

Response of *Arabidopsis thaliana* to 22 ALS inhibitors: baseline toxicity and cross-resistance of *csr1-1* and *csr1-2* resistant mutants

F ROUX, A MATEJICEK & X REBOUD

UMR Biologie et Gestion des Adventices, INRA, Dijon, France

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Summary

The baseline toxicity of 22 acetolactate synthase (ALS)-inhibiting herbicides and the cross-resistance patterns of chlorsulfuron- and imazapyr-resistant (*R*) lines on these 22 ALS-inhibiting herbicides were investigated using the model species *Arabidopsis thaliana*. The 22 herbicides consisted of 18 sulfonyleureas (SU), three imidazolinones (IMI) and one triazolopyrimidine (TP). The ED₅₀ values (doses of herbicides required to reduce dry matter by 50%) of the post-emergence-treated Col and Ler susceptible (*S*) lines ranged from 22 to 4822 mg ha⁻¹ and from 17 to 3143 mg ha⁻¹ respectively. The *csr1-1* chlorsulfuron-resistant line (substitution of Pro₁₉₇ to Ser) conferred a high resistance to the only TP tested as well as to nine SU herbicides (*R*:*S* ratio ≥30), a low resistance

to two SU herbicides (*R*:*S* ≥5 and <30) and little or no resistance to the three IMI and seven other SU herbicides (*R*:*S* <5). This result contradicts the expectation that an ALS mutation selected by an SU herbicide confers high cross-resistance to other SU herbicides. We found that the efficacy of specific ALS inhibitors was different for different species and therefore could not be predicted from our results with *A. thaliana*; however, the cross-resistance patterns in *A. thaliana* were highly correlated with cross-resistance patterns in unrelated species with the same resistance mutation. These results have implications for resistance management.

Keywords: acetolactate synthase, acetoxyacid synthase, herbicide resistance, target site, weed control, herbicide mixture.

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Introduction

Acetolactate synthase (ALS), also referred to as acetoxyacid synthase (AHAS), is the first enzyme in the biosynthesis of the branched-chain amino acids isoleucine, valine and leucine. ALS is the target site of the herbicide family known as ALS inhibitors. Although recent studies indicate that baseline toxicity on weed species can vary greatly among ALS inhibitors (Sibony *et al.*, 2001), the ALS inhibitors are effective at low doses and have a favourable environmental profile and thus they are popular herbicides. Indeed, ALS inhibitors control a broad weed spectrum, have low soil residual activity, wide selectivity in some crops and low mammalian toxicities (Mazur & Falco, 1989; Tranel &

Wright, 2002). As a consequence, the number of developed active ingredients and crops in which they are now registered has made these ALS inhibitors the most widely used selective herbicides in the world (Tranel & Wright, 2002).

The intensive use of the ALS inhibitors, together with an apparently high weed mutation rate (Preston & Powles, 2002) and/or a wide range of resistance alleles (Tranel & Wright, 2002) have resulted in an increased occurrence of weed population resistance. Biotypes of 83 weed species worldwide have evolved resistance to these herbicides (Heap, 2004). In most cases, resistance to ALS-inhibiting herbicides is due to an insensitive site of action caused by one of several possible point mutations in the ALS gene (reviewed by Saari *et al.*, 1994; Devine &

Eberlein, 1997; Tranel & Wright, 2002). Non-target-site resistance due to rapid detoxification of the herbicide has also been reported in *Alopecurus myosuroides* Huds. (Menendez *et al.*, 1997; Letouzé & Gasquez, 2003), *Lolium rigidum* Gaudin (Christopher *et al.*, 1991, 1992; Cotterman & Saari, 1992), *Echinochloa phyllopogon* (Stapf.) Koss. (Fisher *et al.*, 2000) and *Sinapis arvensis* L. (Veldhuis *et al.*, 2000). Cinosulfuron and primisulfuron resistance involving gene amplification has so far only been described in an *in vitro* case (Harms *et al.*, 1992). Resistance caused by an altered ALS is usually classified as three types on the basis of cross-resistance (e.g. Tranel & Wright, 2002): (i) sulfonylurea (SU) and triazolopyrimidine (TP) resistant, (ii) imidazolinone (IMI) and pyrimidinylthiobenzoate (PTB) resistant and (iii) SU, IMI, TP and PTB resistant (described as a 'broad' cross-resistance) representing roughly 49, 30 and 21% respectively of recorded cases of field resistance. For the Ala₁₂₂ and Ala₂₀₅ mutation sites in the ALS gene, the cross-resistance patterns are not detailed enough to determine which of the above types they belong to. In addition, depending on the point mutation conferring resistance, a weed population resistant to an ALS inhibitor class could still be controlled by another ALS inhibitor of an appropriate class. When examined in detail, cross-resistance patterns can be far more complex, with sensitivities varying within each chemical subfamily. For example, mutation at Pro₁₉₇ in *Raphanus raphanistrum* L. confers high resistance to chlorsulfuron (SU) and metosulam (TP), but a low resistance to metsulfuron (SU) and no resistance to flumetsulam (TP) (Yu *et al.*, 2003). On a whole-plant basis, sulfometuron-resistant *Amaranthus retroflexus* L. has *R:S* ratios ranging from 6 to 127 for sulfonylureas and from 4 to 63 for imidazolinones (Sibony *et al.*, 2001).

Establishing the relationships among numerous ALS-inhibiting herbicides by studying on the one hand their baseline toxicity and on the other hand the cross-resistance patterns conferred by several mutations within the ALS gene would be informative for weed management. Such information was obtained in this study on *Arabidopsis thaliana* (L.) Heynh. (thale cress). Although *A. thaliana* is not a noxious weed, this model plant species is a self-fertile diploid from which numerous herbicide-resistant mutants have been isolated (Haughn *et al.*, 1988; Mourad *et al.*, 1994; Jander *et al.*, 2003). Susceptible and resistant lines share the common Columbia (Col) genetic background, allowing unambiguous direct comparisons. The work reported here was therefore undertaken (i) to study the relationships among 22 ALS-inhibiting herbicides using two *A. thaliana* susceptible lines and (ii) to assess the cross-resistance pattern of chlorsulfuron- and imazapyr-resistant lines to these 22 ALS-inhibiting herbicides. The baseline toxicity

and cross-resistance patterns in *A. thaliana* were compared with those observed in other weed species.

Materials and methods

Plant material and chemicals

Two susceptible (*S*) and two resistant (*R*) lines of *A. thaliana* were used in our study. The two commonly used Columbia (Col) and Landsberg (Ler) inbred lines were chosen as the susceptible references. We used the *A. thaliana* chlorsulfuron-resistant (*csr1-1* or GH50) and imazapyr-resistant (*csr1-2* or GH90) mutants isolated by Haughn and Somerville (1986, 1990) from ethylmethane-sulfonate (EMS) mutagenized populations of the wild-type susceptible Col line. The *csr1-1* mutant is resistant due to a point mutation resulting in a Pro to Ser substitution at the 197th amino acid, while the *csr1-2* mutant is resistant due to a point mutation resulting in a Ser to Asn substitution at the 653rd amino acid (Haughn *et al.*, 1988; Sathasivan *et al.*, 1990, 1991). All *A. thaliana* lines were provided by the Nottingham Stock Centre (Nottingham, UK).

The 22 ALS-inhibiting herbicides used in this study consisted of 18 SU, three IMI and one TP (Table 1). Of

Table 1 Herbicides applied to *A. thaliana* in experiments to establish dose-response and cross-resistance pattern

Group*	Herbicide	Commercial formulation	a.i. (% wt/wt)
SU	amidosulfuron ¹	Gratil	75
	azimsulfuron ²	Gulliver	50
	bensulfuron-methyl ²	Londax	60
	chlorsulfuron ²	Glean	75
	cinosulfuron ³	Setoff	20
	flazasulfuron ³	Mission	25
	flupyrsulfuron-methyl ²	Lexus	50
	foramsulfuron ¹	–	22.5
	lodosulfuron ¹	–	10
	mesosulfuron ¹	–	75
	metsulfuron-methyl ²	Allié	20
	nicosulfuron ³	Milagro	4
	rimisulfuron ²	Titus	25
	sulfosulfuron ⁴	Monitor	80
	thifensulfuron-methyl ²	Harmony	75
	triasulfuron ³	Longran	20
	tribenuron-methyl ²	Caméo	75
triflusulfuron-methyl ²	Safari	50	
IMI	imazamox ⁵	Raptor	12
	imazapyr ⁵	Arsenal	25
	imazaquin ⁵	Scepter	70
TP	metosulam ⁶	Eclipse	10

*SU, sulfonylurea; IMI, imidazolinone; TP, triazolopyrimidine. ¹Bayer CropSciences (Lyon, France); ²DuPont de Nemours (Paris, France); ³Syngenta Agro (Saint-Cyr, France); ⁴Monsanto (Bron, France); ⁵BASF Agro (Tassin la Demi Lune, France); ⁶Dow AgroSciences (Sophia Antipolis, France).

the 22 herbicides used in this study, 19 were commercial formulations. Foramsulfuron, iodosulfuron and meso-sulfuron, marketed only as mixtures with other active ingredients, were obtained directly from the marketing companies who provided a formulation containing the ALS inhibitor as the single herbicide active ingredient.

Bioassays

Seeds of *A. thaliana* were sown in 1-L plastic pots filled with a commercial soil (Terreau Semis Bouturage Repiquage; Composana, Roche-les-Beaupré, France). They were grown in the greenhouse at 20/25°C (night/day) under natural light supplemented by artificial sodium light to provide a 16-h photoperiod. To avoid micro-environmental effects the pots were regularly rotated during the growing period. The plants were watered twice a week with a standard nutrient solution. The 22 ALS-inhibiting herbicides (Table 1) were applied post-emergence using a laboratory track sprayer delivering 300 L ha⁻¹ spray solution with a 110-04 nozzle operated at 400 kPa. All herbicides were applied to S and R *A. thaliana* plants at the four- to five-leaf stage. Before spraying, plants were thinned to 40 per pot. Two weeks after treatment plants were cut off at soil level and shoots were oven-dried at 70°C for 48 h. An observation corresponded to the dry shoot biomass of 40 plants per pot. The experiment was conducted in a randomized design with three replicates.

Dose–response experiments on susceptible lines

We constructed a geometric series of herbicide concentrations with the aim of encompassing the range zero to normal recommended dose rates. For each susceptible line (and notwithstanding their various field recommended doses), herbicide application rates were: 0.034, 0.103, 0.309, 0.926, 2.778, 8.33 and 25 g a.i. ha⁻¹. In this way, assuming that no major bias was induced by the various chemical formulations, the respective toxicity response of the 22 ALS inhibitor herbicides could be directly assessed. Data were expressed as percentages of their untreated respective controls to standardize comparisons between Col and Ler lines. For each line a non-linear regression was used to describe the response of lines to ALS-inhibiting herbicides. Following Kudsk and Streibig (1993), we used the equation given below and fitted the dose–response curve using SYSTAT (SYSTAT 10, 2000).

$$W_{ij} = C + \frac{(D - C)}{\left(1 + (x_{ij}/ED_{50i})^{b_i}\right)}$$

where W_{ij} denotes the dry matter at the j th dose of herbicide i ; D and C denote the upper and the lower limits of dry

matter at zero and very large doses of herbicide i ; ED_{50i} denotes the dose of herbicide i required to reduce dry matter by 50% between the upper and lower limits; and b_i is proportional to the slope of the curve around ED_{50i} . An F -test ($P = 0.05$) was used to test significant differences of the regression parameters. Bonferroni's correction was applied to adjust the observed significance level for the fact that multiple comparisons were made (Scherrer, 1984). Comparisons of ED_{50} values among herbicides were carried out by examining the overlap between the 95% Wald's confidence limits. Wilcoxon's signed-rank test was then performed to test the effect of the Col or Ler genetic background of the S line on the ED_{50} (Scherrer, 1984).

Cross-resistance pattern

For each herbicide, Col S, homozygote *csr1-1* R and *csr1-2* R *A. thaliana* plants were treated with four doses: ED_{50} (determined as described above), and 3, 30 and 300 times ED_{50} dose. Resistance factors were defined from the ED_{50} values ($ED_{50R}:ED_{50S}$).

Predictability within and among species

Comparisons of ED_{50} and cross-resistance patterns between *A. thaliana* and other weed species were performed by reference to the available data from literature (list used in the legend of Table 3). Only those weed species were included for which a measure of 50% reduction in dry matter, survival, character or growth inhibition (i.e. ED_{50} , I_{50} , EC_{50} or GR_{50}) were determined for at least three ALS-inhibiting herbicides that were also used in this study. Moreover, comparisons of cross-resistance patterns among *A. thaliana* and other weed species involved species for which target-site mutation at Pro₁₉₇ or Ser₆₅₃ amino acids (based on *A. thaliana* numbering) were clearly identified. Spearman's correlations insensitive to scale dependence (Scherrer, 1984) were calculated to assess the predictability of ED_{50} or cross-resistance patterns between *A. thaliana* and each of the other weed species.

Results and discussion

Baseline toxicity

For each susceptible line the herbicide application rates were sufficient to establish the dose–response curve except for amidosulfuron, imazamox and imazaquin. For these three herbicides a new geometric series of herbicide concentrations (0.309, 0.926, 2.778, 8.33, 25, 75 and 225 g a.i. ha⁻¹) was applied to both Col and Ler susceptible lines. Except for imazamox in the Ler susceptible line, a common slope could be accepted

for all response curves. Hence, ED₅₀ was used to characterize the baseline toxicity of the ALS-inhibiting herbicides studied for *A. thaliana*. The dose–response experiments revealed differences in the reaction of the susceptible lines to the tested herbicides (Table 2). The ED₅₀ values of the Col and Ler susceptible lines ranged from 22 to 4822 mg ha⁻¹ and from 17 to 3143 mg ha⁻¹ respectively. Wilcoxon's signed-rank test on the mean ED₅₀ indicated no effect of the genetic background on the susceptibility of the lines ($z_t = 0.381$, $P = 0.703$). For each *A. thaliana* susceptible line herbicides were ranked according to their ED₅₀. Highly significant Spearman's correlation between Col and Ler lines ($r_s = 0.904$, $P < 0.001$) indicated that the rank observed for one susceptible line correctly predicted the rank observed for the other susceptible line. ALS-inhibiting herbicides were more toxic to *A. thaliana* than to *A. retroflexus* (Table 3). Mean ED₅₀ value was indeed higher for susceptible *A. retroflexus* biotypes than for both Col and Ler lines (Wilcoxon's signed-rank test; Col: $z_t = 2.845$, $P = 0.004$; Ler: $z_t = 2.667$, $P = 0.008$). Triasulfuron was the most toxic herbicide to both *A. thaliana* susceptible lines, while metsulfuron-methyl and chlorsulfuron were the most effective on *A. retroflexus* and *Chlamydomonas reinhardtii* Dangeard

respectively (Table 3). However, nicosulfuron, rimsulfuron and thifensulfuron remained more effective than amidosulfuron or triflurosulfuron for both the susceptible lines of *A. thaliana* and *A. retroflexus*.

Data from 14 species were considered to be suitable for the study of the relationships between ED₅₀ for *A. thaliana* and other weed species (see Table 3). Six SU herbicides (flazasulfuron, flupyrsulfuron, foramsulfuron, iodosulfuron, mesosulfuron and sulfosulfuron) were not included in the comparison because they had not been involved in studies where at least three ALS-inhibiting herbicides were common to those used in this study. Spearman's correlations cover a large range of values from 1 for *A. thaliana*–*Amaranthus blitoides* S. Watson and *A. thaliana*–*Salsola iberica* Sennen & Pau pairs (but on four and three herbicides in common respectively), to –1 for the *A. thaliana*–*Monochoria korsakowii* Regel & Maack pair (Table 4). Mean correlation is 0.06, a non-significant overall value. The toxicity ranking of an ALS inhibitor herbicide is thus similar for two susceptible lines of the same species (*A. thaliana* in our study) but the toxicity ranking observed in the other species cannot be predicted. However, the possible comparisons involved few herbicides and thus statistical power was lacking. Although absorption and translocation of

Table 2 ED₅₀ for the Col and Ler susceptible lines and resistance ratios (R:S) for the chlorsulfuron-resistant *csr1-1* and imazapyr-resistant *csr1-2* lines of *Arabidopsis thaliana* treated with 22 ALS-inhibiting herbicides

Chemical class	Herbicide	<i>Arabidopsis thaliana</i>				<i>csr1-1</i> R:S	<i>csr1-2</i> R:S
		Col		Ler			
		ED ₅₀ (mg ha ⁻¹)	CL* (mg ha ⁻¹)	ED ₅₀ (mg ha ⁻¹)	CL* (mg ha ⁻¹)		
SU	amidosulfuron	2947	1164–4729	387	71–704	555	1
	azimsulfuron	138	88–188	384	112–655	419	1
	bensulfuron-methyl	154	76–233	502	174–830	490	5
	chlorsulfuron	65	34–96	144	51–238	549	1
	cinosulfuron	217	104–330	458	237–680	275	3
	flazasulfuron	325	169–481	1232	420–2044	2	1
	flupyrsulfuron-methyl	311	184–438	732	298–1165	3	1
	foramsulfuron	333	95–570	506	291–722	2	1
	iodosulfuron	132	84–179	287	75–499	11	1
	mesosulfuron	376	175–577	1507	579–2436	2	1
	metsulfuron-methyl	210	109–311	392	234–1149	2	1
	nicosulfuron	106	49–162	119	58–180	3	5
	rimsulfuron	71	36–106	49	18–80	1	1
	sulfosulfuron	81	46–115	61	32–90	119	1
	thifensulfuron-methyl	30	13–48	20	10–31	191	2
triasulfuron	22	9–35	17	6–28	11	1	
tribenuron-methyl	62	31–92	34	15–52	738	6	
triflurosulfuron-methyl	886	423–1349	660	363–958	158	1	
IMI	imazamox	4822	1608–8036	3143	1020–5266	1	151
	imazapyr	212	109–316	218	104–332	3	124
	imazaquin	3687	1287–6086	1740	309–3172	1	97
TP	metosulam	268	116–420	298	96–499	32	1

*CL: 95% Wald confidence limits.

R, resistant; S, susceptible.

Table 4 Spearman's correlation between *A. thaliana* and other weed species for ED₅₀ and resistance ratio (*R*:*S*)

Species compared with <i>A. thaliana</i>	Spearman's correlation		
	ED ₅₀ †	<i>R</i> : <i>S</i>	Mutation
<i>Amaranthus blitoides</i> S. & W.	1*	–	–
<i>Amaranthus retroflexus</i> L.	0.067	0.432*	Pro ₁₉₇ to Leu
<i>Chlamydomonas reinhardtii</i> Dangeard	0.5	–	–
<i>Chlorella vulgaris</i> Beijerinck	–0.5	–	–
<i>Conyza albida</i> Willd.	0.671*	–	–
<i>Cyperus difformis</i> L.	0.5	–	–
<i>Galium spurium</i> L.	–0.6*	–	–
<i>Lolium perenne</i> L.	–0.5	–	–
<i>Monochoria korsakowii</i> Regel & Maack	–1*	–	–
<i>Monochoria vaginalis</i> Presl	0.5	–	–
<i>Raphanus raphanistrum</i> L.	–0.2	0.5*	Pro ₁₉₇ to His
<i>Raphanus raphanistrum</i> L.	–0.2	0.5*	Pro ₁₉₇ to Thr
<i>Raphanus raphanistrum</i> L.	–0.2	0.5*	Pro ₁₉₇ to Ser
<i>Salsola iberica</i> Sennen & Pau	1*	–	–
<i>Stellaria media</i> (L.) Vill. I ₅₀	–0.5	–	–
<i>Stellaria media</i> (L.) Vill. GR ₅₀	0.5	–	–
Mean (SE)	0.06 (0.153)	0.48 (0.017)	

*Significant values of Spearman's correlation at $P < 0.05$.

†The ED₅₀ of the Col susceptible line was used to calculate Spearman's correlations.

ALS-inhibiting herbicides are said to differ among species (Ackley *et al.*, 1999; Gallaher *et al.*, 1999), this absence of conserved ranking between species could be explained in part by the varying abilities of species to conduct detoxification. As an illustration of this variation among species, *A. thaliana* and rice (*Oryza sativa* L.) genomes are estimated to contain as many as 286 and 455 different cytochrome P450 genes respectively (Nelson, 1999, 2004; Xu *et al.*, 2001). Although only a portion of the cytochrome P450 genes may be involved in herbicide detoxification, a change in their expression levels may be expected to be encountered when comparing other phylogenetically distant species (as for instance among weed species). Therefore, the ranking of ED₅₀ values cannot be extended from a single species to other unrelated ones.

Cross-resistance

A cross-resistance pattern could be directly assessed by the inhibition of ALS enzyme activity. Here, the cross-resistance pattern on the 22 ALS-inhibiting herbicides used in our study was assessed for the homozygous chlorsulfuron- and imazapyr-resistant lines by recording plant dry matter. The resistance ratios for the *csr1-1* and *csr1-2* lines are indicated in Table 2. The *csr1-2* imazapyr-resistant line conferred high resistance to the three IMI herbicides (*R*:*S* ratio ≥ 30), low resistance to three SU herbicides (*R*:*S* ratio ≥ 5) and little or no resistance to the TP and 15 other SU herbicides (*R*:*S* ratio < 5). The cross-resistance pattern caused by the substitution of Ser₆₅₃ to Asn was in agreement with the expectation that this ALS mutation selected by an IMI herbicide confers

high cross-resistance to other IMI herbicides but not to SU and TP herbicides (Tranel & Wright, 2002). The *csr1-1* chlorsulfuron-resistant line exhibited high resistance to the only TP tested as well as to nine SU herbicides (*R*:*S* ratio ≥ 30), low resistance to two SU herbicides (*R*:*S* ratio ≥ 5) and little or no resistance to the three IMI and seven SU herbicides (*R*:*S* ratio < 5). The cross-resistance pattern caused by the substitution of Pro₁₉₇ to Ser was in agreement with the expectation that this ALS mutation selected by an SU herbicide conferred high cross-resistance to TP herbicides but not to IMI herbicides. We have also demonstrated that the substitution of Pro₁₉₇ to Ser in *A. thaliana* did not confer a high resistance to the whole group of SU herbicides. This result contradicts the assumption that resistance caused by an altered ALS could be classified into three types on the basis of cross-resistance (e.g. Tranel & Wright, 2002): (i) sulfonylurea (SU) and triazolopyrimidine (TP) resistance, (ii) imidazolinone (IMI) and pyrimidinylthiobenzoate (PTB) resistance and (iii) SU, IMI, TP and PTB resistance (described as a 'broad' cross-resistance). Studying numerous herbicides per ALS inhibitor class is thus necessary to assess correctly the cross-resistance pattern of a mutation. In our case, including more ALS-inhibiting TP herbicides could be expected to yield even wider and more reliable results.

The observed cross-resistance pattern and resistance ratios in *A. thaliana* are consistent with those observed in *A. retroflexus* and *R. raphanistrum* (Table 3), with Spearman's correlation values ranging from 0.432 to 0.5 (Table 4). The cross-resistance pattern found in *A. thaliana* could thus be extended to other resistant weed species with a Pro₁₉₇ substitution. This incomplete

cross-resistance within a chemical family has two consequences for the management of weed species. First, a weed species resistant to a specific SU herbicide could still be managed by other herbicides from the same chemical class. As an example, *R. raphanistrum* chlorsulfuron-resistant populations (Yu *et al.*, 2003) may be controlled by up to seven other SU herbicides depending on crop selectivity. Second, mixtures of two ALS-inhibiting herbicides to which resistance is conferred by different target-site mutations may diminish the frequency of mutants resistant to both active ingredients. With appropriate dosage only those plants having the two target-site mutations (or a rare non-target site resistance mechanism) would be selected in fields, therefore delaying the appearance and propagation of the resistance. However, a target-site mutation conferring a broad cross-resistance, i.e. substitution of Trp₅₇₄ to Leu, could also be selected by such herbicide mixtures. This highlights the importance of extending the analysis to all isolated mutants in order to design the best herbicide combination, i.e. the one that demonstrates the lowest overlap between *R:S* ratios. It is disappointing that, so far, the cross-resistance pattern of ALS mutations is usually limited to a few SU herbicides.

Predictability within and among species

Focusing on the only case species for which both ED₅₀ and *R:S* ratios were available (Table 4), we found that the Spearman's correlations for *R:S* ratios were significantly higher than the Spearman's correlations for ED₅₀ (Friedman test statistic = 4, $P = 0.046$). This result indicates that the toxicity ranking seems to be of much less predictive power outside the species range than the cross-resistance pattern. The cross-resistance pattern seems to remain significant even when comparing more phylogenetically distant species. However, this result would greatly benefit from extended cross-resistance analysis of the available material, the molecular characterization of well-documented herbicide resistance, or the search for new mutants in a well known model organism. Studies on the new ALS-inhibiting herbicide resistant lines in *A. thaliana* (Jander *et al.*, 2003) would fall into this category. The work presented here shows how information from studies based on laboratory mutants such as *A. thaliana* can help with management strategies aimed at containing the development of resistant weed species in the fields.

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