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Review

Agronomic and environmental implications of enhanced *s*-triazine degradation

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Abstract

Novel catabolic pathways enabling rapid detoxification of *s*-triazine herbicides have been elucidated and detected at a growing number of locations. The genes responsible for *s*-triazine mineralization, i.e. *atzABCDEF* and *trzNDF*, occur in at least four bacterial phyla and are implicated in the development of enhanced degradation in agricultural soils from all continents except Antarctica. Enhanced degradation occurs in at least nine crops and six crop rotation systems that rely on *s*-triazine herbicides for weed control, and, with the exception of acidic soil conditions and *s*-triazine application frequency, adaptation of the microbial population is independent of soil physiochemical properties and cultural management practices. From an agronomic perspective, residual weed control could be reduced tenfold in *s*-triazine-adapted relative to non-adapted soils. From an environmental standpoint, the off-site loss of total *s*-triazine residues could be overestimated 13-fold in adapted soils if altered persistence estimates and metabolic pathways are not reflected in fate and transport models. Empirical models requiring soil pH and *s*-triazine use history as input parameters predict atrazine persistence more accurately than historical estimates, thereby allowing practitioners to adjust weed control strategies and model input values when warranted.
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Keywords: enhanced biodegradation; modeling; leaching; weed control; pesticide

1 INTRODUCTION

It is arguable that no highly substituted *s*-triazine compound had been released into the biosphere until the commercial launch of simazine in 1958; thus, it was deemed unlikely that microbial populations would possess the enzymatic ability rapidly to degrade these xenobiotic compounds.[1,2] *s*-Triazine persistence data collected over subsequent decades supported this assumption, and consequently these compounds have been considered recalcitrant. Current weed control programs and pesticide transport models function under this historic paradigm, that is, *s*-triazine herbicides are persistent, provide season-long residual weed control and are susceptible to off-site transport. Howerver, recent data indicate that bacterial adaptations have occurred, thereby enabling rapid mineralization of highly substituted *s*-triazine herbicides. Reviews focusing on the evolutionary significance of these metabolic pathways exist,[3,4] but the scope of this manuscript is to expound on the agronomic and environmental implications of these bacterial adaptations. Specifically, the intention is to:

- characterize the pathways enabling *s*-triazine mineralization;
- describe the agronomic and environmental significance of these pathways;
- delineate the physiochemical properties of soils harboring microbial populations able rapidly to degrade *s*-triazine herbicides;
- project where adaptation may occur based on global soil data;

- determine if cultural practices, that is, herbicide use history, cropping history, residue management practices, and N fertility, affect adaptation;
- identify management practices for improving weed control with *s*-triazine herbicides in affected areas;
- propose input values for modeling atrazine fate, transport and risk assessment;
- describe a multiple linear regression model that predicts atrazine persistence in soil at the global scale.

2 MATERIALS AND METHODS

2.1 Bacterial phylogeny and taxonomy

Bacterial species reported to contain atrazine-metabolizing genes, i.e. *atzABCDEF* and *trzNDF*, or unidentified genes that function like

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atzA or *trzN*, were used to query 16S ribosomal sequences from the Ribosomal Database Project (RDP).[5] When available, a type species was used to represent a reported species; otherwise, the most similar species was used. In cases where the reported species has been renamed or reassigned, the synonyms listed with the National Center for Biotechnology Information GenBank database were used to identify currently accepted taxonomy. In cases where an atrazine-degrading bacterium was not identified at the species level, a type species for that genus was used in its place. These 16S sequences were collected, and a dendogram with bootstrap values was constructed using RDP 10.10 Tree Builder.

2.2 Harmonized world soil database

The harmonized world soil database (HWSD) v.1.1 was used to identify surface soils with sand, silt, clay, organic carbon and pH between the tenth and ninetieth percentile and maximum and minimum values of known *s*-triazine-adapted soils.[6] The HWSD-viewer was used to visualize where these soils occur, and an ARC-GIS shape file containing the latitude and longitude of known adapted soils was overlaid on the HWSD-viewer.

2.3 LEACHM simulations

Two one-dimensional leaching estimation and chemistry models (LEACHMs) were constructed to compare the potential for atrazine and its metabolites to leach through *s*-triazine-adapted and non-adapted soils.[7,8] For the non-adapted simulation, the half-lives for atrazine (60 days), desethylatrazine (DEA) (52 days), deisopropylatrazine (DIA) (36 days) and hydroxyatrazine (HA) (60 days) were assumed to be constant for 0–30 cm depth.[9,10] Persistence estimates increased linearly from 30 to 100 cm depth, with maximum half-life values at the bottom of the root zone threefold higher than that in the upper horizon: 180 days for atrazine, 156 days for DEA, 108 days for DIA and 180 days for HA.[10–12] The percentage of atrazine converted to DEA, DIA and HA was set to reflect historic trends, that is, 72% for DEA, 18% for DIA and 10% for HA.[12] For adapted soils, the half-life for atrazine (6 days), DEA (10 days), DIA (8 days) and HA (6 days) was reduced relative to non-adapted soils and assumed to be constant for 0–30 cm depth.[9–11] Again, persistence estimates increased linearly from 30 to 100 cm depth, but the maximum half-life value at the bottom of the root zone was 11-fold higher than that in the upper horizon: 66 days for atrazine, 110 days for DEA, 88 days for DIA and 66 days for HA.[9–10,13] The proportion of atrazine converted to daughter products was set to reflect alterations in the historic metabolic pathway, that is, 22% for DEA, 7% for DIA and 71% for HA.[9,14]

Ten-year simulations (1995–2004) for the top meter of unsaturated silty loams beneath a field in the Morgan Creek, Maryland, watershed subjected to annual rotations of corn (*Zea mays* L.) and soybean [*Glycine max* (L.) Merr.] were constructed for *s*-triazine-adapted and non-adapted soils. The simulation includes five biennial applications of 2.08 kg ha^{-1} atrazine in the spring, preceding the planting of corn.[15] Physical and chemical properties of atrazine and its metabolites are those described previously.[16]

2.4 Multiple linear regression analysis

2.4.1 Initial data survey from the literature

Atrazine dissipation was assumed to follow first-order kinetics. Reported and derived atrazine half-life values were normalized to 20 °C using a Q_{10} of 2.2. Soils were identified as adapted by one of four methods: (1) *s*-triazine half-life value <30 days;

(2) mineralization assay;[12] (3) dissipation assay;[17] or (4) detection of *atzABCDEF* or *trzNDF* genes. The physiochemical range of *s*-triazine-adapted soils was subsequently identified by constructing box plots for the percentage sand, silt, clay and organic carbon and pH using Sigma plot 10.0. Soils identified as potential outliers via the box-plotting technique, that is, values outside the 10 and 90 percentiles, were omitted from the dataset. Using these criteria, 98 soils with known atrazine use histories and persistence estimates were selected for model development.

2.4.2 Model development

Multiple linear regression was used to determine the importance of soil physiochemical parameters and herbicide use history in predicting atrazine persistence in soil (SAS Proc ReG, stepwise option). Parameters evaluated included percentage sand, silt, clay and organic carbon, soil pH (1:1 aqueous soil solution paste), consecutive years of atrazine applications, ranging from 0 (no applications) to 5 (five consecutive applications), and atrazine use history in the last five years, where soils receiving an atrazine application = 1 and soils not receiving an atrazine application = 2.

3 RESULTS

3.1 Historic *s*-triazine dissipation pathways

s-Triazine dissipation in soil has been the focus of many studies over the last 50 years because of its widespread use and frequent detection in surface and groundwater.[4,18] The existing paradigm on the rate and path of *s*-triazine dissipation is based almost exclusively on work conducted prior to 1993. Thus, many weed scientists assume that *s*-triazine herbicides dissipate in the soil via either non-biological, chemical hydrolysis or biologically mediated *n*-dealklyation reactions, and that these compounds are more persistent as soil pH increases (Fig. 1).

3.1.1 *s-Triazine dissipation prior to 1993*

The basic assumption prior to the 1990s was that the halogen, methylthioether and *N*-alkyl substitutions on the *s*-triazine ring impede the ability of soil microorganisms to metabolize most herbicidal *s*-triazines.[18] Hence, atrazine and other *s*-triazines were considered to be poorly biodegradable in the soil, as was supported by early research. For example, Cook[1] was able to isolate soil bacteria that could grow on cyanuric acid and related *s*-triazines, but these microbes were not able to grow on *s*-triazine herbicides.

The proposed pathway of *s*-triazine degradation prior to the discovery of microbes that could rapidly degrade these herbicides is reported in Fig. 1. There were two branches in this pathway: (1) a chemical hydrolysis pathway that resulted in the formation of hydroxylated products, and (2) a biological system that *N*-dealkylated the side chains. The biological pathway does not detoxify the herbicides because the metabolites are still phytotoxic, so it was proposed that the primary method for detoxifying *s*-triazines in the soil was through chemical hydrolysis.[19,20]

3.1.2 *Chemical hydrolysis of s-triazines in the soil*

The hydroxyl analogs of atrazine, simazine and propazine can be produced in strongly acid or basic solutions via chemical hydrolysis.[21] Early researchers studying the route of *s*-triazine herbicide dissipation in soil found the hydroxylated metabolites of atrazine, propazine and simazine.[22–25] The formation of these metabolites occurred more rapidly in the presence of soil than
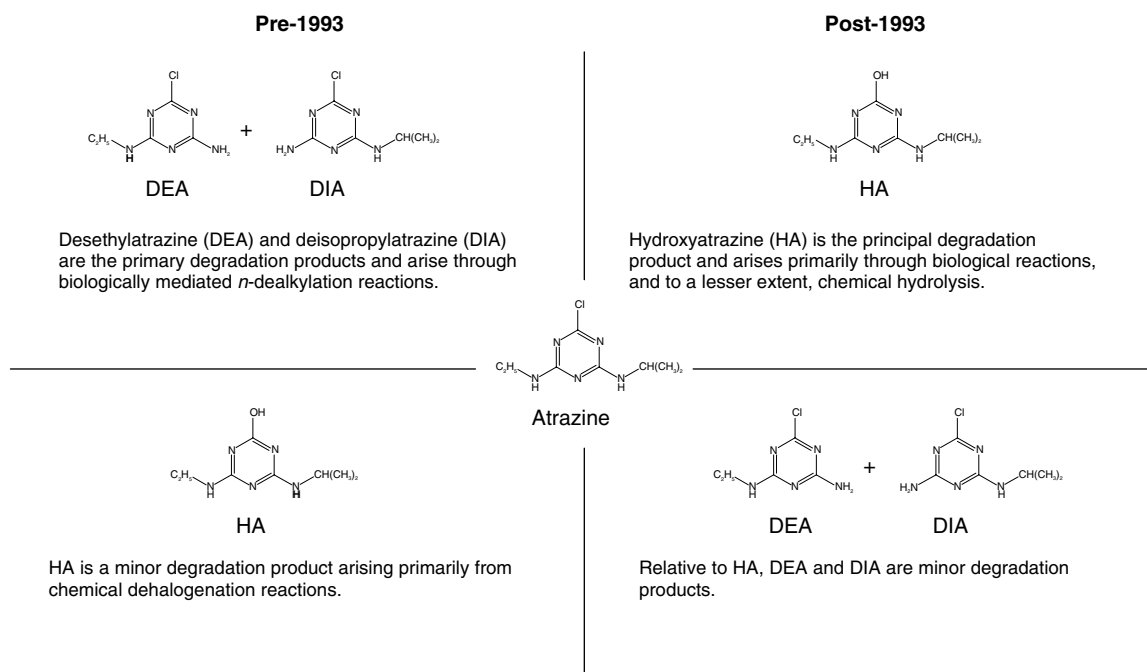
Pre-1993

Post-1993

DEA   +   DIA

HA

Desethylatrazine (DEA) and deisopropylatrazine (DIA) are the primary degradation products and arise through biologically mediated *n*-dealkylation reactions.

Hydroxyatrazine (HA) is the principal degradation product and arises primarily through biological reactions, and to a lesser extent, chemical hydrolysis.

Atrazine

HA

DEA   +   DIA

HA is a minor degradation product arising primarily from chemical dehalogenation reactions.

Relative to HA, DEA and DIA are minor degradation products.

Figure 1. Degradation pathway for atrazine in soil, pre- and post-1993. Adapted from Wackett *et al*[4].

in an aqueous solution.[22] Treating soil with either 200 ppm of sodium azide or heat (95 °C) did not decrease the formation of the hydroxylated metabolites of atrazine, simazine and propazine, thus supporting the assumption that this process was not biologically mediated.[23]

Russell *et al*.[26] demonstrated how montmorillonite clay could catalyze the formation of hydroxytriazines from atrazine and propazine. The hydrolysis occurs in three steps. First, the chloro-*s*-triazine is sorbed to a soil surface through hydrophobic interactions of the alkyl side chain. Then a proton is transferred to one of the ring N atoms. This protonation facilitates nucleophilic attack on the C atom in the 2-position, resulting in the formation of the hydroxylated metabolite.[27]

The rate of abiotic hydrolysis is dependent on pH, with greater hydrolysis as pH decreases. Research prior to the 1990s showed that the persistence of the triazines was the shortest in soils with pH less than 6 and greatest in soils with pH greater than 7. The conclusion from these results was that the primary method of detoxification of the *s*-triazines was chemical hydrolysis, as the hydroxylated metabolites are tightly bound to soil and are non-phytotoxic.[24,28]

3.1.3 Biological degradation of s-triazines

Initial work on the metabolism of ^{14}C ring-labeled atrazine in the soil showed minimal or no release of $^{14}CO_2$ from the soil (Fig. 2).[24,29] The rate of $^{14}CO_2$ release was related to the level of organic matter in the soil and general microbial activity.[30] Researchers at the time concluded that ring cleavage of the *s*-triazine played little or no role in the dissipation of the herbicide.

Sirons *et al*.[20] found that atrazine was microbially converted into de-ethylated atrazine (DEA) as a major metabolite and into a de-isopropylated metabolite (DIA) as a minor metabolite. Krutz *et al*.[11] also found that there was more DEA formed compared with DIA in non-adapted soils from Colorado and Mississippi. DEA and DIA bind loosely to soil, and DEA has been frequently
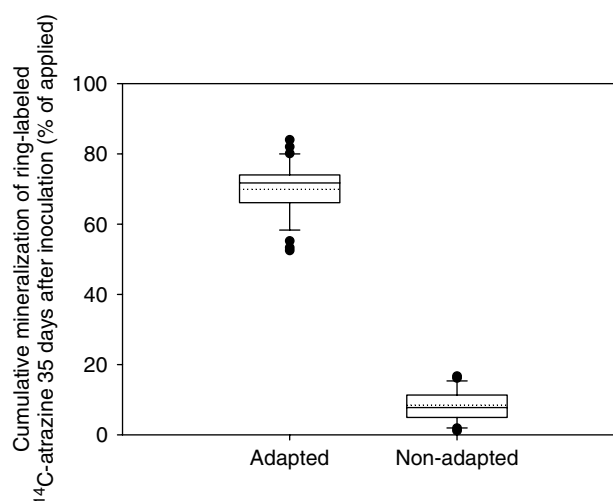
Figure 2. Box plot comparisons of published data for cumulative $^{14}CO_2$ evolution of ^{14}C-ring-labeled atrazine 35 days after inoculation in *s*-triazine-adapted and non-adapted soil. Aadapted from Krutz *et al*.[12] Boundary of box closest to zero indicates the 25th percentile, a solid line within the box marks the median, a dashed line within the box delineates the mean and the boundary of the box furthest from zero indicates the 75th percentile. Error bars above and below the box indicate the 90th and 10th percentile, and solid dots indicate outliers. The number of independent observations is 22 for non-adapted soils and 54 for *s*-triazine-adapted soils.[75,85,107–110,121,122,125,154–157].

found in soil water at greater depths and at higher concentrations in groundwater than atrazine or DIA.[31] Early studies showed that more $^{14}CO_2$ was released when the radiolabel was on the aminoethyl substituent than when it was in the ring or on the aminoisopropyl substituent.[21] However, *N*-dealkylation was not considered to be a detoxification step, as DEA and DIA are still phytotoxic. It was proposed that part of the long-term activity of atrazine was due to the formation of these metabolites.[20] Field
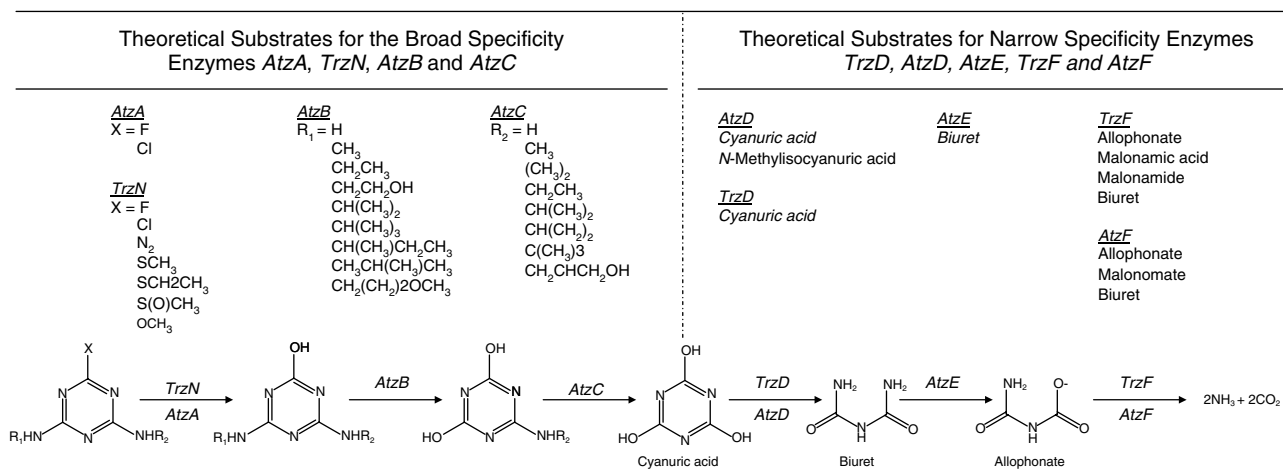
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Figure 3. *s*-Triazine metabolism by soilborne bacteria with *atzABCEEF* and *trzNDF* gene homologs. Adapted from Shapir *et al*[3].

work from the 1970s to the 1990s supported this interpretation, as atrazine and other *s*-triazines were shown to have very long residual activity, with half-lives ranging from 28 to 178 days, depending on soil and rate applied.[32–35] Hiltbold and Buchanan[36] estimated that atrazine persistence increased from 9 to 29 days with each unit increase in pH, depending on soil. They also reported that atrazine degraded more rapidly in acid soils compared with basic soils, but that microbes played a greater role in degradation as pH increased.[34,36] Based on these observations, the average atrazine half-life was estimated to be 60 days.[13]

The authors submit that the dissipation pathway in Fig. 1 is accurate for soils that do not have an *s*-triazine application history or where *s*-triazine herbicides are rarely applied. For example, Krutz *et al.*[11] compared atrazine metabolism in non-adapted and adapted soils from Colorado and Mississippi. They found that in soils with no history of atrazine use the half-life was between 32 and 128 days, depending on temperature and moisture. Conversely, in soils with an *s*-triazine application history the atrazine half-life values varied from 1 to 12 days. Moreover, the pattern of metabolite formation in non-adapted soils was similar to that reported by earlier researchers, with an accumulation of DEA, DIA and HA. Thus, in non-adapted soils, the widely accepted paradigm on *s*-triazine dissipation in soil is probably correct. However, in soils harboring microbial populations able rapidly to mineralize *s*-triazine herbicides, the historic pathway is likely overshadowed by a new, biologically mediated dehalogenation pathway.

3.2 *atzABCDEF* and *trzNDF* homologs
3.2.1 Novel s-triazine catabolic pathways
The discovery of two bacterial isolates, *Pseudomonas* sp. ADP and *Nocardioides* sp. *C190*, changed current understanding of atrazine's fate in the environment. *Pseudomonas* sp. ADP was isolated from an atrazine-contaminated soil collected from an agricultural chemical dealership in Little Falls, Minnesota, USA,[37–39] and *Nocardioides* sp. *C190* was isolated from a Canadian agricultural soil with a prior atrazine use history.[40,41] The isolation of *Pseudomonas* sp. ADP was unprecedented in that the bacterium mineralized atrazine, a feat deemed unlikely by microbiologists from 1958 until the bacterium's isolation in 1994. Conversely, *Nocardioides* sp. *C190* could not mineralize atrazine but degraded a broader range of *s*-triazine herbicides than did *Pseudomonas* sp. ADP.[40,41] In the succeeding years, the underlying principles regulating *s*-triazine

catabolism by these bacteria were elucidated. Herein, a review is given of the bacterial genes *atzABCDEF* and *trzNDF* and the corresponding enzymes *AtzABCDEF* and *TrzNDF* responsible for rapid *s*-triazine mineralization in soil.

The *atzABCDEF* and *trzNDF* genes code for unique enzymes that catabolize diverse *s*-triazine herbicides (Fig. 3). In this pathway, atrazine degradation is initiated by either *AtzA* or *TrzN*, Fe(II) and Zn(II) metalloenzymes respectively, which dehalogenate atrazine, yielding hydroxyatrazine.[41–46] *AtzA* and *TrzN* share less than 27% sequence identity and have different substrate specificities.[42,43,45] *AtzA*'s substrate range includes six chlorotriazine herbicides, i.e. atrazine, simazine, propazine, terbuthylazine, mesoprazine and sebuthylazine, and their mono-*n*-dealkylated metabolites. Conversely, *TrzN*'s substrate range encompasses the six chlorotriazines above, in conjunction with six methoxytriazines, i.e. atraton, methometon, prometon, secbumeton, simeton and terbumeton, four methylthiotriazines, i.e. ametryn, desmetryn, methoprotyne and terbutryn, the mono-*N*-dealkylated metabolites of the preceding chloro, methoxy and methylthiotriazines herbicides and a few pyrimidine compounds. The succeeding enzyme in the pathway, hydroxyatrazine *N*-ethylaminohydrolase (*AtzB*), is a Zn(II) metalloenzyme that hydrolytically converts hydroxyatrazine to *N*-isopropylammelide.[47] *AtzB*'s substrate range includes the preceding hydroxylated chloro, methoxy and methylthiotriazine herbicides and their corresponding hydroxy-mono-*N*-dealkylated metabolites.[48] The third enzyme in the pathway, *N*-isopropylaminohydrolase (*AtzC*), is a Zn(II) metalloenzyme that converts *N*-isopropylammelide and related *s*-triazine metabolites to cyanuric acid.[49,50] One of two cyanuric acid hydrolases, *AtzD* or *TrzD*, then cleaves the *s*-triazine ring, yielding biuret.[51–53] Biuret is converted to allophanate via *AtzE*, at which point one of two enzymes, *AtzF* or *TrzF*, hydrolyze allophanate, yielding 2 mol of ammonia and carbon dioxide respectively.[54–57] Relative to the upstream genes in this pathway, *AtzDEF* and *TrzDF*'s substrate range is comparatively restricted.[44,52–55,57]

3.2.2 Biological and spatial distribution of the s-triazine catabolism genes
The *atzABCDEF* and *trzNDF* genes occur in diverse phyla and are highly conserved and globally dispersed (Fig. 4; Table 1). Specifically, bacterial isolates from four phyla originating from six
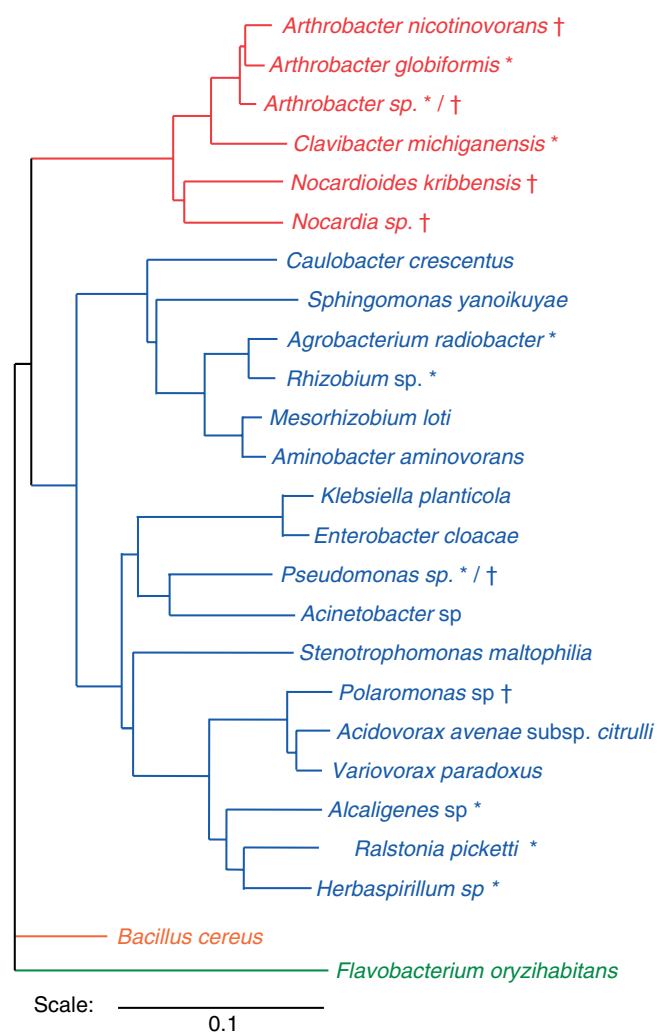
www.interscience.wiley.com/journal/ps · This article is a US Government work and is in the public domain in the USA · *Pest Manag Sci* 2010; **66**: 461–481

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Figure 4. Phylogenetic relationship of bacterial species reported to contain atrazine-metabolizing genes, i.e. *atzABCDEF* or *trzNDF*. When available, a type species was used to represent a reported species; otherwise, the most similar species was used. In cases where the reported species has been renamed or reassigned, the synonyms listed with the National Center for Biotechnology Information GenBank database were used to identify currently accepted taxonomy. In cases where an atrazine-degrading bacterium was not identified at the species level, a type species for that genus was used in its place. Red, blue, orange and green denotes Actinobacteria, Proteobacteria, Firmicutes and Bacteroidetes phyla respectively. The symbols * and † denote detection of *atzA* and *trzN* gene respectively.

continents bear homologs of *s*-triazine catabolism genes with high sequence identity to those of *Pseudomonas* sp. ADP or *Arthrobacter aurescens* TC1. With noted exceptions,[58–61] most isolates contain some or all of the *atzABCDEF* and *trzNDF* homologs on one or more self-transmittable plasmids.[55,59,60,62] The prevailing explanation for the detection of *atzABCDEF* and *trzNDF* homologs in diverse phyla, therefore, is horizontal gene transfer arising primarily from plasmid conjugation[40,55,59–61,63–67] and, to a lesser extent, transduction.[67,68] The highly conserved sequence identity among *atzABC* and *trzN* homologs is consistent with a recent, single origin with subsequent global dispersal.[59,69,70] Intercontinental transport of *atzABCDEF* and *trzNDF* genes is undocumented, but airborne microbial communities are colonized by soil bacteria which remain viable after transatlantic atmospheric deposition.[71,72]

3.3 Agronomic implications of enhanced degradation

3.3.1 Reduced residual weed control

Detecting *atzABCDEF* and *trzNDF* gene homologs in bacterial isolates spanning diverse phyla and continents indicates potential for widespread enhanced *s*-triazine degradation. Enhanced degradation is the phenomenon whereby a soil-applied pesticide is rapidly degraded by a population of microorganisms that have developed the ability to use the compound as a carbon, energy and (or) nutrient source because of previous exposure to it or an analog. Agronomic implications of enhanced degradation, namely reduced residual weed control, are compounded when structurally similar pesticides also degrade rapidly in adapted soils, a phenomenon referred to as cross-adaptation. As the substrate range of this catabolic pathway encompasses most commercially available *s*-triazine herbicides, cross-adaptation among compounds in this herbicide class is also likely (Fig. 3). In the following section, the agronomic implications of enhanced *s*-triazine degradation are discussed, the geographic distribution of adapted soils is described, basic soil physiochemical properties associated with enhanced degradation are delineated, the impact of agronomic practices on enhanced degradation is determined and, finally, strategies to curtail adverse agronomic effects attributed to this phenomenon are proposed.

Reduced residual weed control is the primary agronomic concern for soils exhibiting enhanced degradation. Residual weed control depends on the herbicide concentration required for efficacy and the herbicide's persistence in soil, which are pest and soil specific.[73] Atrazine half-life values in *s*-triazine-adapted soil are approximately tenfold lower than historic estimates, 60 days (Fig. 5); thus, for a given soil–pest complex, residual weed control may be tenfold lower in *s*-triazine-adapted soil than in non-adapted soil. For example, if an atrazine concentration of 0.4 mg kg^{-1} is required to achieve ≥85% control of pitted morningglory in a Dundee silt loam, then acceptable residual weed control would last 80 days in a non-adapted soil but only 8 days in an *s*-triazine-adapted soil (Fig. 6).

Enhanced degradation can reduce atrazine's residual control of sensitive weed species.[12,74–77] Under laboratory and greenhouse conditions, *Nasturtium officinale* R.Br. and *Solanum nigrum* L. died 15 days after sowing in soil containing atrazine incorporated at 4 mg kg^{-1}.[77] Inoculating this same soil with atrazine-degrading bacteria prior to incorporating atrazine at 4 mg kg^{-1} resulted in normal plant growth.[77] Under greenhouse conditions, the application of atrazine at 1.82 kg ha^{-1} to an *s*-triazine-adapted Dundee silt loam from Mississippi, USA, harboring native bacteria possessing *atzABC* and *trzN* gene homologs did not control *Ipomoea lacunosa* L.[74] Moreover, under typical Mississippi Delta, USA, corn production systems, neither atrazine nor simazine applied at 1.2 kg ha^{-1} controlled *Sida spinosa* L. in an *s*-triazine-adapted field site.[12,75] It was postulated, therefore, that instances of reduced residual weed control with atrazine in soils with a prior use history in the USA, including Colorado, Hawaii, Louisiana, Mississippi, Tennessee and Texas, were associated with enhanced degradation and not *s*-triazine-resistant weed biotypes, improper application techniques or lack of activation.[12] Consequently, there is a need to identify where soils exhibiting enhanced *s*-triazine degradation presently occur.

3.3.2 Distribution of adapted soils

For this section, soils were designated as adapted by one of four methods, that is, whole soil extracts or bacterial isolates

Table 1. Bacteria reported to contain the *atzABCDEF* and (or) *trzNDF* genes

Bacterium	Genes	Accession number	Reference
Acidovorax avenae subsp. *Citrulli*	*trzD*	AF086815.2	51
Acidovorax sp. JLS4	*trzD*		52
Acinetobacter sp. C-1	*atzB*	AM901597.2	159
Agrobacterium J14a	*atzABC*		160,161
Agrobacterium radiobacter	*atzD*		52
Agrobacterium radiobacter J14a	*atzA* like		143,162
Agrobacterium sp.	*atzABCDEF*		59
Agrobacterium sp.	*atzA*		99
Agrobacterium tumefaciens	*atzB*		96,99,
Alcaligenes sp. SG1	*atzABC, trzD*		52,143,161
Aminobacter aminovorans	*atzC, trzD*		163
Arthrobacter sp.	*trzN*		59
Arthrobacter aurescens TC1	*trzN, atzBC*	CP000475.1	46,64,164
		AY456696.1	
Arthrobacter crystallopoietes strain Cit2	*atzBC*	AF364904.1	163
Arthrobacter globiformis	*trzN, atzC*		97
Arthrobacter nicotinovorans	*atzB*	AY650036.1	79
Arthrobacter nicotinovorans strain HIM	*atzABC*	AY650035.1	79
Arthrobacter sp.	*trzN*		59
Arthrobacter sp.	*trzN, atzBC*		165
Arthrobacter sp.	*trzN, atzBCD*		61
Arthrobacter sp.	*trzN, atzBC*		166
Arthrobacter sp.	*atzB*		96
Arthrobacter sp.	*trzN, atzBCD*		61
Arthrobacter sp. AD1	*atzA*		58
Arthrobacter sp. AD1	*atzA*	AF543694.1	58
Arthrobacter sp. AD25	*trzN*	DQ989289.1	Li and Cai, unpublished
	atzD	DQ989288.1	
Arthrobacter sp. AD26	*trzN*	EU091479.1	Li *et al.*, unpublished
	atzB	EU621846.1	Zhu *et al.*, unpublished
	atzC	EU621847.1	Zheng *et al.*, unpublished
Arthrobacter sp. AD26-2	*trzN*	EU400620.1	Wang *et al.*, unpublished
Arthrobacter sp. AD30	*trzN, atzBC*	FJ161691.1	Zheng *et al.*, unpublished
		FJ161693.1	
		FJ161695.1	
Arthrobacter sp. AG1	*trzN, atzBC*		165
Arthrobacter sp. MCMB-436	*trzN, atzBCD*	AY589015.1	61
		AY589016.1	
		AY589013.1	
		AY594331.1	
Beta proteobactria	*atzA*	AB194097.1	167
Beta proteobactria	*atzB*	AB194098.1	167
Caulobacter crescentus	*atzB*		96
Chelatobacter heintzii = *Aminobacter aminovorans*	*trzD, atzABC*		168
Chelatobacter heintzii strain Cit1	*atzABCD*	AF364900.1	163
		AF364901.1	
		AF364902.1	
		AF364903.1	
Clavibacter			161
Clavibacter michiganensis ATZ1	*atzABC*		127,143,161
Enterobacter cloacae	*trzD*	AF342826.1	51,169
Flavobacterium oryzihabitans	*atzB*		96
Flavobacterium sp.	*atzBC*		96
Herbaspirillum sp. B601	*atzABC*	DQ089655.2	Bazhanov unpublished
		AY965854.2	
		AY965855.2	

Table 1.   (*Continued*)

| Bacterium	Genes	Accession number	Reference
Isolate 38/38	*atzABC*		161
Klebsiella planticola 99	*trzD*		52
Klebsiella sp.	*atzA*		99
Mesorhizobium loti	*atzF*	BA000012.4	170
Nocardia sp.	*trzN, atzB*		96
Nocardioides kribbensis	*trzN, atzBC*		97
Nocardioides panacihumi (three isolates)	*trzN, atzC*		97
Nocardioides sp.	*trzN*		59
Nocardioides sp.	*trzN, atzBC*		59
Nocardioides sp.	*trzN, atzBC*		171
Nocardioides sp. C190	*trzN*		40,41
Nocardioides sp. SP12	*trzN; atzBC*	AF537327 AF537330 AF537329	171
Nocardioides sp. AN3	*trzN*	AB427184.1	172
Nocardioides sp. C190	*trzN*	AF416746.1	41
Nocardioides sp. CMU5	*trzN, atzB*	EF088652.1 EF088653.1	97
Nocardioides sp. MTD22	*trzN*	AB427183.1	172
Nocardioides sp. SP12	*trzN, atzBC*	AF537328.1 AF537330.1 AF537329.1	119,171
Polaromonas sp.	*trzN, atzBC*		59
Pseudaminobacter sp.	*AtzABC*		40
Pseudaminobacter sp. C147	*AtzABC*		39,40, 173
Pseudaminobacter sp. C195	*AtzABC*		39,40,173
Pseudomonas putida	*AtzB*		96
Pseudomonas sp. CN1	*TrzD*		52
Pseudomonas sp. NRRLB-12227	*TrzD*		53
Pseudomonas sp. NRRLB-12228	*TrzD*		52
Pseudomonas sp.	*atzABCDEF*		175
Pseudomonas sp. AD39	*AtzC*	FJ161696.1	Zheng *et al.*, unpublished
Pseudomonas sp. AD39	*AtzB*	FJ161694.1	Zheng *et al.*, unpublished
Pseudomonas sp. AD39	*AtzC*	FJ161696.1	Zheng *et al.*, unpublished
Pseudomonas sp. AD39	*TrzN*	FJ161692.1	Zheng *et al.*, unpublished
Pseudomonas sp. ADP	*atzABCDEF*		39
Pseudomonas sp. ADP	*atzABCDEF*	U66917.2	47
Pseudomonas sp. C-15	*AtzB*	AM901596.2	159
Pseudomonas sp. CN1	*AtzAC*		87
Pseudomonas sp. YAYA6	*atzA* like		174
Ralstonia M91-3	*AtzABC*		161,176,177
Ralstonia picketti D	*atzA* like		143
Ralstonia picketti D	*AtzD*		52
Ralstonia sp. M91-3	*TrzD*		52
Rhizobium leguminosarum	*AtzBC*		96
Rhizobium sp.	*atzA*		178
Rhizobium sp.	*AtzC*		96
Rhizobium sp. PATR	*AtzA*		143,178
Sinorhizobium sp.	*trzN, atzBC*		59
Sphingomonas yanoikuyae	*AtzB*		96
Stenotrophomonas maltophilia	*AtzA*		163
Stenotrophomonas sp.	*AtzAB*		179
Variovorax paradoxus	*AtzB*		96
ß-proteobacterium	*atzABCDEF*		167
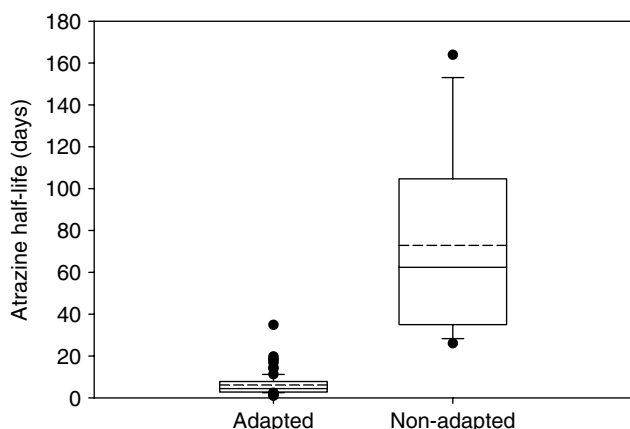
Figure 5. Box plot comparisons of published data for atrazine dissipation in *s*-triazine-adapted and non-adapted soil. Boundary of box closest to zero indicates the 25th percentile, a solid line within the box marks the median, a dashed line within the box delineates the mean and the boundary of the box furthest from zero indicates the 75th percentile. Error bars above and below the box indicate the 90th and 10th percentile, and solid dots indicate outliers. The number of independent observations is 14 for non-adapted soils and 84 for *s*-triazine-adapted soils.[11,82,84,85,91,104,105,107,108,125,158] .
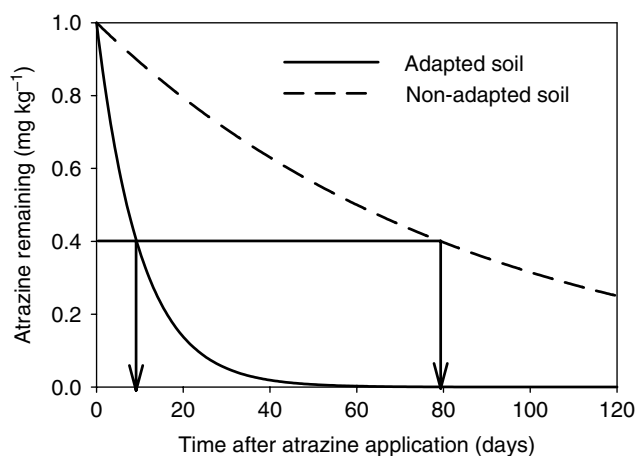

Figure 6. Theoretical example of residual weed control with atrazine in adapted and non-adapted soils. In this example, 0.4 mg kg^{-1} atrazine is required for 85% weed control. In a non-adapted soil, residual atrazine does not decline to 0.4 mg kg^{-1} until 80 days after atrazine application, while the atrazine concentration will decline to that same level in an adapted soil in only 10 days.

Table 2.	Intercontinental reports of enhanced *s*-triazine degradation	
Continent	Country/region	Reference
Asia	China	58,165,166,180,181
	Croatia	59,98,182
	India	61,183
	Iran	184
	Israel	185
	Japan	94,167,172,186–188
Australia	New South Wales	91,189
Africa	Kenya	190
Europe	Belgium	82,88,191,192
	France	40,59,62,83,90,95,103,110, 117–119,125,126;163, 171,178,193
	Hungary	88,194
	Ireland	88
	Italy	159
	Spain	93,195–197
	Switzerland	174,198,199
North America	California	17,96,127,143,161
	Colorado	9,11,17,104,112
	Delaware	89
	Florida	200
	Illinois	17,201
	Indiana	161,202
	Iowa	153
	Louisiana	161
	Maryland	16
	Mexico	128
	Minnesota	37–39,161,203
	Mississippi	9,11,12,17,75,85,105,114
	Nebraska	97,160,161,204
	North Carolina	205
	North Dakota	99
	Ohio	68,111,123,161,176,177,206,207
	Ontario	40,41,83
	Pennsylvania	208
	Quebec	40,46,209,210
	South Dakota	211,212
	Tennessee	68,84
	Texas	109,113
South America	Argentina	107,108,121,122,213
	Chile	175,179,214,215
	Colombia	216

were positive for *atzABCDEF* or *trzNDF* gene homologs, dissipation assays,[17] mineralization assays[12] or they were declared as such by the author. Within this context, *s*-triazine-adapted soils are reported for the Hawaiian island (Shaner, private communication), the New Zealand islands[78–81] and all continents except Antarctica (Table 2; Fig. 7). Enhanced *s*-triazine degradation is not, therefore, a geographically localized problem, and, in order to determine where adaptation may occur in the future, the physiochemical range of known adapted soils was delineated.

3.3.3 Physiochemical ranges
Soil data from the aforementioned studies were evaluated to delineate the physiochemical range of *s*-triazine-adapted soils. Enhanced degradation occurs in soils ranging in texture from 3 to 90% sand, 4 to 60% silt and 3 to 56% clay, which covers

all possible soil texture classes (Fig. 8). However, 90% of soils exhibiting enhanced degradation have textures ranging from 5 to 60% sand, 19 to 74% silt and 15 to 41% clay. Transposing these data onto the textural triangle reveals that 90% of known *s*-triazine-adapted soils group into the silty clay, clay loam, silty clay loam, sandy clay loam, loam, silt loam or sandy loam classes (Fig. 9). Furthermore, adaptation has been documented in soils with organic carbon contents ranging from 0.5 to 46%, with 90% of these soils having total organic carbon levels between 1.0 and 2.8% (Fig. 8). Soil pH levels ranging from 5.1 to 8.6 are reported for *s*-triazine-adapted soils, with 90% of the soils having a pH between 5.8 and 8.1 (Fig. 8).
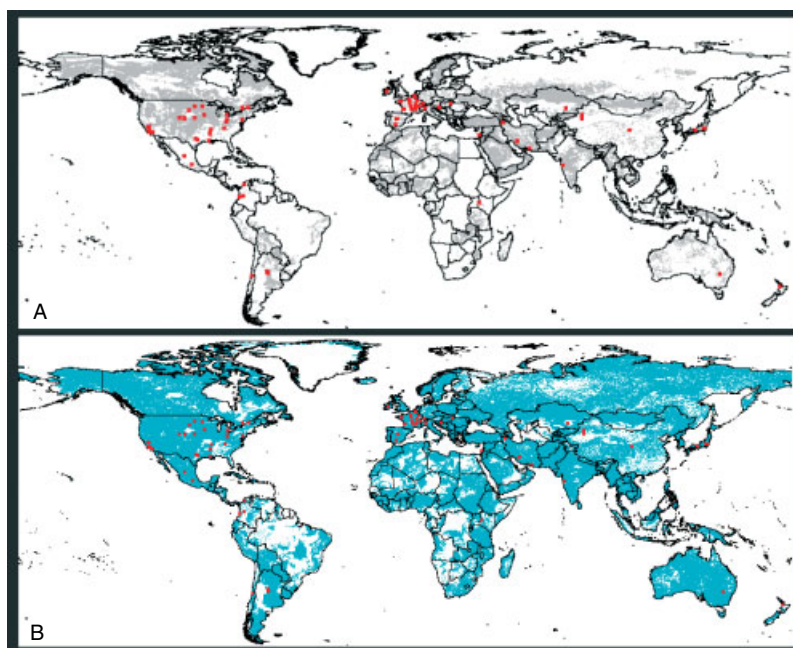
Figure 7. Global distribution of surface soils with physiochemical properties (percentage sand, silt, clay and organic carbon, and pH) consistent with those known to be exhibiting enhanced *s*-triazine degradation. Locations known to be exhibiting enhanced *s*-triazine degradation are noted with red dots. Panel A has highlighted in grey those soils with physiochemical properties within the 10–90th percentile range of known adapted soils. Panel B has highlighted in blue those soils with physiochemical properties within the minimum–maximum range of known adapted soils.
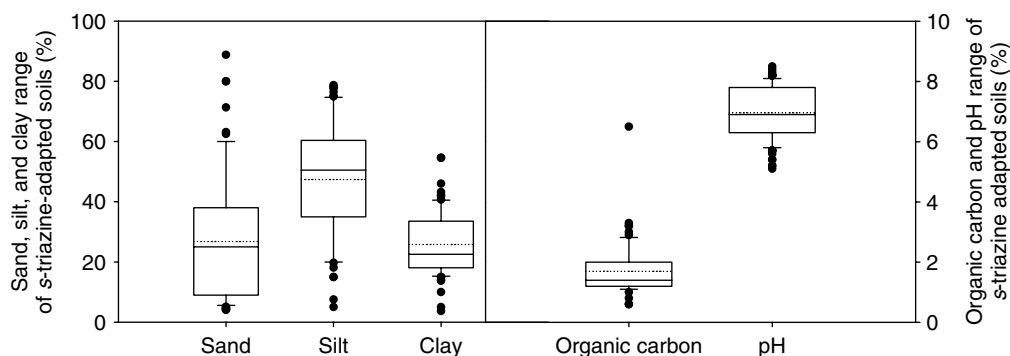

Figure 8. Physicochemical range of known *s*-triazine-adapted soils.

3.3.4 Soil pH as the principal physiochemical driver

Of the evaluated soil physiochemical properties, only acidic soil conditions restrict adaptation, particularly in Europe.[82,83] In two European screening studies spanning 83 soils varying widely in texture and pH, enhanced degradation was not observed in soils with pH <6.1.[82,83] In North America, enhanced degradation occurs in soils with pH ≥5.1, but the dissipation rate is reduced at pH <5.5.[84,85] For instance, the average atrazine half-life for a Tennessee, USA, *s*-triazine-adapted soil with a pH ranging from 5.7 to 7.5 was 2.8 days ($n = 20$; SD = 0.27).[84] When the soil pH ranged from 5.1 to 5.4, however, the average atrazine half-life was 11.4 days ($n = 4$; SD = 1.29). Thus, atrazine half-life increased fourfold at pH ≤5.4, yet was still fivefold lower than the historic persistence estimate, 60 days.

The pH effects on microbial ecology likely contribute to increased *s*-triazine persistence in adapted soils. For example, *s*-triazine catabolism in agricultural soils is mediated primarily by bacterial consortia.[68,86–99] Soil bacterial diversity and richness are maximal at neutral pH but decline as pH decreases, a trend consistent from field to continent scale.[100,101] Moreover, as soil pH declines, the microbial population shifts from bacterial to fungal dominated.[102] Thus, increased *s*-triazine persistence in acidic soils and (or) the inability of acidic soils to exhibit enhanced degradation are likely a function of decreased diversity and richness among the atrazine-degrading bacterial community.

3.3.5 Herbicide use history as the principal cultural management practice

A prerequisite for enhanced degradation is the selection of soilborne bacteria with an adaptation that enables rapid pesticide degradation.[9] To date, atrazine-degrading bacteria or the *atzABCDEF* and *trzNDF* gene homologs have been detected in agricultural soils only when these have been previously exposed to *s*-triazine herbicides.[17,83,85,103,104] This likely signifies that *s*-triazine herbicides are the primary agents responsible for the selection of *s*-triazine-degrading bacteria with *atzABCDEF* and *trzNDF* gene homologs. The development of enhanced atrazine degradation in
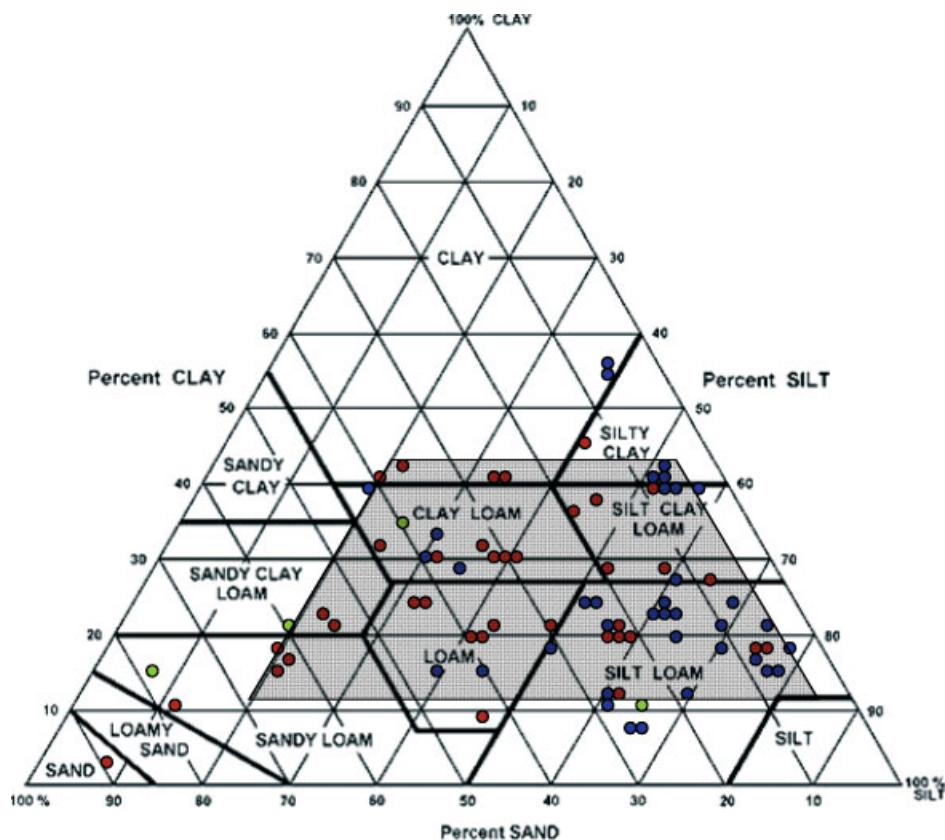
Figure 9. Textural triangle revealing 10–90th percentile of known *s*-triazine-adapted soils.

agricultural soils will depend primarily on herbicide use history, particularly *s*-triazine application frequency.

Not only do recurrent *s*-triazine applications select for bacteria able rapidly to degrade the herbicide, but frequent application increases the degrader population and (or) their activity. The relationship between application frequency and atrazine-degrader number/activity is inferred from European and North American screening studies,[17,82,83,85,104] but is clearly illustrated with data from a 6 year replicated field study conducted in Mississippi, USA, on a Dundee silt loam.[105] Prior to receiving an *s*-triazine application, the Dundee silt loam mineralized 9.2% of ring-labeled ^{14}C-atrazine 30 days after application, which is typical for non-adapted soil (Figs 3 and 10). One year after the first atrazine application, the soil mineralized 55% of ring-labeled ^{14}C-atrazine by 30 days after application, a value representative of *s*-triazine-adapted soils. Atrazine mineralization increased incrementally with successive applications until year 5, at which time it plateaued at approximately 80%. Results from this replicated field study indicate that one atrazine application, even at a typical field rate for corn, can induce enhanced degradation, and that successive applications increase either the atrazine-degrader population and (or) their activity.

Decreasing the *s*-triazine application frequency may slow the development and (or) minimize the adverse agronomic effects associated with enhanced degradation. One *s*-triazine application every 2 years, however, does not retard adaptation,[74,82,83,105–110] or increase residual weed control.[74] Atrazine dissipation in soil with an application frequency of once every 36 months is reduced relative to soils receiving annual applications,[111,112] but weed control data are lacking for this application interval. The authors suspect that
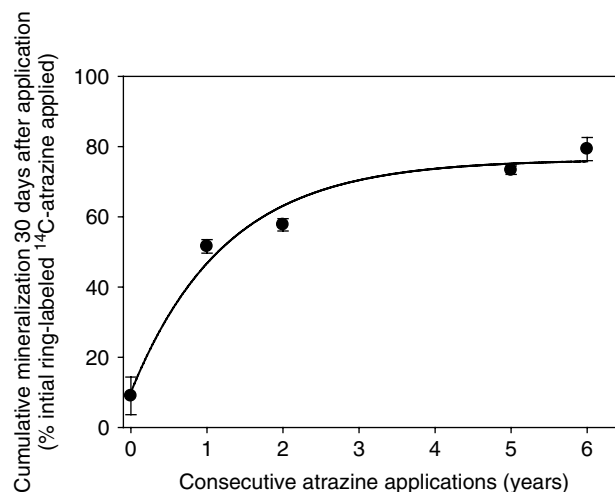
Figure 10. Mineralization of ring-labeled ^{14}C-atrazine as a function of *s*-triazine exposure history. One exposure increased cumulative atrazine mineralization from 10 to 50%. After five exposures, cumulative atrazine mineralization approached 80%. Adapted from Zablotowicz *et al*[114].

an application frequency of once every 48 months may suppress adaptation, but field data are required for verification.

The coapplication of glyphosate or glufosinate with atrazine increases the herbicide's persistence in *s*-triazine-adapted soils. For instance, at 8 and 12 days after the application of atrazine (3.86 mg kg^{-1}) and glyphosate (0, 43, 86, 129, 172 or 215 mg kg^{-1}) to a Weswood silt loam, extractable atrazine was positively correlated with glyphosate rate; yet, regardless of glyphosate rate,

Table 3. Crop and crop rotations exhibiting enhanced *s*-triazine degradation

Crop	Reference
Monocultures	
Avocado	215
Citrus	Hanson, unpublished
Corn	9,11,12,17,62,68,74,75,82–86,88–90, 95,97,103–105,109–112,114,117–119, 121–123,125–127,163,171,178,181,184, 191,192,200,207,209–211
Grape	17
Olive	93
Pineapple	Shaner, unpublished
Sorghum	113
Sugar cane	158,190
Turf	88,167,205
Rotations	
Corn–barley	82,110
Corn–cotton	17,74,85,105
Corn–sorghum	17,109
Corn–soybean	17,82,104,107,108,121,122,153,207
Corn–sunflower	104
Corn–wheat	82,89,103,110
Corn–wheat–soybean	111,123,207
Corn–wheat–fallow	112
Corn–wheat–sunflower	112

atrazine half-life was ≤8 days.[113] Similarly, 5 days after the application of atrazine (2.5 mg kg^{-1}) and glufosinate (0, 10, 20 or 40 mg kg^{-1}) to a Dundee silt loam, extractable atrazine was positively correlated with glufosinate rate, but, regardless of treatment, atrazine's half-life was ≤5 days.[114] These data demonstrate that the co-application of glyphosate or glufosinate with atrazine transiently increases the herbicide's persistence in *s*-triazine-adapted soils. It is unlikely, however, that field application rates of glyphosate or glufosinate increase the residual activity of *s*-triazine herbicides under field conditions.

3.3.6 Cropping history

Enhanced degradation is reported for nine crops and six crop rotation systems that rely on *s*-triazine herbicides for residual weed control (Table 3). The authors submit, therefore, that enhanced degradation can occur in any cropping system where this class of herbicides remains a component of the weed control program if soil pH is greater than approximately 5.4 and the application frequency is greater than once every 4 years. Other crops potentially affected by enhanced *s*-triazine degradation include those treated with ametryn, i.e. bananas and plantains, with atrazine, i.e. macadamia nuts, conifers and guava, with prometon, i.e. industrial sites and railroad rights of way, with prometryn, i.e. cotton, celery and pigeon peas, and with simazine, i.e. strawberries, almonds, nectarines, apples, blueberries, caneberries, cranberries, established Christmas trees, filberts, lemons, pears, pecans, shelterbelts, sour cherries, walnuts, peaches, plums, sweet cherries, lemons, oranges and grapefruit.[115]

Although no production system impedes *s*-triazine adaptation, there is evidence that corn facilitates adaptation through a rhizosphere effect. The rhizosphere is a zone of soil under the direct influence of plant roots; thus, it is carbon enriched and maintains a larger and more diverse microbial population than non-rhizosphere soil.[116] Accordingly, atrazine-degrader numbers, diversity, activity and survival are often greater in the rhizosphere of corn than in bulk soil.[86,95,117–119] It is unknown whether the rhizosphere of other crops exhibit a similar stimulatory effect on the *s*-triazine-degrader population, as these studies are lacking.

3.3.7 Residue management systems

Residue management practices, that is, production systems that leave 30% or more of the crop residues on the soil surface after planting, can alter pesticide persistence relative to conventional tillage systems.[120] Residue management systems evaluated to date do not appear to retard the development of enhanced *s*-triazine degradation. *s*-Triazine-adapted soils occur in conventional tillage,[85,107–109] reduced tillage,[9,11–12,74–75,105,121] no-tillage[84,105,107,108,121,122] and cover crop production systems.[121,123] Studies comparing enhanced atrazine degradation among residue management systems are limited, but, when evaluated, atrazine dissipation is similar among NT, CT and cover crop systems.[121] Thus, residue management systems do not appear to impede adaptation when *s*-triazine herbicides are a component of the weed control program.

3.3.8 Exogenous N

Isolation techniques and ^{15}N-labeling studies reveal that catabolizing bacteria can utilize *s*-triazine herbicides as an N source.[124] It was postulated that this relationship could be exploited to alter *s*-triazine persistence in adapted soils. For example, under laboratory conditions, the addition of exogenous N to pure culture and native *s*-triazine-adapted soils transiently suppressed *s*-triazine degradation by adapted microbial populations, with the suppression level depending primarily on the bacterial population responsible for degradation and the N source applied.[82,89,114,125–130] Additionally, atrazine-catabolizing genes, particularly *atzDEF*, are subject to a complex regulatory circuit that is modulated by N availability and the presence of degradation products, e.g. cyanuric acid.[129,130] Thus, coinciding N fertilization with *s*-triazine applications could increase herbicide persistence. Field data from Colorado, Tennessee and Mississippi, USA, indicate that *s*-triazine half-life values are ≤9 days under typical corn production systems, even when N is applied at recommended field rates, approximately 150 kg ha^{-1}.[12,75,84,104] It is unlikely, therefore, that synchronizing *s*-triazine applications with N fertilization will appreciably extend the residual activity of *s*-triazine herbicides in adapted soils.

3.3.9 Minimizing effects of enhanced s-triazine degradation

There are three historical approaches to dealing with enhanced pesticide degradation:[131]

- increase pesticide application rate and frequency;
- switch to pesticides from alternative chemical families;
- integrated pest management (IPM), that is, utilize a combination of pest control strategies including mechanical, physical, genetic, biological, cultural and chemical.

Applying the approaches described above to the problem of enhanced *s*-triazine degradation reveals one feasible solution, IPM. For example, increasing application rate and frequency is not a viable option in that regulatory limits may be violated. Switching to another herbicide is not a sensible option because of cross-adaptation among *s*-triazine herbicides, regional and

global registration restrictions on alternative herbicide families and limited economically available alternative herbicides, particularly for corn. Integrated pest management, therefore, is the most feasible option for controlling enhanced atrazine degradation.

In the context of IPM, two strategies can be employed:[131,132]

- prevent the development of enhanced degradation;
- circumvent the consequence of enhanced degradation in adapted soils.

In the case of enhanced *s*-triazine degradation, the present analysis indicates that the phenomenon can occur in any cropping system if (1) the soil pH is above approximately 5.4 and (2) *s*-triazines are applied more than once every 3–4 years. Consequently, if the aim is to prevent enhanced degradation, then *s*-triazine herbicides should not be applied more than once every 4 years. This will necessitate the use of alternative herbicides, which, as stated above, may not be feasible owing to regional registration restrictions and limited economically viable alternative chemistries. Conversely, if the objective is to maintain acceptable weed control in the context of existing enhanced degradation, then a number of interconnected strategies could be employed:

- supplement *s*-triazine residual weed control with additional pre-emergent chemistries from alternative herbicide families;
- plant herbicide-resistant varieties and rely on post-emergence applications and (or) cultivation for weed control;
- apply *s*-triazine herbicides post-direct, thereby taking advantage of foliar uptake.

3.4 Environmental implications of enhanced degradation

3.4.1 Fate, transport and environmental risk assessment predictions

Pesticide persistence is an integral component of fate, transport and risk assessment models. Sensitivity analysis, that is, investigations into how model performance varies along with changes in the key assumptions on which the model projections are based, indicates that inaccurate persistence estimates adversely affect model predictions.[80,133–141] The primary environmental concern associated with enhanced *s*-triazine degradation, therefore, is that historic *s*-triazine persistence estimates, if used as default input parameters, will result in erroneous fate, transport and environmental risk predictions for adapted soils. Herein, the differences in atrazine persistence between adapted and non-adapted soils are described, how these differences will affect environmental modeling and risk assessment is discussed and alternative default input parameters for *s*-triazine-adapted soils are recommended.

3.4.2 Historic atrazine persistence estimates

The USEPA indicates that atrazine's average half-life in soil under aerobic laboratory conditions is 3–4 months,[142] which is consistent with Wauchope's review of the literature[13] and the present authors' analysis of published data for non-adapted soils (Fig. 5). Conversely, the average atrazine half-life under aerobic laboratory conditions in *s*-triazine-adapted soils is 6 days (Fig. 5). The differences in atrazine persistence between non-adapted and *s*-triazine-adapted soils are significant in that a theoretical twofold decrease in herbicide persistence can reduce the predicted off-site loss tenfold.[134]

3.4.3 Metabolite formation and persistence in adapted and non-adapted soils

The concentration of atrazine's mono-*N*-dealkylated metabolites is typically lower in *s*-triazine-adapted soil than in non-

adapted soil, which has been attributed to two competing processes.[9,11] First, atrazine-degrading consortia containing *atzABCDEF* and *trzNDF* gene homologs circumvent biologically mediated *N*-dealkylation reactions, thereby focusing atrazine dissipation primarily through the novel *s*-triazine pathway (Fig. 3).[11] This hypothesis is supported by modeling data, which indicate that, in *s*-triazine-adapted soils, 71, 23 and 6% of the parent compound is converted to HA, DEA and DIA respectively.[14] Conversely, in non-adapted soils, the projected conversion of atrazine to daughter products is 10% for HA, 72% for DEA and 18% for DIA.[9] Second, the mono-*N*-dealkylated metabolites of atrazine are substrates for *atzA* and *trzN*, and thus, in *s*-triazine-adapted soils, DEA and DIA could be quickly converted to hydroxy-*s*-triazine intermediates.[44,143] For example, the mean half-life value for DEA and DIA in non-adapted soil is 52 and 36 days respectively.[144–146] In *s*-triazine-adapted soil, however, the mean half-life is 10 days for DEA and 8 days for DIA.[9] The differences in metabolite formation and dissipation between *s*-triazine-adapted soils are significant in that the *n*-dealklyated metabolites are included in atrazine's risk assessment.[142]

3.4.4 Temperature and moisture effects on atrazine degradation in adapted soils

Prominent transport models contain submodules that predict pesticide degradation as a function of temperature and moisture:[11,147]

$$C_t = C_0 \, e^{-k(T, \theta)} \tag{1}$$

$$T_{1/2} = 0.693/k \tag{2}$$

$$k_{(T, \theta)} = k_{ref}Q_{10}[(T - T_{ref})/10](\theta/\theta_{ref})\beta \tag{3}$$

$$Q_{10} = T_{1/2\,20\,°C}/T_{1/2\,10\,°C} \tag{4}$$

$$\beta = -[\log(T_{1/2\,70\%FC}) - \log(T_{1/2\,40\%FC})]/\log(70/40) \tag{5}$$

where C_t is the pesticide concentration in soil at time t (mg kg^{-1}), C_0 is the initial pesticide concentration (mg kg^{-1}), $k_{(T,\theta)}$ is the rate constant at actual temperature T (°C) and soil moisture θ, k is the first-order rate constant in reciprocal days (day^{-1}) at reference temperature and moisture, Q_{10} is the factor by which degradation increases when T increases by 10 °C, T is the actual temperature (°C), T_{ref} is the reference temperature, θ is the actual soil moisture [% field capacity (FC)], θ_{ref} is the reference soil moisture (40% FC) and β is the moisture exponent. FOCUS, which reviewed 148 Q_{10} estimates for various pesticides, concluded that Q_{10} does not vary significantly among pesticides, and recommended a default Q_{10} of 2.20, which is standard for most fate and transport models.[148] On review of subsequent data, however, the Panel on Plant Protection Products and their Residues (PPR-Panel) reversed their decision:

- There are group-specific and compound-specific differences in Q_{10}.
- One median Q_{10} value for all pesticides should not be assumed.
- Compound-specific Q_{10} values should be used rather than default values.
- Until such data are available, a default Q_{10} value of 2.58 is recommended.[149]

To date, one study has evaluated temperature and moisture effects on atrazine degradation in *s*-triazine-adapted soils, and separate Q_{10} values for adapted (3.00) and non-adapted soils (2.20) were recommended.[11] Until these findings are substantiated,

Table 4. Leaching estimation and chemistry model (LEACHM) simulations for the Morgan Creek, Maryland, watershed. Projections are based on 10 year simulations (1995–2004) for the top metre of s-triazine-adapted or non-adapted unsaturated silty loams when subjected to annual rotations of corn (*Zea mays* L.) and soybean [*Glycine max* (L.) Merr.]. Five biennial applications of 2.08 kg ha^{-1} of atrazine were applied in the spring preceding the planting of corn

Soil	Compound	MF ratio[a]	$T_{1/2}$[b] (days)	V_{mhf}[c]	Applied/produced (g ha^{-1})	Leached (g ha^{-1})	Residual (g ha^{-1})	Leached (% applied)
Adapted	Atrazine		6	11	10 400	23	0	0.22
	Desethylatrazine	0.22	10	11	2350	25	0	0.24
	Deisopropylatrazine	0.07	8	11	600	4	0	0.04
	Hydroxyatrazine	0.71	6	11	7420	9	0	0.09
	Total residues					61	0	0.59
Non-adapted	Atrazine		60	3	10 400	284	35	2.73
	Desethylatrazine	0.72	52	3	7250	464	69	4.46
	Deisopropylatrazine	0.18	36	3	1810	67	8	0.64
	Hydroxyatrazine	0.10	60	3	1010	22	14	0.21
	Total residues					837	126	8.04

[a] MF ratio is the ratio of atrazine converted to desethylatrazine, deisopropylatrazine and hydroxyatrazine.[9,12,14]
[b] $T_{1/2}$ is the near-surface half-life for atrazine, desethylatrazine, deisopropylatrazine and hydroxyatrazine.[9–11]
[c] V_{mhf} is the maximum multiple of the near-surface half-life that the half-life reaches at 100 cm.[10]

it is prudent to adjust default Q_{10} values from 2.20 to 2.58, regardless of herbicide use history. Conversely, moisture effects on atrazine degradation indicate that differential β values are not required for modeling atrazine fate and transport in *s*-triazine-adapted and non-adapted soils.[11] This finding corroborates that of FOCUS, which recommended a default β of +0.8 for pesticide fate and transport modeling, as variations in measurements of β for individual pesticides are as great as variations between pesticides.[148]

3.4.5 Soil depth effects on atrazine degradation in adapted soils

Atrazine persistence, regardless of herbicide use history, generally increases with depth in the soil profile.[9,78,81,108,145,146,150–153] Models like RZWQM and LEACHM, therefore, allow the user to vary the maximum half-life factor, that is, the multiple of the near-surface half-life that the half-life reaches at 100 cm.[10] Data from Colorado and Mississippi, USA, indicate that the maximum half-life factor will be at least 3–4-fold higher in *s*-triazine-adapted soil than in non-adapted soil.[9] For example, if the maximum half-life factor for non-adapted soil is 3, then a maximum half-life factor ranging from 9 to 12 would be appropriate for the same soil exhibiting enhanced degradation.

3.4.6 LEACHM predictions for adapted and non-adapted soils

LEACHM predictions were notably different between *s*-triazine-adapted and non-adapted soils (Table 4). By accounting for the altered metabolic pathway and reduced persistence estimates for the parent and daughter products in *s*-triazine-adapted soil, leaching potential relative to non-adapted soil was reduced 12.3-fold for atrazine, 18.5-fold for DEA, 16.8-fold for DIA, 2.4-fold for HA and 13.7-fold for total *s*-triazine residues. LEACHM simulations demonstrate that, if historic dissipation pathways and rate constants are assumed for *s*-triazine-adapted soils, then herbicide fate, transport and risk assessment errors will be considerable. Given the number of soils known to exhibit enhanced degradation, coupled with the projections on where this phenomenon can occur (Fig. 7), there is a need to develop a reliable method to predict atrazine persistence in agricultural soils.

3.5 Predicting atrazine persistence across the agricultural landscape

A multiple linear regression model able to predict atrazine half-life values in soil was constructed using the stepwise procedure in SAS (model 1):

$$\log_{10}(T_{1/2}) = 0.18080 - [0.62740 \times \log_{10}(\text{pH})]$$
$$- [0.32927 \times \log_{10}(C_{yr})]$$
$$+ 0.87216h \qquad \text{(model 1)}$$

where $T_{1/2}$ is the atrazine half-life value standardized to 20 °C via the Arrhenius equation (days), pH is minus the decimal logarithm of the hydrogen ion activity in a 1 : 1 aqueous soil solution paste, C_{yr} is the consecutive years of atrazine applications, ranging from 0 (no applications) to 5 (five consecutive applications), and h is the atrazine use history in the last 5 years, where soils receiving an atrazine application $= 1$ and soils not receiving an atrazine application $= 2$. The model and all parameters contained in it were highly significant: $P_{0.05}$ for full model <0.0001, $P_{0.05}$ for pH <0.0001, $P_{0.05}$ for $C_{yr} = 0.0206$ and $P_{0.05}$ for $h < 0.0001$. Conversely, tillage, soil texture and organic carbon did not significantly contribute to model 1 (data not reported).

Model 1 grouped 90 soils from five continents, including Africa, Australia, Europe, North America and South America, into one of three categories:

- adapted, i.e. atrazine half-life $\leq$15 days;
- intermediate, i.e. atrazine half-life ranging from 15 to 30 days;
- non-adapted, i.e. atrazine half-life >30 days (Table 5).

The model accurately grouped 11 of 12 non-adapted soils and 73 of 77 adapted soils. Model deficiencies were primarily associated with discriminating between adapted and intermediately adapted soils, particularly in Europe. Moreover, model 1 was a better predictor of atrazine half-life values than the historic estimate (Table 5). It is likely, therefore, that this model, or an updated version of it, could be used to generate initial atrazine persistence estimates for environmental and risk assessment modeling.

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Table 5. Observed and predicted atrazine half-life ($T_{1/2}$) values in soils from five continents that have or have not been exposed to atrazine

Continent	Country/region	$T_{1/2}$ predicted (days)[a]	$T_{1/2}$ observed (days)	Error reduction (%)[b]	Reference
		Soils with an atrazine use history			
Africa	Africa	5.3	18.7	149.3	158
South America	Argentina	4.5	11.3	370.7	107
Australia	Australia	3.2	7.0	703.0	91
Europe	Belgium	4.5	2.3	2400.7	82
Europe	Belgium	4.7	2.8	2012.2	82
Europe	Belgium	4.5	3.1	1800.6	82
Europe	Belgium	3.6	3.2	1768.5	82
Europe	Belgium	2.7	4.2	1299.2	82
Europe	Belgium	4.0	4.3	1290.8	82
Europe	Belgium	3.0	4.3	1269.0	82
Europe	Belgium	4.8	4.5	1224.0	82
Europe	Belgium	4.5	4.6	1197.0	82
Europe	Belgium	2.8	4.6	1160.0	82
Europe	Belgium	4.4	5.0	1101.6	82
Europe	Belgium	4.2	6.3	823.5	82
Europe	Belgium	4.7	6.7	764.4	82
Europe	Belgium	5.1	7.9	621.9	82
Europe	Belgium	5.6	8.5	575.0	82
Europe	Belgium	5.5	9.5	492.2	82
Europe	Belgium	5.8	9.5	495.5	82
Europe	Belgium	5.6	10.2	441.6	82
Europe	Belgium	5.4	10.5	426.2	82
Europe	Belgium	4.1	10.9	388.2	82
Europe	Belgium	3.9	14.5	240.3	82
Europe	Belgium	5.6	16.8	190.0	82
Europe	Belgium	6.4	19.8	135.6	82
Europe	Belgium	5.6	34.9	−11.8	82
North America	Colorado	6.1	1.5	3595.3	104
North America	Colorado	3.5	2.0	2824.0	104
North America	Colorado	2.8	2.0	2862.2	104
North America	Colorado	4.1	2.2	2543.0	104
North America	Colorado	3.0	2.5	2281.6	104
North America	Colorado	3.0	2.6	2194.1	104
North America	Colorado	3.2	4.0	1380.1	11
North America	Colorado	5.8	4.2	1290.6	104
North America	Colorado	10.6	10.4	474.6	104
Europe	France	2.7	8.9	504.1	125
North America	Mississippi	3.8	1.0	5618.1	11
North America	Mississippi	3.5	3.1	1821.8	105
North America	Mississippi	6.4	3.2	1676.3	105
North America	Mississippi	6.4	4.1	1306.2	85
North America	Mississippi	5.3	4.4	1242.5	85
North America	Mississippi	3.8	4.6	1187.8	85
North America	Mississippi	5.3	4.8	1140.4	85
North America	Mississippi	4.8	4.8	1149.5	85
North America	Mississippi	6.7	4.9	1087.8	85
North America	Mississippi	4.4	5.0	1088.6	105
North America	Mississippi	5.1	5.3	1027.6	105
North America	Mississippi	7.0	5.5	964.1	85
North America	Mississippi	6.4	5.5	973.7	85
North America	Mississippi	5.1	5.7	941.4	105
North America	Mississippi	7.0	5.9	898.8	85
North America	Mississippi	6.4	5.9	909.2	105
North America	Mississippi	5.3	6.2	859.4	85
North America	Mississippi	6.4	7.7	662.0	85
North America	Mississippi	6.7	10.0	467.0	85

Table 5. (*Continued*)

Continent	Country/region	$T_{1/2}$ predicted (days)[a]	$T_{1/2}$ observed (days)	Error reduction (%)[b]	Reference
North America	Mississippi	8.8	10.7	442.6	85
North America	Mississippi	9.6	14.1	293.5	85
North America	Mississippi	6.4	18.0	168.7	85
North America	Tennessee	3.2	2.4	2366.4	84
North America	Tennessee	4.0	2.5	2238.6	84
North America	Tennessee	4.8	2.6	2123.2	84
North America	Tennessee	3.8	2.6	2161.8	84
North America	Tennessee	3.4	2.6	2176.6	84
North America	Tennessee	4.9	2.7	2039.4	84
North America	Tennessee	4.6	2.7	2051.5	84
North America	Tennessee	4.0	2.7	2072.8	84
North America	Tennessee	4.1	2.7	2069.2	84
North America	Tennessee	3.3	2.7	2098.6	84
North America	Tennessee	4.7	2.8	1974.5	84
North America	Tennessee	3.6	2.8	2014.6	84
North America	Tennessee	4.2	3.0	1858.5	84
North America	Tennessee	4.2	3.0	1860.8	84
North America	Tennessee	4.4	3.1	1794.7	84
North America	Tennessee	5.6	3.2	1698.6	84
North America	Tennessee	7.3	3.3	1597.6	84
North America	Tennessee	4.8	3.5	1578.1	84
North America	Tennessee	5.9	3.7	1463.3	84
North America	Tennessee	4.2	3.7	1506.9	84
North America	Tennessee	6.3	10.6	425.1	84
		Soils with no known atrazine use history			
Africa	Africa	51.3	164.0	−5.3	190
Africa	Africa	130.0	88.0	−15.9	190
South America	Argentina	56.3	121.8	−3.1	107
South America	Argentina	48.6	142.2	−8.0	108
Europe	Belgium	71.4	53.0	−21.5	82
North America	Colorado	44.1	26.1	60.9	104
North America	Colorado	40.6	36.0	54.0	11
Europe	France	37.0	99.0	−23.2	125
North America	Mississippi	52.8	30.5	23.5	85
North America	Mississippi	51.3	32.0	27.1	11
North America	Mississippi	47.4	37.0	34.2	105
North America	Mississippi	48.6	38.2	29.9	85
North America	Mississippi	51.3	71.8	−12.1	85
North America	Mississippi	47.4	81.0	−15.6	85

[a] Predicted values are based on model 1: $\log_{10}(T_{1/2}) = 0.18080 - [0.62740 \times \log_{10}(\text{pH})] - [0.32927 \times \log_{10}(C_{yr})] + 0.87216h$, where $T_{1/2}$ is the atrazine half-life value standardized to $20\,^{\circ}$C via the Arrhenius equation (days), pH is minus the decimal logarithm of the hydrogen ion activity in a 1 : 1 aqueous soil solution paste, C_{yr} is the consecutive years of atrazine applications, ranging from 0 (no applications) to 5 (five consecutive applications), and h is the atrazine use history in the last 5 years, where soils receiving an atrazine application = 1 and soils not receiving an atrazine application = 2. The model and all parameters contained in it were highly significant: $P_{0.05}$ for full model <0.0001, $P_{0.05}$ for pH <0.0001, $P_{0.05}$ for $C_{yr} = 0.0206$ and $P_{0.05}$ for $h < 0.0001$.
[b] Reduction in error afforded by model 1 relative to historic estimate, 60 days, that is, error reduction = [(60 days − observed)/observed] ×100 − [(model 1 projection − observed)/observed] ×100. Negative values indicate less error associated with historic estimate relative to model 1.

4   CONCLUSIONS AND FUTURE RESEARCH NEEDS

Basic and applied research indicates a recent bacterial adaptation that enables rapid detoxification of *s*-triazine herbicides. The genes that allow bacteria to use *s*-triazine herbicides as a sole C and (or) N source are primarily plasmid mediated, are susceptible to horizontal gene transfer, occur in at least four bacterial phyla and have been identified on six continents. The substrate range of these metabolic pathways encompasses most commercially available *s*-triazine herbicides and has been linked to enhanced degradation in agricultural soils from around the globe.

Linking these microbial adaptations with enhanced degradation has significant agronomic implications. Decreased residual weed control with *s*-triazine herbicides has been confirmed under laboratory, greenhouse and field conditions. With the exception of *s*-triazine application frequency and soil pH, cultural management practices and soil physiochemical properties do not retard adaptation. Thus, it appears that enhanced degradation can occur

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in any agricultural soil with a pH greater than approximately 5.4, given sufficient exposure to *s*-triazine herbicides.

From an environmental perspective, the development and subsequent spread of enhanced *s*-triazine degradation will require the historic default input values to be changed if accurate fate, transport and risk assessment modeling is desired. For example, when the altered metabolic pathway and reduced persistence estimates for the parent and daughter products in *s*-triazine-adapted soil were accounted for, atrazine's leaching potential was reduced 13.7-fold relative to LEACHM simulations that relied on historic input values.

At completion of this manuscript, several questions remain:

- How widespread is enhanced *s*-triazine degradation?
- Does atrazine provide an economic return on investment when applied to soils exhibiting enhanced degradation?
- What are typical half-life values for atrazine, DEA, DIA and HA as a function of their geographic position, depth in the soil profile and herbicide use history?
- What are regulatory agencies using as estimates for the conversion of atrazine to daughter products, regardless of *s*-triazine use history?
- Will the suggested modeling parameters better predict atrazine fate and transport under actual field conditions?

Given the potential impact of these new metabolic pathways on *s*-triazine efficacy and off-site transport, the authors submit that a large-scale screening study is required to address these concerns.

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