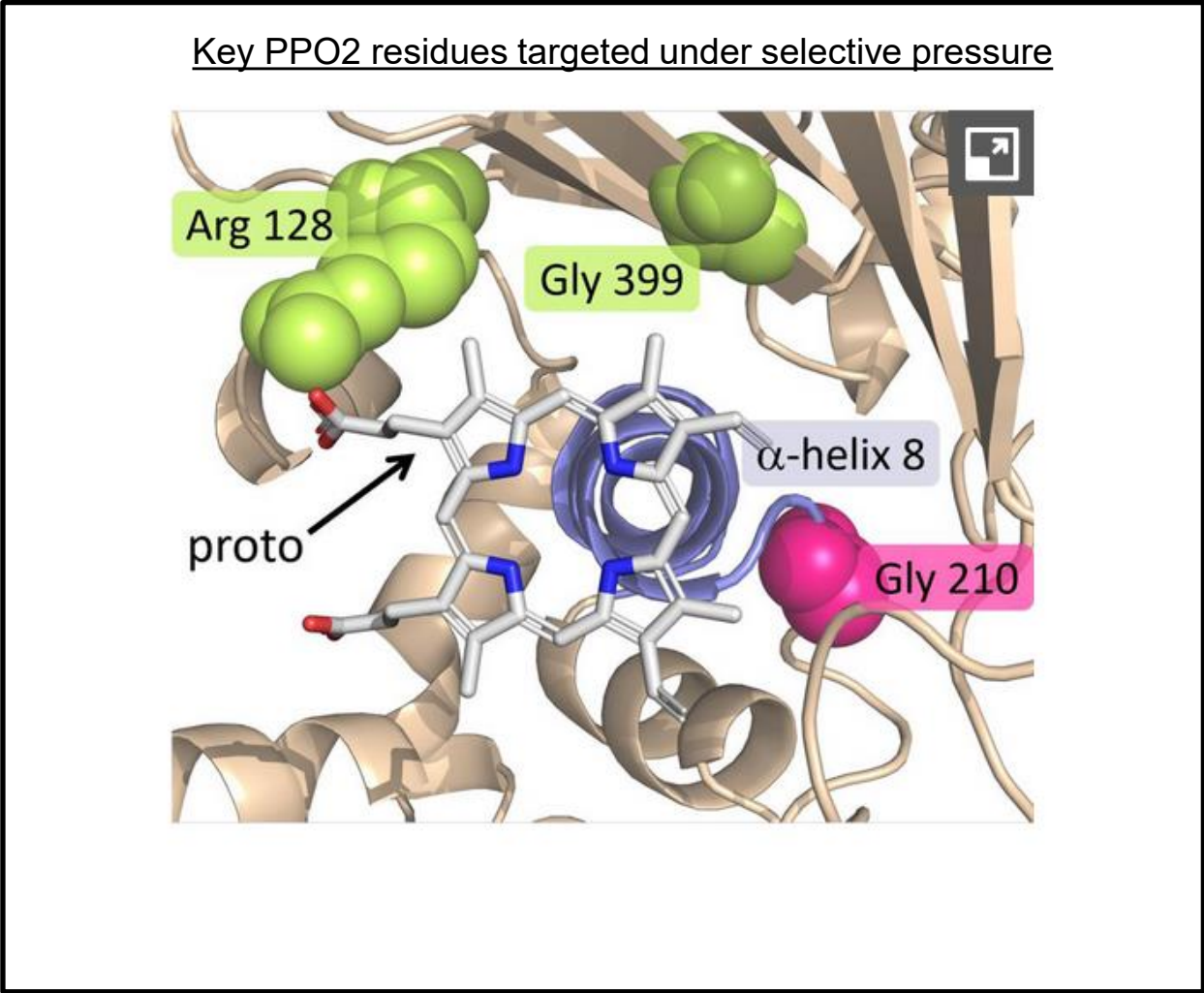
Grasses:

E. indica, *A. fatua*, *P. annua*, *Lolium*

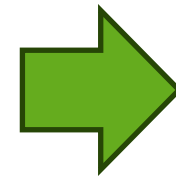
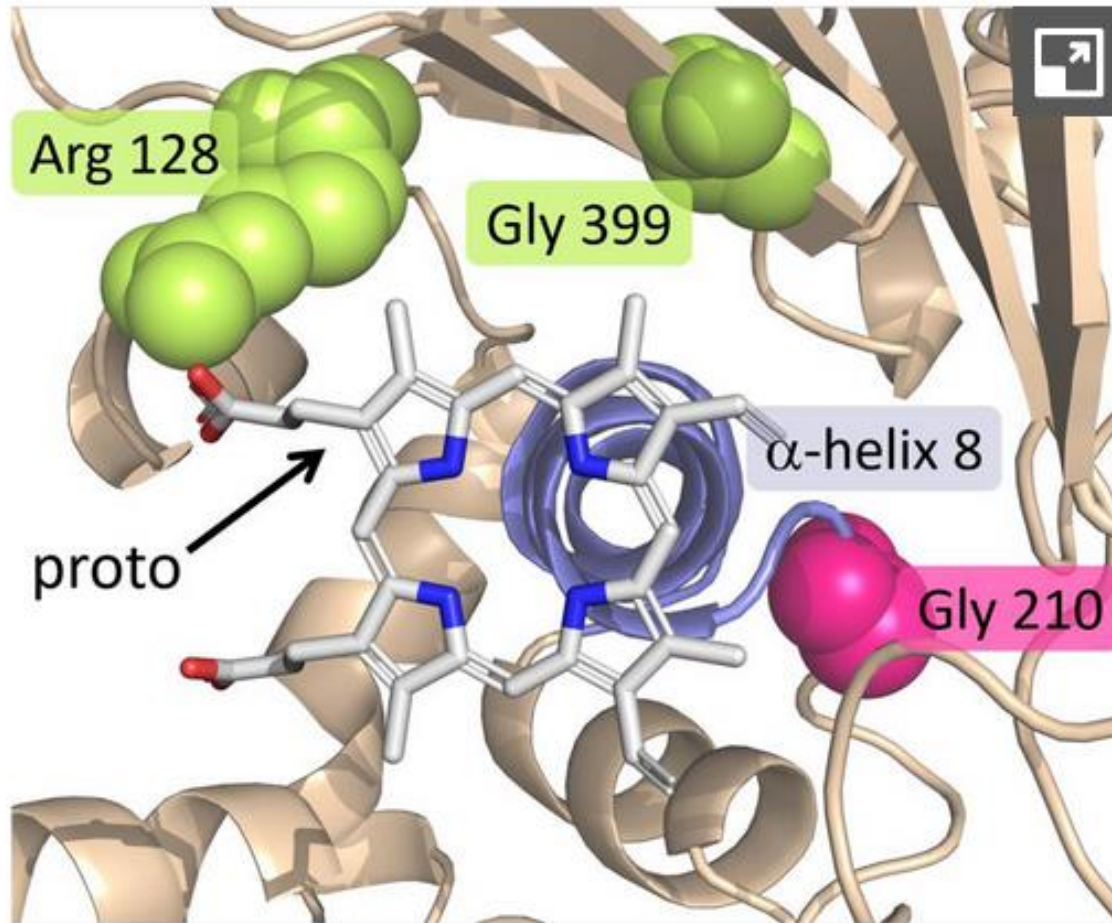


- ❖ PPO resistance was first reported in 2001, in common waterhemp from Kansas. However, it became a major problem mainly between 2013 and 2017. This happened because PPO inhibitors were increasingly used to control weeds already resistant to ALS inhibitors and glyphosate, creating strong selection pressure.

PPO target-site resistance dominates—but the picture is increasingly complex



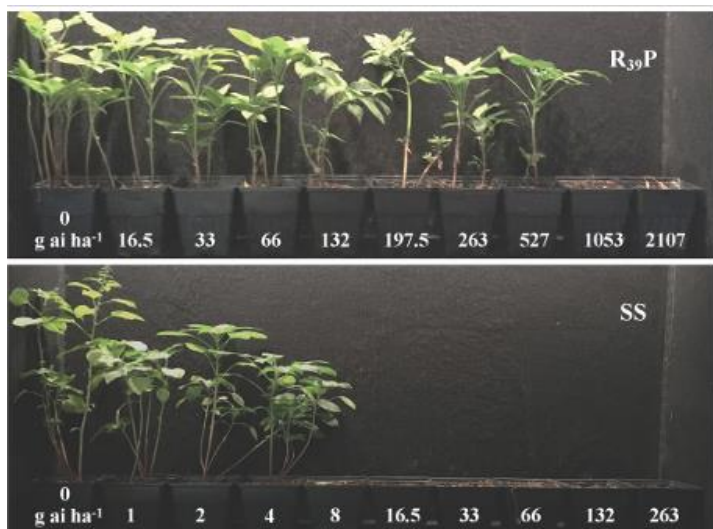
PPO2 became the primary target of resistance evolution



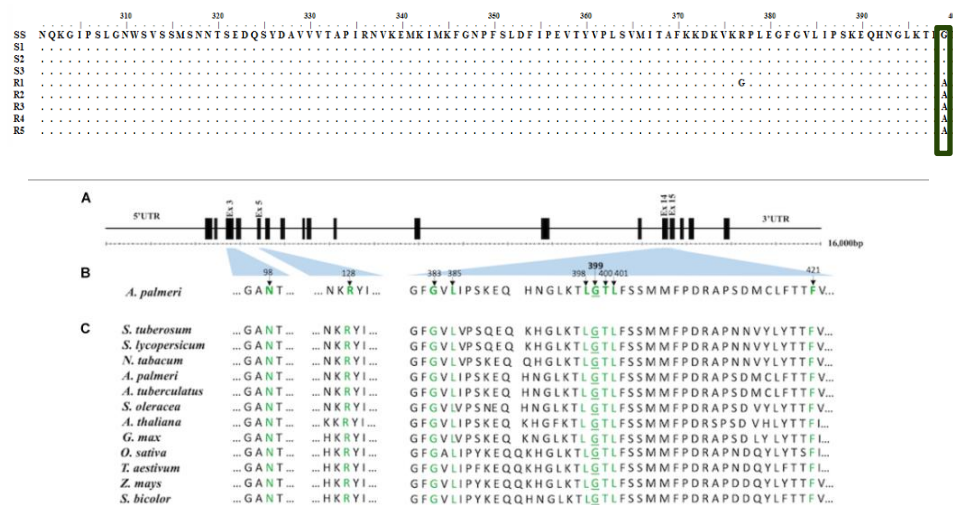
Target-site
mutation in PPO2

- ❖ Δ G210
- ❖ R128G, L, M
- ❖ G399A
- ❖ V361A

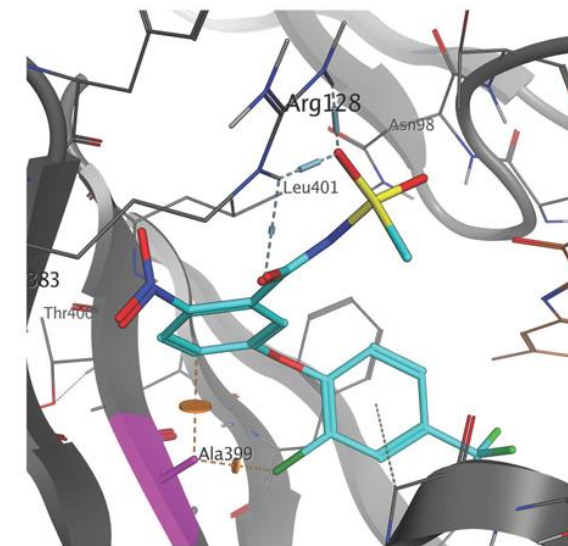
A novel catalytic-domain mutation G399A in *Amaranthus palmeri*



R39P *A. palmeri* showed high resistance to fomesafen.



R₃₉P *A. Palmeri* showed a G399A substitution in PPO2



The additional methyl (-CH₃) group of A399 creates repulsive interactions that weaken the inhibition of the PPO2 enzyme by fomesafen.

Recommended

A Novel Single-Site Mutation in the Catalytic Domain of Protoporphyrinogen Oxidase IX (PPO) Confers Resistance to PPO-Inhibiting Herbicides

Gulab Rangani¹, Reiofeli A. Salas-Perez¹, Raphael A. Aponte², Michael Knapp², Ian R. Craig², Thomas Mietzner², Ana Claudia Lango², Matheus M. Nogueira¹, Simone Porri² and Nilda Roma-Burgos^{1*}



Multiple PPO2 mutations can coexist in the same weed plant

Table 1. Summary of mutations in 115 *Amaranthus palmeri* accessions from the US Mid-south surviving the application of 264 g a.i. ha⁻¹ fomesafen +0.5% v/v nonionic surfactant, Fayetteville, Arkansas, USA, 2019

Mutation ^a		Accessions per state							
No. of mutations	ΔG210	R128G	R128M	G399A	AR	MO	MS	TN	Total
0	—	—	—	—	2	1	23	4	30
1	+	—	—	—	9	2	5	1	17
	—	+	—	—	4	2	3	1	10
	—	—	+	—	0	0	0	2	2
	—	—	—	+	2	1	0	1	4
2	+	+	—	—	2	5	2	0	9
	+	—	+	—	0	1	0	0	1
	+	—	—	+	11	7	0	0	18
	—	+	—	+	0	2	0	0	2
	—	+	+	—	0	0	1	0	1
3	+	+	—	+	1	11	0	0	12
	+	—	+	+	2	4	0	0	6
4	+	+	+	+	1	2	0	0	3

^a Up to 25 survivors per accession were genotyped individually for the ΔG210 mutation and DNA from up to eight survivors was pooled to detect the presence of other known mutations in the population. Accessions were collected in 2017 and 2018. AR, Arkansas; MO, Missouri; MS, Mississippi; TN, Tennessee. The green shade highlights the presence of each mutation in a population.

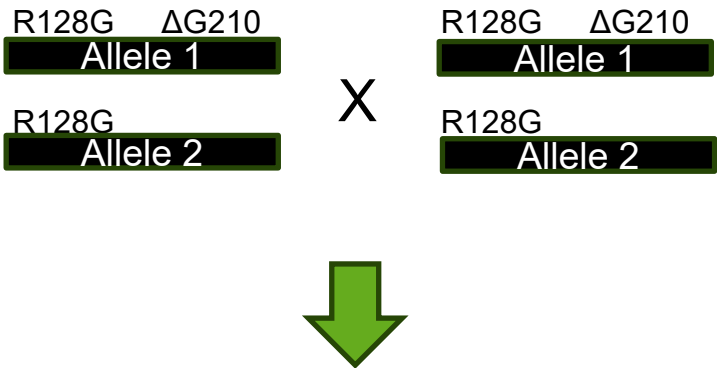
Recommended

Functional PPO2 mutations: co-occurrence in one plant or the same *ppo2* allele of herbicide-resistant *Amaranthus palmeri* in the US mid-south

Matheus M Noguera,^a Gulab Rangani,^a James Heiser,^b Taghi Bararpour,^c Lawrence E Steckel,^d Michael Betz,^e Aimone Porri,^e Jens Lerchl,^e Sophie Zimmermann,^e Robert L Nichols^f and Nilda Roma-Burgos^{a*}



Mutation accumulation has functional limits



- ❖ Sequencing of the *A. palmeri* PPO2 gene of more than 800 collected Palmer amaranth biotypes revealed
 - ❖ Population LIH1640 carrying R128G/ΔG210 double mutation on the same allele
 - ❖ R128G being homozygous
- ❖ Four male x female crosses did not reveal any homozygous double mutant among 400 offspring lines analyzed

Table 1. Segregation of mutant alleles in F ₁ and F ₂ generations of PPO-herbicide-resistant <i>A. palmeri</i> genotypes								
		Number of plants						
		Expected			Observed			
F ₁ line	Genotyped	128G +/+ Δ210 –/–	128G +/+ Δ210 +/-	128G +/+ Δ210 +/+	128G +/+ Δ210 –/–	128G +/+ Δ210 +/-	128G +/+ Δ210 +/+	χ ² Calc
LIH11640 F ₁	100	25	50	25	71	29	0	118.46
LIH11640 F ₂ -1	100	25	50	25	65	35	0	93.50
LIH11640 F ₂ -2	100	25	50	25	73	27	0	127.74
LIH11640 F ₂ -3	100	25	50	25	70	30	0	114.00
Grand total	400	100	200	100	279	121	0	451.62

No homozygous double mutants could be found and the ΔG210 mutation is much lower represented as compared to Mendelian segregation pattern

Recommended

Research Article

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(wileyonlinelibrary.com) DOI: 10.1002/ps.6850

Can double PPO mutations exist in the same allele and are such mutants functional?

Aimone Porri,^{a*} Matheus M Noguera,^{b*} Michael Betz,^a Daniel Sälinger,^a Frank Brändle,^c Steven J Bowe,^d Jens Lerchl,^a Lucie Meyer,^a Michael Knapp^a and Nilda Roma-Burgos^{b*}

109/10/2026

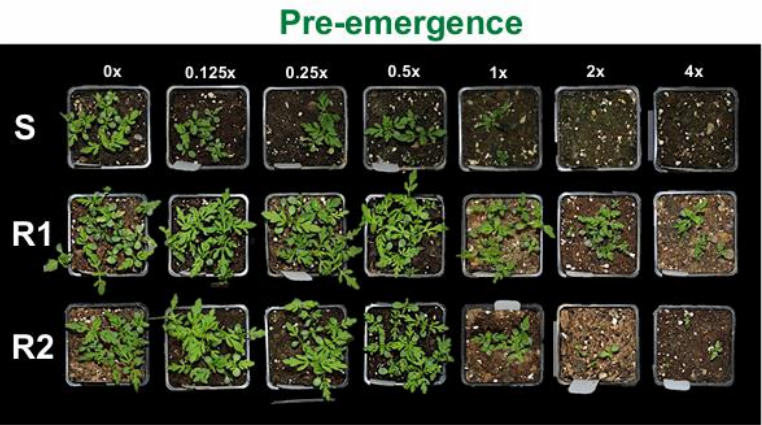
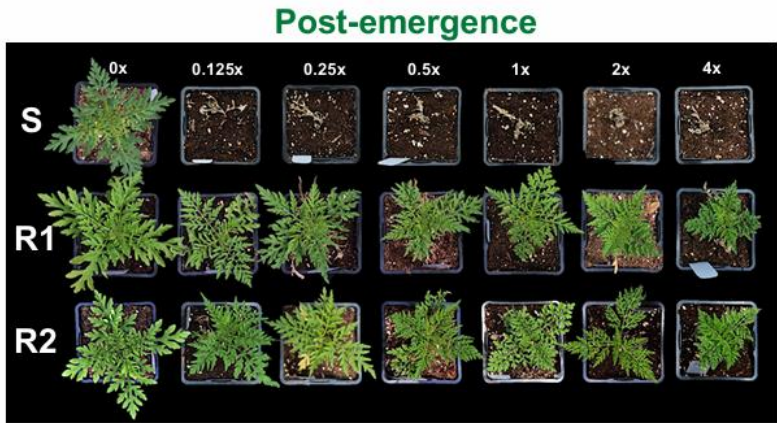
Internal

BASF

We create chemistry

New PPO2 substitutions continue to emerge

Sulfentrazone



A Novel Arg99Gln substitution in PPO2

R99Q

S -----CCAATTCACAGCACAAACGGACATTGTAAGAAA
R GTTGACAGCAATTCACAGCACAAACAGACATTGTAAGAAA

S cca att tca cag cac aag cgg tac att gta aga
P I S Q H K R Y I V R

R cca att tca cag cac aag cag tac att gta aga
P I S Q H K Q Y I V R

Sequence alignment of PPO gene shows a **single nucleotide substitution** (highlighted in green) associated with resistance to PPO-inhibiting herbicides (such as sulfentrazone). The mutation occurs at **codon 99**, where the **wild-type codon CGG** (arginine, R) is replaced by **CAG** (glutamine, Q) in resistant individuals.

Recombinant expression and *in vitro* enzyme inhibition analysis

We performed an *in vitro* inhibition study using recombinant PPO2 proteins from both the susceptible and Arg99Gln mutant variants of *A. artemisiifolia*. Proteins were expressed and purified in *E. coli*, and enzyme activity was evaluated against 14 different PPO-inhibiting herbicides to assess the functional impact of the mutation. Results showed that the **R99Q** substitution markedly reduces sensitivity to **saflufenacil** and **sulfentrazone**.

Table 1. Effects of protoporphyrinogen oxidase (PPO) inhibitors on *in vitro* enzyme activity of recombinant *Ambrosia artemisiifolia* PPO2 wild-type and Arg99Gln mutant variants.

Herbicide	Susceptible R99	Resistant R99Q
Diphenyl ethers		
Fomesafen	1.67E-06	1.28E-07
Lactofen	6.51E-09	1.46E-09
Nitrofen	2.81E-07	5.09E-08
Acifluorfen	1.57E-06	2.70E-07
N-Phenylimides		
Butafenacil	2.55E-10	2.64E-08
Saflufenacil	3.56E-08	>1.00E-05
N-Phenyltriazolinones		
Trifludimoxazin	2.72E-10	1.39E-08
Tiafenacil	1.01E-09	7.99E-07
Epyrifenacil	4.71E-10	4.81E-07
Others		
Sulfentrazone	7.51E-07	>1.00E-05
Carfentrazone-ethyl	6.76E-10	1.14E-06
Pyraclozil	2.25E-08	1.05E-06
Pyraflufen-ethyl	3.00E-10	1.64E-08
Oxadiazon	1.78E-09	3.29E-07

New Results

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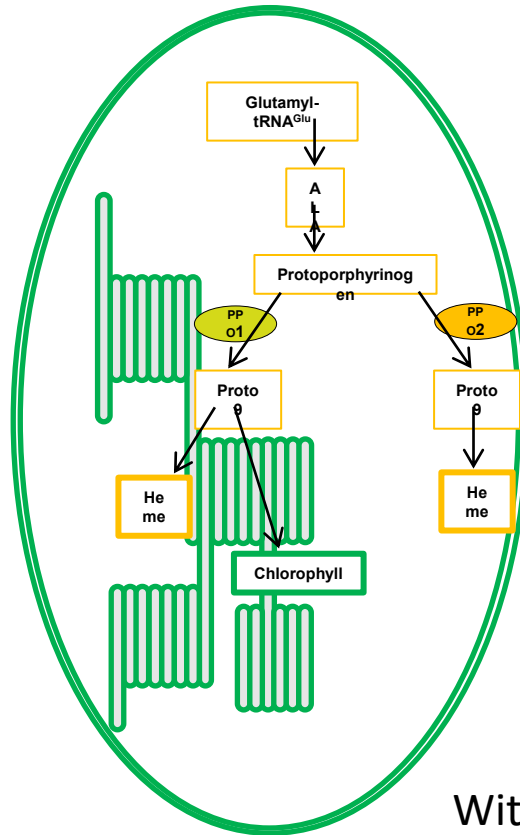
Characterization of a novel R98Q mutation that confers resistance to sulfentrazone in common ragweed (*Ambrosia artemisiifolia*) populations from Michigan

Sara Álvarez-Rodríguez, Michael Ozolins, Aimone Porri, Jens Lerchl, Eric Patterson

doi: <https://doi.org/10.64898/2026.07.31.742095>

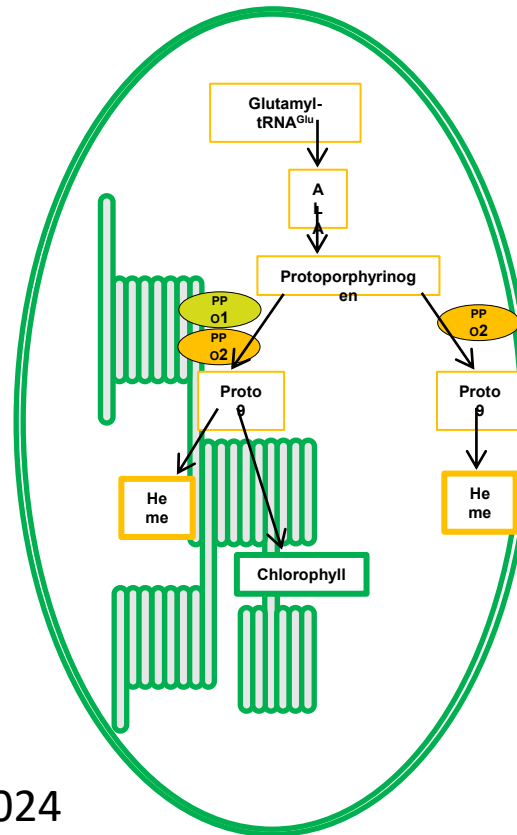
Why the PPO isoform matters?

Arabidopsis & Tobacco



Wittmann *et al.*; 2024

Amaranth



- ❖ In Amaranthus, PPO2 is localized in the envelope and thylakoid membranes, while PPO1 is localized only in the thylakoid membrane
- ❖ PPO1 accounts for most of total PPO activity in green tissue. Target site resistance mutations would have a detrimental effect on plant fitness.
- ❖ This is the reason why PPO2 and not PPO1 target site resistance mutations were selected through selective pressure.

PPO1 can also become a resistance target; the *E. indica* case

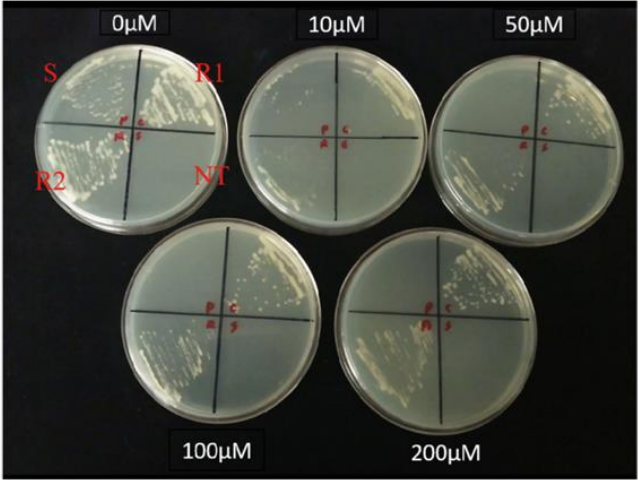
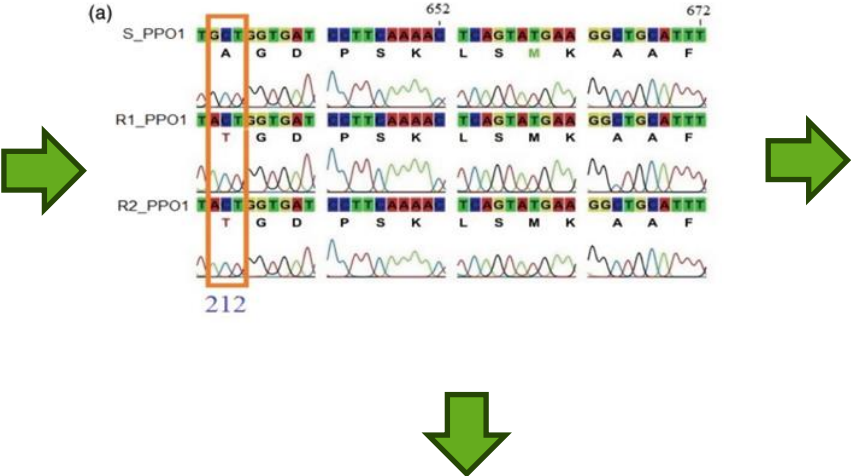
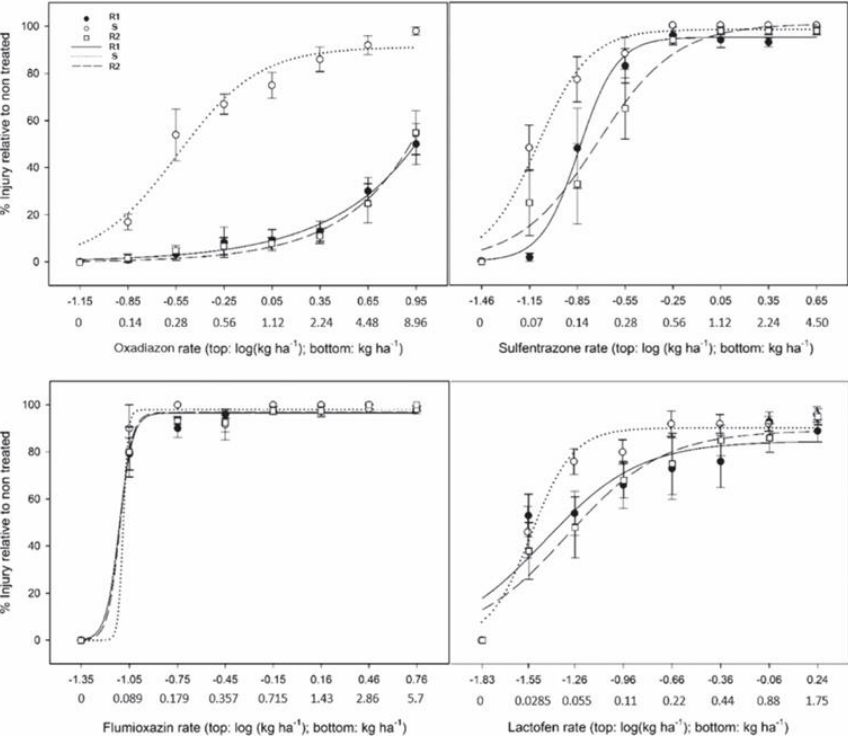


Table 2. Effects of protoporphyrinogen oxidase (PPO) inhibitors on *in vitro* enzyme activity of recombinant *E. indica* wild-type PPO1 and variant A212T PPO1 enzyme

PPO chemical family	Herbicides	Wild-type <i>E. indica</i> PPO1 sensitivity (IC ₅₀) (M)	Variant <i>E. indica</i> PPO1 A212T sensitivity (IC ₅₀) (M)	Wild-type <i>E. indica</i> PPO1 % inhibition ^a (%)	Variant <i>E. indica</i> PPO1 A212T % inhibition ^a (%)
Oxadiazoles	Oxadiazon	2.47 × 10 ⁻⁸	>1.00 × 10 ⁻⁵	>90	16
Triazolinones	Sulfentrazone	2.75 × 10 ⁻⁷	1.87 × 10 ⁻⁶	>90	76
Pyrimidinediones	Saflufenacil	1.94 × 10 ⁻⁸	4.49 × 10 ⁻⁷	>90	>90
Diphenyl ethers	Lactofen	2.64 × 10 ⁻⁸	3.62 × 10 ⁻⁷	>90	>90
<i>N</i> -Phenyl-phthalimides	Flumioxazin	1.67 × 10 ⁻⁸	1.34 × 10 ⁻⁸	>90	>90
	Trifludomoxazin	1.71 × 10 ⁻⁸	2.65 × 10 ⁻⁸	>90	>90

^a % inhibition with the rate of the highest herbicide concentration 10⁻⁵ M.

Research Article

Received 20 October 2019 | Revised 26 November 2019 | Accepted article published 1 December 2019 | Published online in Wiley Online Library 12 January 2020

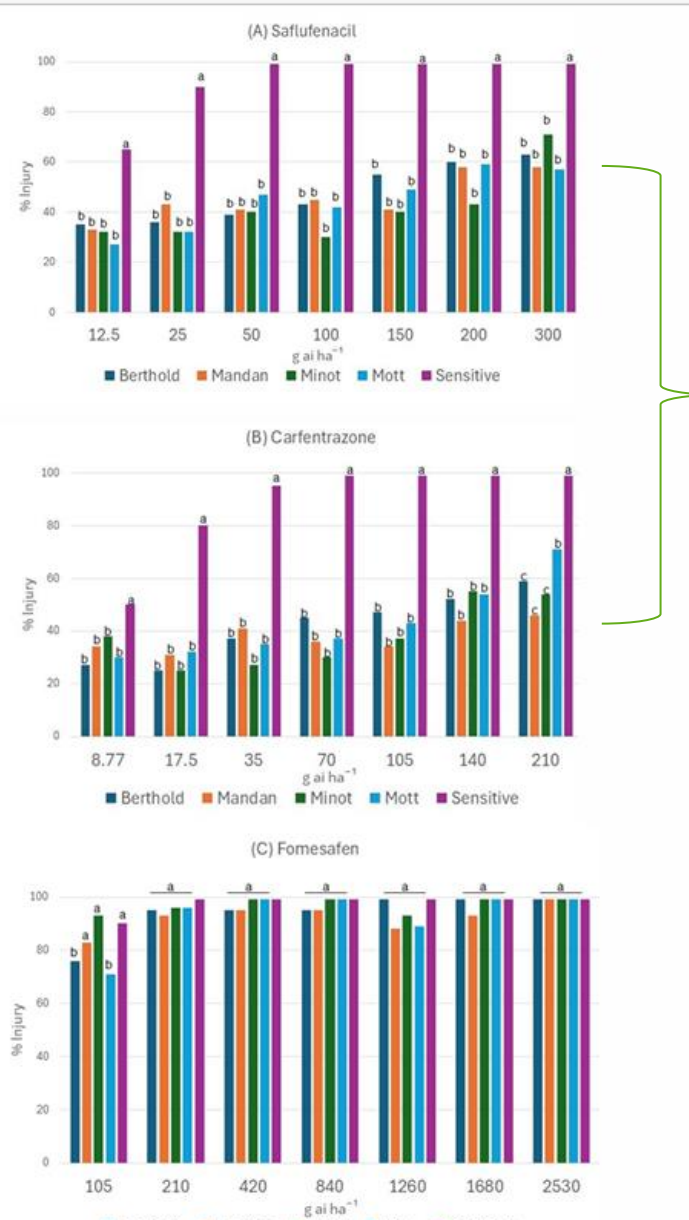
DOI 10.1002/ps.5703

A novel mutation A212T in chloroplast Protoporphyrinogen oxidase (PPO1) confers resistance to PPO inhibitor Oxadiazon in *Eleusine indica*

Bo Bi,^a Qiang Wang,^b Jeffrey J Coleman,^b Aimone Porri,^c John M Peppers,^a Jinesh D Patel,^a Michael Betz,^c Jens Lerchl^d and J Scott McElroy^{a,*}

Recommended

PPO1 resistance has now evolved in *Bassia scoparia* from North Dakota

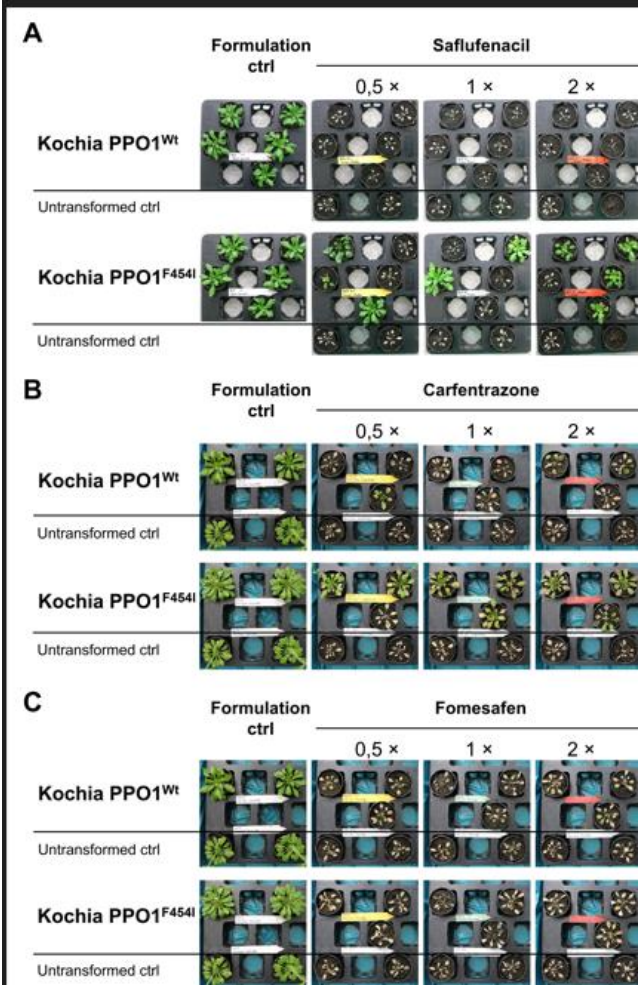


Population	Survivor ID	<i>B. scoparia</i> PPO1	F454
Berthold	Survivor 1		◇ I
	Survivor 2		◇ F/I
	Survivor 3		◇ I
Mandan	Survivor 1		◇ I
	Survivor 2		◇ I
	Survivor 3		◇ F/L
Minot	Survivor 1		◇ I/L
	Survivor 2		◇ L
	Survivor 3		◇ V
Mott	Survivor 1		◇ L
	Survivor 2		◇ F/I
	Survivor 3		◇ F/I

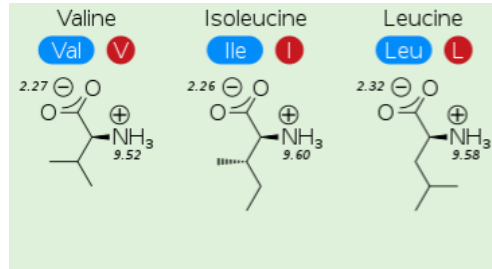
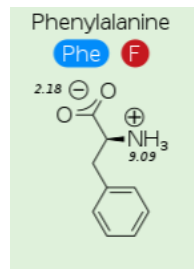
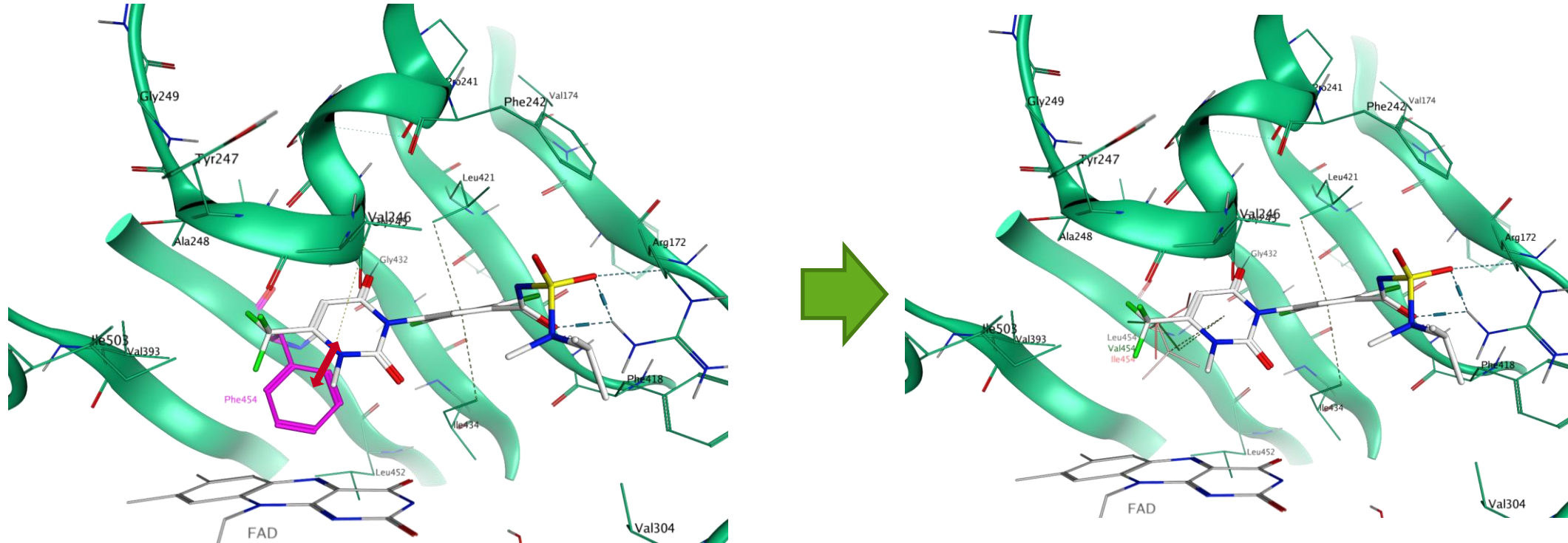
iNovel amino acid substitutions in Protoporphyrinogen oxidase 1 endow resistance to PPO-inhibiting herbicides in *Bassia scoparia*

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Aimone Porri¹Quincy D. Law²Charles M. Geddes³



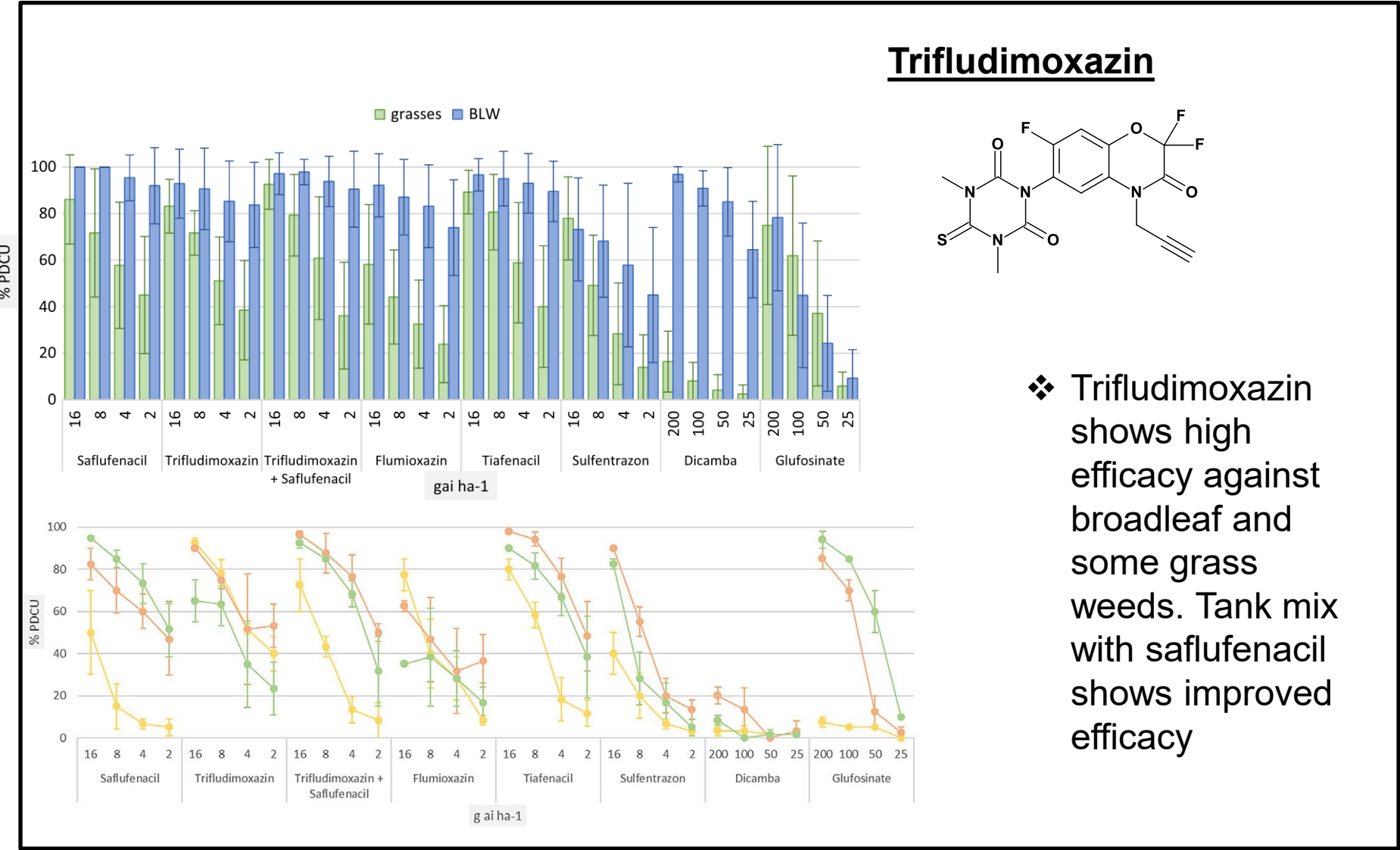
Modeling of saflufenacil into the *B. scoparia* PPO1 active site



The phenyl group of F454 is involved in pi-stacking interaction with saflufenacil

Substitutions of F454 to L, V and I disrupt the pi-stacking interaction

New PPO inhibitors to combat resistance; Trifludimoxazin



❖ Trifludimoxazin shows high efficacy against broadleaf and some grass weeds. Tank mix with saflufenacil shows improved efficacy

BASF has a solid portfolio of next-generation PPO herbicides to manage resistance



Recommended

Complementary activity of trifludimoxazin and saflufenacil when used in combination for postemergence and residual weed control

Published online by Cambridge University Press: 13 November 2024

Liliana Parra Rapado, Frederik Uwe Gerhard Kölpin, Silke Zeyer, Ulrike Anders, Laurent Piccard, Simone Porri and Scott Asher



BASF
We create chemistry

Trifludimoxazin is a potent inhibitor of PPO1 and PPO2 isoforms

Comparative enzyme inhibition studies



PPO2 Inhibition*

Herbicide	IC50 (nM)
Fomesafen	7.72
Saflufenacil	3.58
<u>Trifludimoxazin</u>	0.71



PPO1 Inhibition*

Herbicide	IC50 (nM)
Fomesafen	64.2
Saflufenacil	41.2
<u>Trifludimoxazin</u>	9.8

*IC₅₀ against wildtype AMATU-PPO,
overexpressed in *E. coli*; for PPO1
additional non binding site modifications

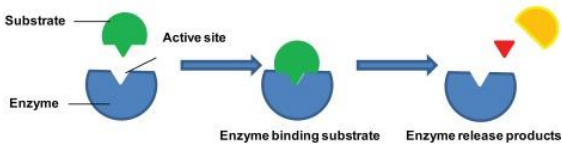
Trifludimoxazin is highly active against the two PPO isoforms

Trifludimoxazin retains activity against the most widespread PPO2 target site mutations

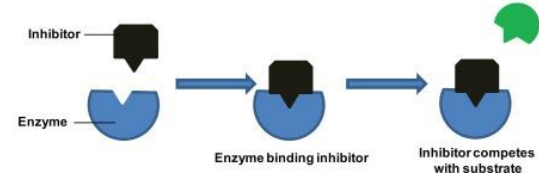
Selection of PPO2 mutations: ΔG210, R128G and G399A

 Expression of *A. Tuberculatus* PPO2 mutant enzymes in *E. Coli*

(a) Reaction



(b) Inhibition



- Enzyme assay
- ▶ Inhibitor concentrations:
 - 1.00E10-05
 - 2.00E10-06
 - 4.00E10-07
 - 8.00E10-08
 - 1.60E10-08
 - 3.20E10-09
 - 6.40E10-10
 - 1.28E10-10
 - 2.56E10-11
 - 5.12E10-12



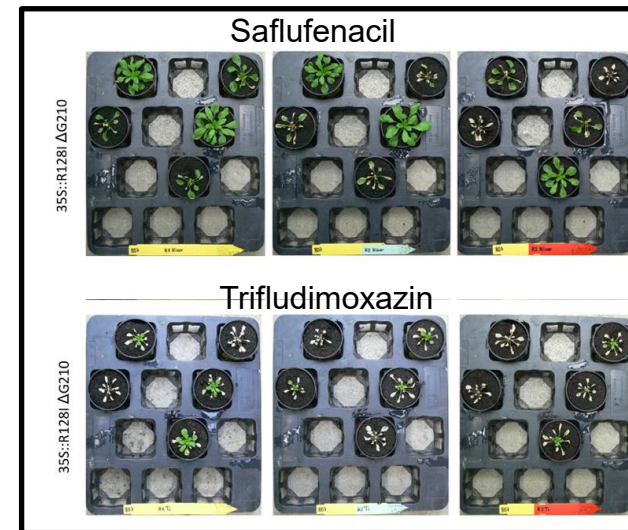
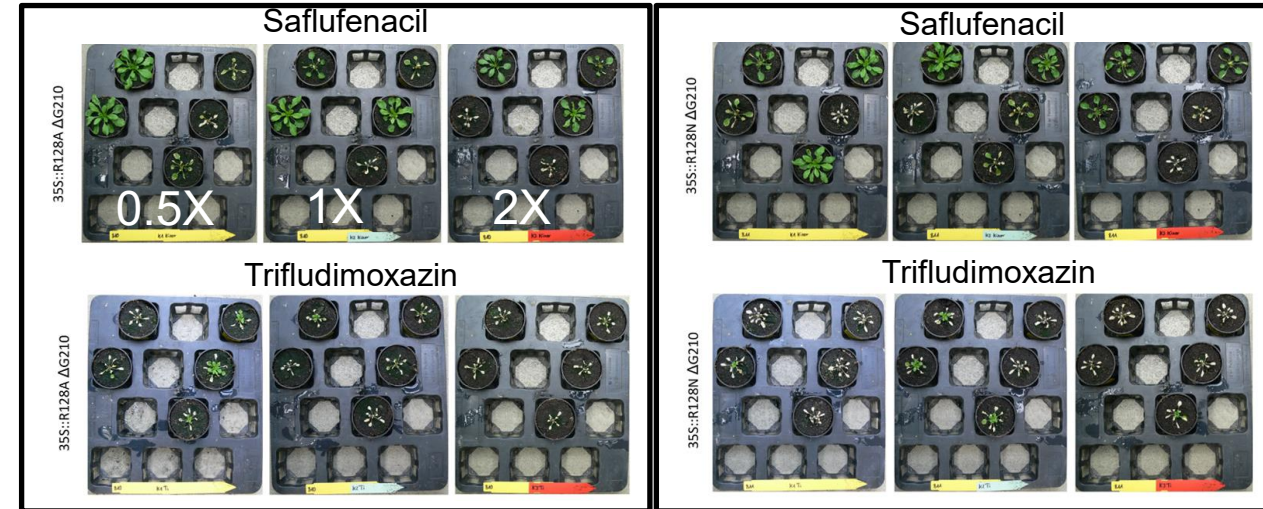
IC50 [M]

Enzyme	Variant AMAPA	Trifludimoxazin	Epyrifenacil	Tiafenacil	Oxadiazon	Saflufenacil	Remaining protein activity %
PPO2	Wild type	4.79E-10	2.11E-10	6.27E-10	5.89E-09	7.76E-10	100
PPO2	ΔG210	1.78E-09	2.56E-07	1.43E-07	1.09E-06	2.04E-06	10
PPO2	R128G	4.80E-10	1.59E-09	4.56E-09	8.57E-09	6.61E-07	60
PPO2	G399A	3.48E-010	6.87E-08	7.30E-08	5.89E-07	6.27E-07	15

Trifludimoxazin retains efficacy against PPO2 double mutants, *in vitro* and *in vivo*

IC50 [M]

Mutant Variant	Trifludimoxazin IC50 (M)	Saflufenacil IC50 (M)	Lactofen IC50 (M)	Fomesafen IC50 (M)
<u>Wild type</u>	2.45E-10	1.89E-09	4.15E-10	7.13E-09
<u>ΔG210</u>	1.83E-09	2.04E-06	5.44E-7	>1E-05
R128A ΔG210	3.30E-08	>1E-05	>1E-05	>1E-05
R128C ΔG210	3.11E-08	>1E-05	2.15E-06	>1E-05
R128D ΔG210	Inactive	Inactive	Inactive	Inactive
R128F ΔG210	Inactive	Inactive	Inactive	Inactive
R128G ΔG210	Inactive	Inactive	Inactive	Inactive
R128H ΔG210	3.02E-08	>1E-05	1.59E-06	>1E-05
R128I ΔG210	9.93E-08	>1E-05	1.95E-06	>1E-05
R128E ΔG210	8.61E-09	>1E-05	6.49E-07	>1E-05
R128K ΔG210	Inactive	Inactive	Inactive	Inactive
R128L ΔG210	5.68E-09	>1E-05	6.49E-7	>1E-05
R128M ΔG210	1.82E-07	>1E-05	>1E-05	>1E-05
R128N ΔG210	1.75E-07	>1E-05	>1E-05	>1E-05
R128Q ΔG210	1.45E-08	>1E-05	1.75E-06	>1E-05
R128T ΔG210	7.48E-08	>1E-05	3.92E-06	>1E-05
R128Y ΔG210	Inactive	Inactive	Inactive	Inactive
R128P ΔG210	Inactive	Inactive	Inactive	Inactive
R128S ΔG210	Inactive	Inactive	Inactive	Inactive
R128V ΔG210	Inactive	Inactive	Inactive	Inactive
R128W ΔG210	Inactive	Inactive	Inactive	Inactive



Pest Management
Science



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Inhibition profile of trifludimoxazin towards PPO2 target site mutations

Aimone Porri, Michael Betz, Kathryn Seebruck, Michael Knapp, Philipp Johnen, Matthias Witschel, Raphael Aponte, Rex Liebl, Patrick J. Tranel, Jens Lerchi

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BASF
We create chemistry

Trifludimoxazin retains activity against the *Amaranth* Δ G210 PPO2 mutant biotype

post-emergence
spray at 4–6-inch
AMAPA plants
(photo taken 20
days after
treatment)



Recommended

Tirexor[®]—design of a new resistance
breaking protoporphyrinogen IX
oxidase inhibitor

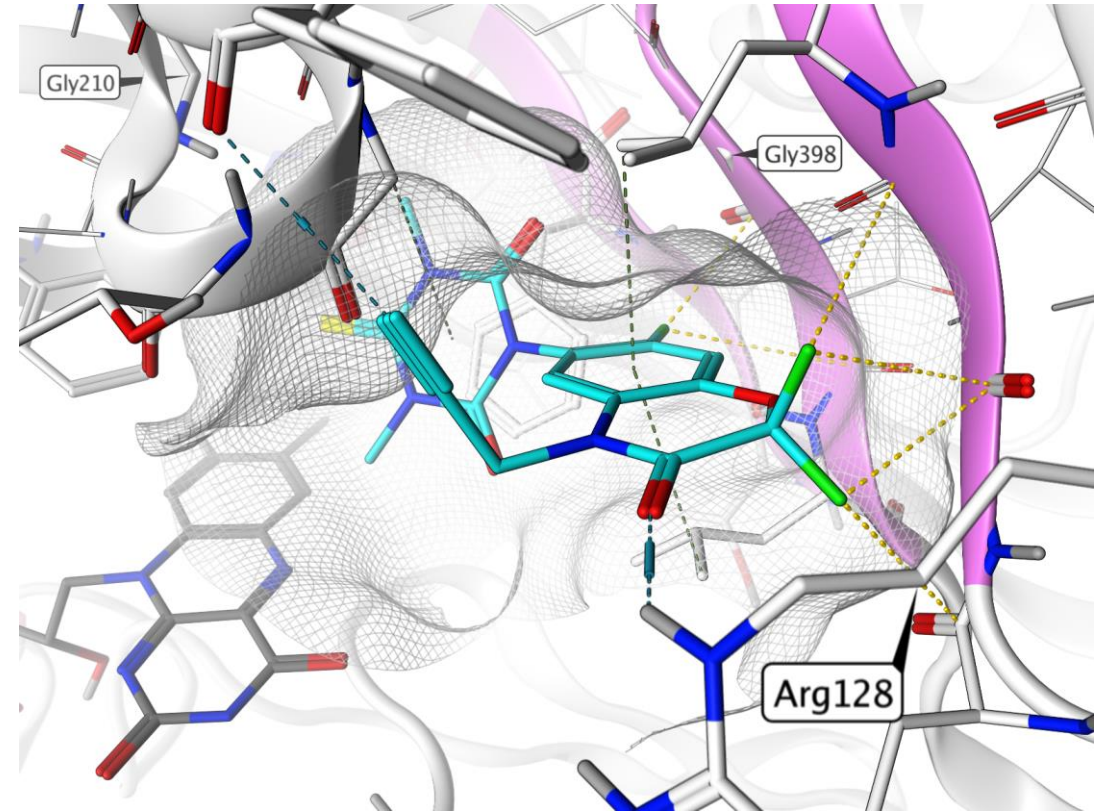
Matthias Witschel*, Raphael Aponte, Gregory Arnel,
Peter Bowerman, Thomas Mietzner, Trevor Newton,
Aimone Porri, Anja Simon and Thomas Seitz



Trifludimoxazin shows high performance in the field against
Amaranthus mutant harboring Δ G210 deletion

Why does trifludimoxazin control PPO2 mutations better?

- ❖ Trifludimoxazin has more binding interactions than other PPO actives
- ❖ Quantity and quality of binding moieties increase binding strength
- ❖ 2 fluorine atoms bind to the beta wall
- ❖ Propargyl group allows additional binding



Trifludimoxazin based products like Tirexor® can be expected to be a powerful tool against PPO target site resistant weeds in the near future

New PPO inhibitors to combat resistance; Fendioxypyracil

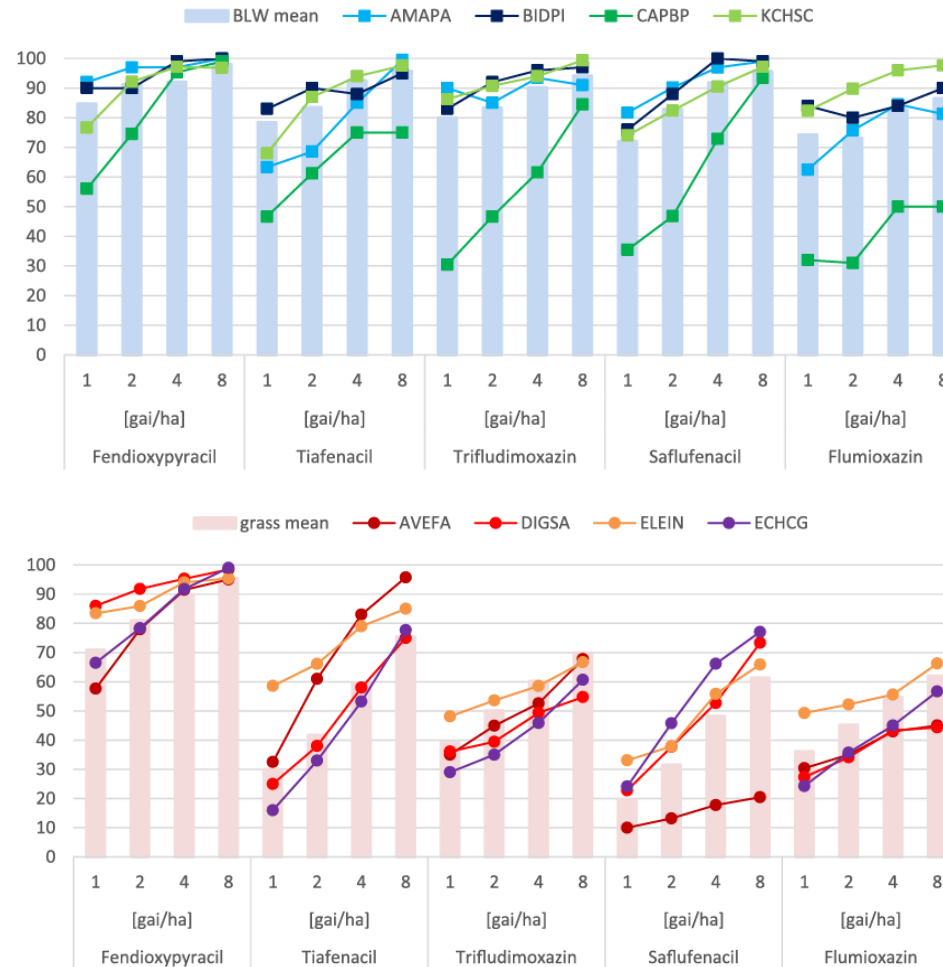
BASF has a solid portfolio of next generation PPO herbicides to manage resistance



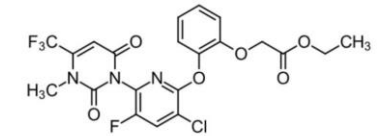
Recommended

Fendioxypyracil, a new and systemic PPO-inhibiting herbicide for X-spectrum weed control

Tobias Seiser,¹ Aimone Porri,² Philipp Johnen,³ Silke Zeyer,⁴ Judith Wahrheit,⁵ Michael Betz,⁶ Breght Vandenberghe,⁷ Scott Asher,⁸ and Lilliana Parra Rapado⁹



Fendioxypyracil



- ❖ Fendioxypyracil shows high efficacy against broadleaf and grass weeds.

Fendioxypyracil is a potent inhibitor of PPO1 and PPO2

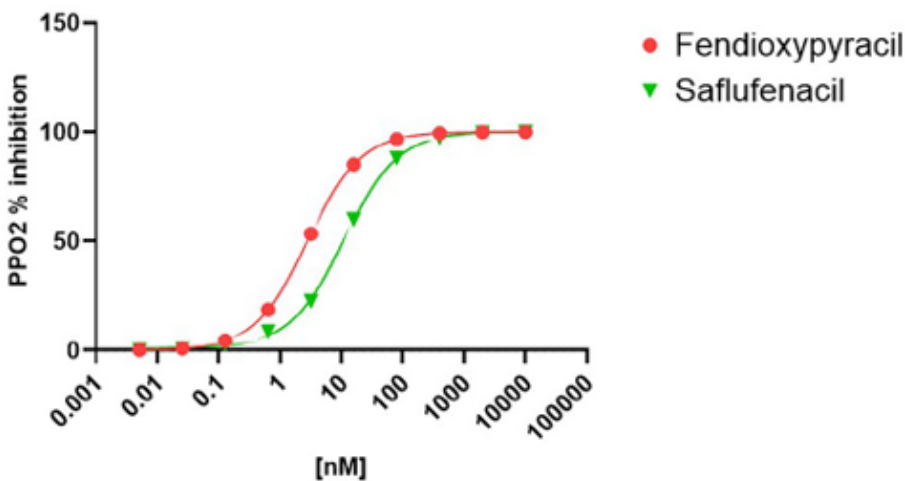
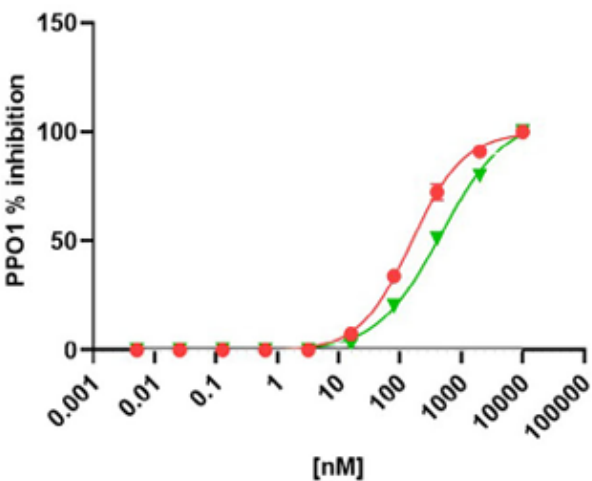
Table 4. *In vitro* inhibition of *Amaranthus tuberculatus* PPO1 and PPO2 wild-type enzymes by fendioxypyracil and the benchmark herbicide saflufenacil. IC₅₀ values (nM) represent the herbicide concentration required to reduce enzyme activity by 50%, as estimated from nonlinear regression of dose–response curves. Saflufenacil was included as a reference compound. % inhibition values were determined at a saturating concentration of 10 µM, at which both enzymes were fully inhibited. Each value is an average of three replications (*P* < 0.0001)

PPO isoform	Enzyme backbone	Fendioxypyracil IC50[nM] <i>P</i> < 0.0001	Saflufenacil IC50[nM] <i>P</i> < 0.0001	Fendioxypyracil % Inhibition at saturation	Saflufenacil % Inhibition at saturation
PPO1	AMATU	195	412	100	100
PPO2	AMATU	2.8	11	100	100

Recommended

Fendioxypyracil, a new and systemic PPO-inhibiting herbicide for X-spectrum weed control

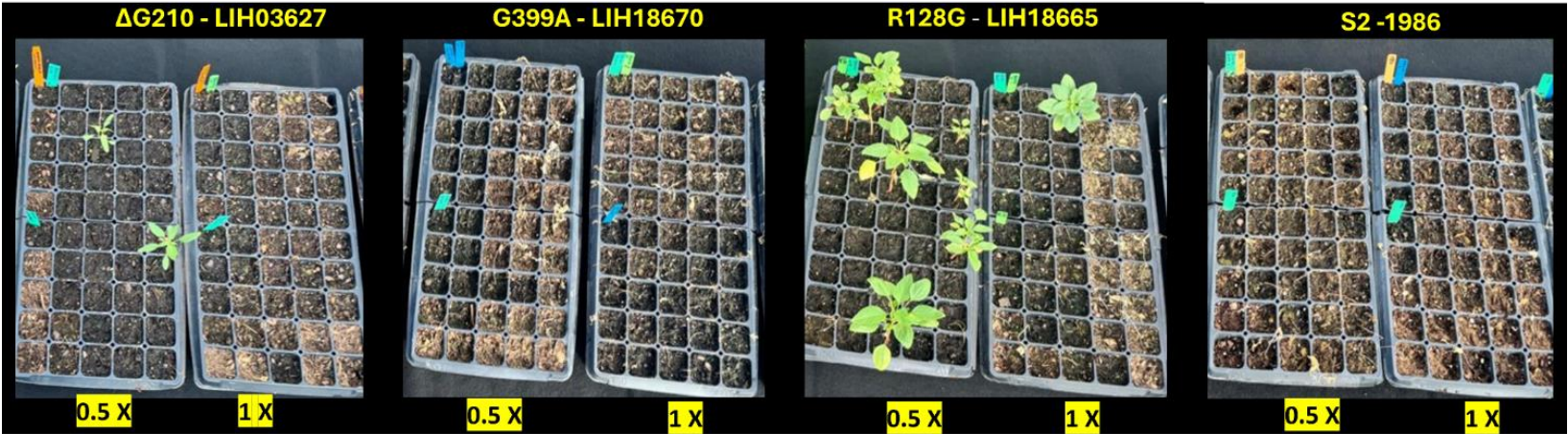
Tobias Seiser,¹ Almone Porri,² Philipp Johnen,³ Silke Zeyer,⁴ Judith Wahrheit,⁵ Michael Betz,⁶ Breght Vandenberghe,⁷ Scott Asher,⁸ and Liliana Parra Rapado¹



Fendioxypyracil expands the resistance-breaking potential

IC50 [M]

Enzyme	Variant (AMAPA)	Fendioxypyracil	Epyrifenacil	Tiafenacil	Oxadiazon	Saflufenacil	Remaining protein activity %
PPO2	Wild type	4.79E-10	2.11E-10	6.27E-10	5.89E-09	7.76E-10	100
PPO2	ΔG210	1.14E-07	2.56E-07	1.43E-07	1.09E-06	2.04E-06	10
PPO2	R128G	4.20E-09	1.59E-09	4.56E-09	8.57E-09	6.61E-07	60
PPO2	G399A	2.58E-08	6.87E-08	7.30E-08	5.89E-07	6.27E-07	15



Fendioxypyracil

Fendioxypyracil retains activity against engineered grass PPO2 target site mutations

G210 of *A. palmeri* corresponds to A213 in *S. viridis*

IC50 [M]

	Compounds	Fendioxypyracil	Epyrifenacil	Tiafenacil	Oxadiazon	Pyraclozil	Saflufenacil	Remaining protein activity %	Likelihood
PPO2	WT SETVI	2,32E-09	1,81E-09	2,62E-09	1,53E-08	3,15E-08	1,12E-08	100,00	
PPO2	A213N	4,52E-08	4,11E-08	4,66E-07	1,59E-05	6,33E-09	6,32E-09	2,60	more than 1 nucleodite change
PPO2	A213C	4,00E-09	1,83E-09	5,26E-09	8,26E-08	4,90E-08	1,33E-07	100,00	more than 1 nucleodite change
PPO2	A213Q	2,12E-06	2,56E-06	1,00E-05	1,00E-05	1,00E-05	1,00E-05	0,60	more than 1 nucleodite change
PPO2	A213G	8,41E-10	8,70E-10	2,48E-09	7,23E-08	5,19E-08	9,36E-09	69,00	1 nucleodite change
PPO2	A213I	1,54E-09	3,30E-09	4,68E-09	2,65E-07	9,78E-08	1,89E-07	3,70	more than 1 nucleodite change
PPO2	A213L	3,03E-09	8,49E-09	2,05E-08	1,52E-06	2,11E-07	9,51E-07	14,00	more than 1 nucleodite change
PPO2	A213M	2,76E-08	1,17E-07	1,83E-07	6,23E-06	4,42E-06	3,06E-06	8,60	more than 1 nucleodite change
PPO2	A213F	1,25E-07	1,82E-07	5,67E-06	1,00E-05	1,00E-05	1,00E-05	0,80	more than 1 nucleodite change
PPO2	A213P	4,67E-07	1,80E-06	4,93E-06	1,00E-05	1,00E-05	1,00E-05	1,00	1 nucleodite change
PPO2	A213S	8,63E-10	1,22E-09	2,90E-09	4,57E-08	8,71E-08	1,01E-08	17,00	1 nucleodite change
PPO2	A213T	1,27E-09	2,16E-09	2,88E-09	4,22E-07	2,31E-07	1,34E-07	13,70	1 nucleodite change
PPO2	A213V	6,59E-10	1,87E-09	4,15E-09	5,69E-07	1,67E-07	1,97E-07	11,70	1 nucleodite change

IC50 [M] G399 of *A. palmeri* corresponds to G409 in *S. viridis*

		Fendioxypyracil	Epyrifenacil	Tiafenacil	Saflufenacil	Oxadiazon	Pyraclozil	Remaining protein activity %	Likelihood
	WT SETVI	2,32E-09	1,67E-09	1,81E-09	1,02E-08	1,31E-08	2,29E-08	100,00	
PPO2	G409A	2,20E-09	1,06E-09	2,89E-08	3,81E-07	1,07E-06	1,00E-05	6,80	1 nucleodite change
PPO2	G409E	1,11E-09	5,52E-10	1,98E-08	3,33E-07	8,29E-07	1,00E-05	0,23	1 nucleodite change
PPO2	G409S	4,06E-09	3,91E-09	3,52E-08	2,74E-07	4,46E-07	6,17E-07	0,35	more than 1 nucleodite change

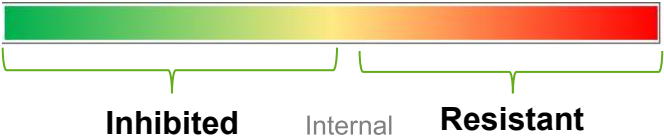


Fendioxypyracil retains activity against engineered grass PPO2 target site mutations

R128 of *A. palmeri* corresponds to R128 in *S. viridis*

IC50 [M]

		Epyrifenacil	Fendioxypyracil	Tiafenacil	Oxadiazon	Pyraclonil	Saflufenacil	Remaining protein activity %	Likelihood
PPO2	WT SETVI	1,67E-09	2,32E-09	1,81E-09	1,31E-08	2,29E-08	1,02E-08	100,00	more than 1 nucleodite change
PPO2	R128A	2,39E-10	2,97E-10	1,85E-09	7,09E-09	1,31E-08	1,38E-07	4,60	more than 1 nucleodite change
PPO2	R128N	1,26E-09	1,92E-09	6,55E-09	1,59E-08	4,43E-08	2,65E-07	23,60	more than 1 nucleodite change
PPO2	R128D	6,24E-09	4,40E-09	1,83E-08	1,38E-07	2,97E-07	2,42E-06	8,20	more than 1 nucleodite change
PPO2	R128C	7,36E-10	1,29E-09	5,16E-09	1,27E-08	4,00E-08	4,06E-07	20,30	1 nucleodite change
PPO2	R128E	1,87E-09	3,83E-09	3,68E-09	1,81E-08	2,68E-08	7,67E-07	0,60	more than 1 nucleodite change
PPO2	R128Q	5,88E-10	7,84E-10	5,46E-09	1,07E-08	1,47E-08	2,86E-07	5,90	more than 1 nucleodite change
PPO2	R128G	3,48E-10	7,36E-10	6,21E-09	4,94E-08	5,08E-08	3,70E-07	4,30	1 nucleodite change
PPO2	R128H	4,28E-10	1,03E-09	3,36E-09	4,55E-09	1,09E-08	4,87E-08	23,50	1 nucleodite change
PPO2	R128I	5,70E-10	1,27E-09	1,77E-09	8,09E-09	3,39E-08	2,00E-07	12,30	more than 1 nucleodite change
PPO2	R128L	7,05E-10	2,03E-09	3,36E-09	1,22E-08	1,11E-07	6,54E-07	10,20	1 nucleodite change
PPO2	R128K	1,09E-09	2,07E-09	5,11E-09	1,94E-08	4,41E-08	3,03E-07	7,20	more than 1 nucleodite change
PPO2	R128M	4,95E-10	1,09E-09	5,93E-09	1,60E-08	1,80E-08	7,57E-07	9,40	more than 1 nucleodite change
PPO2	R128F	5,65E-10	1,02E-09	4,98E-09	1,45E-08	3,00E-08	5,05E-07	5	more than 1 nucleodite change
PPO2	R128P	2,62E-10	8,61E-10	1,40E-09	2,57E-09	1,74E-08	1,16E-07	4	1 nucleodite change
PPO2	R128S	9,41E-10	1,72E-09	5,69E-09	2,41E-08	1,78E-08	2,68E-07	78	1 nucleodite change
PPO2	R128T	4,30E-10	8,37E-10	7,05E-10	6,70E-09	3,88E-08	6,02E-08	23	more than 1 nucleodite change
PPO2	R128W	3,78E-09	7,63E-09	3,77E-08	2,37E-08	3,32E-07	1,93E-06	2,86	more than 1 nucleodite change
PPO2	R128Y	5,71E-10	1,23E-09	2,90E-09	7,39E-09	4,26E-08	7,53E-07	4,44	more than 1 nucleodite change
PPO2	R128V	1,51E-09	3,98E-09	4,07E-09	8,02E-09	5,13E-08	2,98E-07	30	more than 1 nucleodite change



What has nearly 10 years of PPO-resistance research taught us?

- ❖ PPO resistance continues to evolve — and diversify new target-site mutations are emerging in both PPO2 and PPO1.
- ❖ Resistance evolution is constrained by PPO function. Not every mutation — or combination of mutations — is compatible with maintaining sufficient PPO activity and plant fitness.
- ❖ The PPO isoform matters. The biological role and cellular localization of PPO1 and PPO2 strongly influence which resistance mechanisms can evolve.
- ❖ New PPO inhibitors can overcome established resistance. Compounds such as trifludimoxazin and fendioxpyracil retain activity against important PPO target-site mutations and expand the resistance-breaking potential of the mode of action.
- ❖ Staying ahead of resistance requires more than chemistry. Mechanistic understanding + functional validation + new chemistry + stewardship will be essential to preserve the PPO mode of action.

BASF Global R&D has a globally recognized team studying herbicide resistance

“over 50 research reports on herbicide resistance published since 2019”

A Novel Single-Site Mutation in the Catalytic Domain of Protoporphyrinogen Oxidase IX (PPO) Confers Resistance to PPO-Inhibiting Herbicides

Gulab Rangani¹*, Reiofeli A. Salas-Perez¹, Raphael A. Aponte², Michael Knapp³, Ian R. Craig⁴, Thomas Mietzner⁴, Ana Claudia Langaro³, Matheus M. Nogueira¹, Almone Porri² and Nilda Roma-Burgos^{1*}

Pest Management Science

Research Article

A novel mutation A212T in chloroplast Protoporphyrinogen oxidase (PPO1) confers resistance to PPO inhibitor Oxadiazon in *Eleusine indica*

Bo Bi, Qiang Wang, Jeffrey J Coleman, Almone Porri, John M Peppers, Jinesh D Patel, Michael Betz, Jens Lerchl, J. Scott McElroy

Open Access Article

Field-Evolved $\Delta G210$ -ppo2 from Palmer Amaranth Confers Pre-emergence Tolerance to PPO-Inhibitors in Rice and *Arabidopsis*

by Pamela Carvalho-Moreira¹, Gulab Rangani¹, Ana Claudia Langaro², Vilha Brivastava¹, Almone Porri³, Steven J. Bown⁴, Jens Lerchl³ and Nilda Roma-Burgos¹

¹ Department of Crop, Soil, and Environmental Sciences, University of Arkansas, Fayetteville, AR 72704, USA

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⁴ BASF Agricultural Solutions, Research Triangle Park, NC 27713, USA

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Genes 2022, 13(6), 1044; <https://doi.org/10.3390/genes13061044>

Pest Management Science

Research Article

Functional PPO2 mutations: co-occurrence in one plant or the same ppo2 allele of herbicide-resistant *Amaranthus palmeri* in the US mid-south

Matheus M Nogueira, Gulab Rangani, James Heiser, Taghi Bararpour, Lawrence E Steckel, Michael Betz, Almone Porri, Jens Lerchl, Sophie Zimmermann, Robert L. Nichols, Nilda Roma-Burgos

First published: 29 September 2020 | <https://doi.org/10.1002/ps.6111> | Citations: 15

Pest Management Science

Research Article | Open Access

Inhibition profile of trifludimoxazin towards PPO2 target site mutations

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First published: 30 September 2022 | <https://doi.org/10.1002/ps.7216> | Citations: 4

Almone Porri and Michael Betts contributed equally to this work

Open Access Article

Assessment of Efficacy and Mechanism of Resistance to Soil-Applied PPO Inhibitors in *Amaranthus palmeri*

by Gulab Rangani¹*, Almone Porri², Reiofeli A. Salas-Perez¹, Jens Lerchl², Srikanth Kumar Karaiikal¹, Juan Camilo Velásquez¹ and Nilda Roma-Burgos¹

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Discovery, mode of action, resistance mechanisms, and plan of action for sustainable use of Group 14 herbicides

Published online by Cambridge University Press: 29 March 2023

Abigail L. Barker, John Pawlak, Stephen O. Duke, Roland Beffa, Patrick J. Tranel, Joe Wuerfel, Bryan Young, Almone Porri, Rex Liebl, Raphael Aponte, Douglas Findley, Michael Betz, Jens Lerchl, Stanley Culppepper, Kevin Bradley and Frank E. Dayan

Pest Management Science

Research Article | Open Access

Can double PPO mutations exist in the same allele and are such mutants functional?

Almone Porri, Matheus M Nogueira, Michael Betz, Daniel Sälinger, Frank Brändle, Steven J Bown, Jens Lerchl, Lucie Meyer, Michael Knapp, Nilda Roma-Burgos

First published: 27 February 2022 | <https://doi.org/10.1002/ps.6850> | Citations: 4



Recent Highlights in the Discovery and Optimization of Crop Protection Products
2021, Pages 501-509



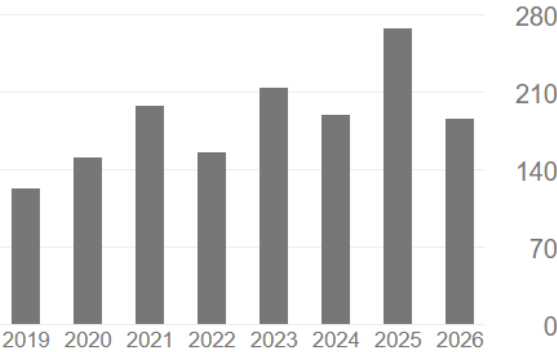
Chapter 34 - Tirexor®—design of a new resistance breaking protoporphyrinogen IX oxidase inhibitor

Matthias Witschel, Raphael Aponte, Gregory Armel, Peter Bowerman, Thomas Mietzner, Trevor Newton, Almone Porri, Anja Simon, Thomas Seitz

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