Selection Pressure, Cropping System, and Rhizosphere Proximity Affect Atrazine Degrader Populations and Activity in s-Triazine–Adapted Soil

L. Jason Krutz, Robert M. Zablotowicz, and Krishna N. Reddy*

A field study was conducted on an s-triazine–adapted soil to determine the effects of s-triazine exclusion interval (1, 2, 3, or 4 yr), crop production system (continuous corn or continuous soybean), and rhizosphere proximity (bulk or rhizosphere soil) on atrazine degrader populations and activity. Atrazine degrader populations were quantified by a radiological Most Probable Number technique, while degrader activity was assessed via mineralization of ring-labeled 14C-atrazine. As the s-triazine exclusion interval increased, atrazine degrader populations declined exponentially, regardless of crop or rhizosphere proximity. Crop and exclusion interval interacted to affect degrader populations (P = 0.0043). Pooled over rhizosphere and bulk soil, degrader populations were 1.5-fold higher and declined 2.8-fold faster in soybean than corn. An interaction between rhizosphere proximity and exclusion interval was also noted (P = 0.0021), whereby degrader populations were 1.9-fold higher and declined 2.8-fold slower in rhizosphere compared with bulk soil, regardless of crop. The time required for 50% mineralization of ring-labeled 14C-atrazine (DT50) following exclusion of s-triazine herbicides increased linearly at a rate of 2.2 d yr^-1. In contrast, the DT50 for this site prior to a known s-triazine application was 85 d and declined exponentially over 5 yr of successive atrazine applications: 24.5 d after 1 yr, 10.8 d after two successive years, and 3.8 d after five successive atrazine applications. Omitting s-triazines can reduce degrader populations and activity in adapted soils, but more than 4 yr is required to return mineralization kinetics to nonadapted levels, regardless of crop or rhizosphere proximity.

Nomenclature: Atrazine; s-triazine; corn, Zea mays L.; soybean, Glycine max (L.) Merr.

Key words: persistence, mineralization, atrABCDEF, half-life, trzN.

Atrazine is a soil and foliar applied, s-triazine herbicide that provides residual control of sensitive weeds in corn, sorghum [Sorghum bicolor (L.) Moench], sugarcane (Saccharum officinarum L.), and turf. The herbicide’s residual control of sensitive weeds is a function of the compounds persistence in soil, DT50 approximately 60 d in nonadapted soil (Krutz et al. 2010). Traditionally, it was believed that atrazine’s halogen and N-alkyl substituents impeded microbial degradation of the s-triazine ring thereby contributing to the compound’s moderate persistence in soil and the inability to isolate atrazine-catabolizing bacteria (Wackett et al. 2002). In the mid-1990s, however, atrazine-catabolizing bacteria were isolated, and an enzymatic pathway for atrazine mineralization was subsequently elucidated (Krutz et al., 2010; Mandelbaum et al. 1995; Radosевич et al. 1995).

Today, atrazine-mineralizing bacteria inhabit agricultural soils on all continents except Antarctica, and many of these soils exhibit enhanced degradation (Krutz et al. 2010). 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 pesticide as an energy and nutrient source because of previous exposure to it or an analogue. Moreover, in atrazine-adapted soils, other s-triazine herbicides can also undergo rapid dissipation, a mechanism referred to as cross-adaptation (Krutz et al. 2008; Shaner et al. 2010). Decreased persistence of s-triazine herbicides in adapted soils can reduce the compounds’ residual weed control, as was noted for atrazine and simazine under field and greenhouse conditions (Krut et al. 2007, 2008, 2009). Consequently, a means of extending atrazine persistence and residual weed control in adapted soils is needed.

Three cultural management practices may serve as a means to decrease s-triazine degrader populations or activity in adapted soils: (1) exclude s-triazine herbicides from the weed control program, (2) rotate away from corn production systems, and (3) alter nitrogen dynamics by planting leguminous crops. Excluding s-triazine herbicides from the weed control program may reduce degrader populations and activity because s-triazine herbicides are likely the primary agents for the selection of s-triazine–degrading bacteria (Krutz et al. 2010). Thus, omitting the selective agents may return degrader populations or their activity to background levels, as was noted for other pesticides undergoing enhanced degradation (Anderson and Lafuerza 1992; Arebélí and Fuentes 2007; Roeth 1986; Suet et al. 1993). Second, atrazine-degrader populations, activity, diversity, and survival are greater in the rhizosphere of corn than in bulk soil, which may be unique to this crop (Alvey and Crowley 1996; Krutz et al. 2010; Marchand et al. 2002; Lopez-Gutierrez et al. 2005; Martin-Laurent et al. 2006; Piutti et al. 2002). Rotating from corn to other crops, therefore, may decrease degrader populations or activity. Finally, since nitrogen can suppress the activity of atrazine-degrading bacteria, then higher exogenous N levels in the rhizosphere of leguminous crops may suppress atrazine degrader populations or activity (Zablotowicz et al. 2008).

The objectives of this field study conducted at an s-triazine–adapted location were (1) to determine the effects of s-triazine exclusion time (1, 2, 3, or 4 yr), crop (continuous corn or continuous soybean), and rhizosphere proximity (bulk or rhizosphere soil) on atrazine degrader populations and activity, and (2) to compare atrazine degrader activity from the present study (Phase II, 2007 to 2010) with values observed previously (Phase I, 1999 to 2005) (Zablotowicz et al. 2007).

Materials and Methods

Field Preparations. An 11-yr field study was conducted from 1999 through 2010 at the USDA Crop Production Systems Research Unit farm, Stoneville, MS (33°26′N). The soil was a Dundee silt loam (fine-silty, mixed, thermic Aeric Ochraqualf) with pH 6.7, 1.1% organic matter, a CEC of

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15 cmol, kg\(^{-1}\), and soil textural fractions of 26% sand, 55% silt, and 19% clay. Before initiation of the study, the experimental area was under glyphosate-resistant soybean production and had no known atrazine exposure for at least 20 yr. Field preparation consisted of disking, subsoiling, disk, and bedding in the fall of 1999. The old seedbeds (1-m wide) were raised (rebabeled) in the fall of each year beginning in 2000 after harvest. Beds were smoothed as needed to plant crops in the spring.

In Phase I (i.e., the adaptation period), the field experiment was conducted as a randomized complete block with four replications of each treatment as previously described (Reddy et al. 2006; Zablottlewicz et al. 2007). Briefly, experimental units consisted of eight rows spaced 1 m apart and 45.7 m long. From 2000 through 2005 soils of three cropping systems were evaluated for the development of enhanced atrazine degradation: (1) a continuous conventional corn cropping system that received applications of atrazine each year (CC); (2) a conventional cotton (\textit{Gossypium hirsutum} L.)–corn rotation (1:1) in which plots received applications of atrazine every 2 yr starting in 2001; and (3) a no atrazine history soil from a glyphosate-resistant (GR) corn–cotton rotation system that did not receive an application of atrazine until the sixth year of the study when plots were planted in a conventional corn variety.

During Phase I the experimental area was treated with paraquat (1.1 kg ai ha\(^{-1}\)) 1 to 4 d before planting to control existing weeds. Conventional and GR cultivars of cotton and corn were planted in 1-m-wide rows. Cotton and corn varieties, planting dates, herbicides and application timings, and harvest dates are presented elsewhere (Reddy et al. 2006). A glyphosate-based program in GR cultivars and non–glyphosate-based program in non-GR (conventional) cultivars were used for weed control. Herbicide treatments were applied with a tractor-mounted sprayer with TeeJet 8004 standard flat spray tips (TeeJet 11002 flat-fan nozzles, Spraying Systems Co., Wheaton, IL) delivering 187 L ha\(^{-1}\) water at 179 kPa. Fertilizer application and insect control programs were standard for cotton and corn production (Anonymous 2005; Reddy 2004). Crops were furrow irrigated as needed.

Phase II (i.e., determining the duration of enhanced degradation), was initiated in the fall of 2005 with disking, subsoiling, disking, and bedding of the Phase I experimental area. This was done to break up seedbeds and mix soil to ensure even dispersal of atrazine degrader populations across the experimental area. In the spring of 2006 the beds from the Phase II experimental area were smoothed as needed, planted with GR corn, and treated with 1.82 kg ha\(^{-1}\) of atrazine at planting and 0.95 kg ha\(^{-1}\) as POST. Starting in the spring of 2007 through 2010, all s-triazine applications were omitted and the duration of enhanced degradation was evaluated in two cropping systems, continuous GR corn and continuous GR soybean.

During Phase II the experimental area was treated with paraquat (1.1 kg ai ha\(^{-1}\)) 1 to 4 d before planting to control existing weeds. GR corn and GR soybean cultivars were planted in 1-m-wide rows. Corn and soybean varieties, planting dates, herbicides and application timings, and harvest dates are presented in Table 1. A glyphosate-based program in GR cultivars was used for weed control. The experiment was conducted in a randomized complete block design with four replications. Herbicide treatments were applied with a tractor-mounted sprayer with TeeJet 8004 standard flat spray tips delivering 187 L ha\(^{-1}\) water at 179 kPa. Fertilizer application and insect control programs were standard for corn and soybean production (Reddy et al. 2006). Crops were furrow irrigated as needed.

### Collection of Bulk and Rhizosphere Soils
Composite bulk soil samples were collected from three locations in each plot to a depth of 10 cm. At each location, one sample was collected from in the plant row and one sample was collected from the

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**Table 1. Production practices used in glyphosate-resistant (GR) corn and soybean grown continuously at Stoneville, MS, 2007 to 2010.**

<table>
<thead>
<tr>
<th>Year</th>
<th>Variety</th>
<th>Planting date</th>
<th>PRE (at planting)</th>
<th>EPOST (3–4 WAP)</th>
<th>LPOST/PD (6–7 WAP)</th>
<th>Harvest date</th>
<th>Corn</th>
<th>Soybean</th>
</tr>
</thead>
<tbody>
<tr>
<td>2007</td>
<td>Variety DKC69-72 (RR2)</td>
<td>19 March</td>
<td>Glyphosate</td>
<td>Glyphosate</td>
<td>13 August</td>
<td>P94B73RR</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2008</td>
<td>Variety DKC69-72 (RR2)</td>
<td>21 March</td>
<td>Glyphosate</td>
<td>Glyphosate</td>
<td>18 August</td>
<td>AG4605RR/S</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2009</td>
<td>Variety DKC69-72 (RR2)</td>
<td>23 March</td>
<td>Glyphosate</td>
<td>Glyphosate</td>
<td>25 August</td>
<td>AG4605RR/S</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2010</td>
<td>Variety DKC69-72 (RR2)</td>
<td>24 March</td>
<td>Glyphosate</td>
<td>Glyphosate</td>
<td>16 August</td>
<td>AG4605RR/S</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\textsuperscript{a} Abbreviations: EPOST, early POST; LPOST, late POST, PD, POST directed to base of the corn plant; PRE, pre-emergence; WAP, weeks after planting corn or soybean.

\textsuperscript{b} Rates of herbicides, g ai (ae for glyphosate) ha\(^{-1}\): pendimethalin, 1,120 + metolachlor, 1,120 as PRE and glyphosate, 870 in corn and soybean.

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furrow between the beds. Rhizosphere soil was recovered from roots using a procedure similar to that described elsewhere (Zablótowicz et al. 1997). Approximately 25 roots were excavated from each plot using a shovel to a depth of approximately 20 cm, placed in paper bags, and processed in the laboratory within an hour of collection. Coarse soil aggregates were gently removed by gently shaking and by forceps and discarded. The outer rhizosphere soil was removed using surface sterilized camel hair brushes. Corn rhizospheres were recovered from both the nodal and seminal roots. After recovery of rhizosphere soil from soybean roots, all nodules that fell off were removed using forceps.

**Mineralization of 14C-Ring-Labeled Atrazine.** Mineralization of 14C-ring-labeled atrazine was evaluated in biometer flasks as previously described (Kruz et al. 2007). Briefly, soil (25 g dry weight equivalent) was fortified with a solution of technical grade atrazine (99% purity, Chemservice, Lancaster, PA) and 14C-atrazine (ring-labeled 14C-atrazine, 115 μCi/mmole specific activity, 94% radiological purity, Sigma Chemical Co., St. Louis, Mo.) in deionized water. The initial herbicide concentration was 1 mg L⁻¹, and the initial radioactivity was 86,580 Bq kg⁻¹. Final soil moisture contents were adjusted to 30% moisture (w/w) by addition of deionized water, and biometers were incubated in the dark at 28 ± 2 C. Evolved 14CO₂ was trapped in NaOH and quantified by liquid scintillation spectrometry using Hionic-Fluor (Perkin Elmer, Shelton, CT). To avoid saturation by CO₂, NaOH was replaced after each sampling.

**Most-Probable-Number (MPN) of Atrazine-Mineralizing Bacterial Population.** The MPN of atrazine-mineralizing bacterial populations were estimated for soils using 14C-ring labeled atrazine (0.05 mg L⁻¹ and 2,000 Bq L⁻¹) as previously reported (Zablótowicz et al. 2006), except that the carbon and nitrogen-limited media were based on mineral salts from Vogel's media (Vogel 1964). For each soil MPN, analysis was conducted on four replicate samples. Soils were serially diluted in phosphate buffer, and shell vials containing 800 μl of atrazine media were inoculated with 100 μl of soil dilution (1:10 to 1:10,000 dilution, five replicates per dilution). The shell vials were plugged with sterile polypropylene foam, placed in scintillation vials containing 1.0 ml of 1 N NaOH, and capped with aluminum foil-lined caps. The MPN assemblies were incubated at 25 C for 28 d. Mineralization was determined by removal of the inoculated shell vial, addition of 15 ml of Hi-ionic cocktail and counting by liquid scintillation spectrometry. Estimates of atrazine-mineralizing bacterial populations were calculated using MPN tables (Woomer 1994).

**Soil chemical analysis.** Chemical analysis was conducted on air-dried soil that was passed through a 2-mm sieve and uniformly milled in a Wiley mill. Total organic carbon and total N content were determined on duplicate samples using a Flash EA 1112 elemental analyzer (CE Elantech, Lakewood, NJ). For nitrate-N analysis, soil (2 g) was extracted in 10 ml 0.1 M KCl, for 1 h, clarified by centrifugation, and determined colorimetrically (Cataldo et al. 1975).

**Soil Enzymatic Activity.** Soil hydrolytic enzyme activity was determined using the fluorescein diacetate (FDA) hydrolysis assay. Assays were conducted in triplicate on freshly collected soil as previously described, corrected for a no substrate control, and reported on an oven dry-weight basis (Zablótowicz et al. 2007).

**Statistical Analysis.** Analysis of variance for all evaluated parameters was performed in PROC MIXED (Statistical software package, SAS Institute, Cary, NC). For soil C and N, FDA, and atrazine degrader populations, a split-split-plot design with four replications of each treatment was utilized whereby years of excluding s-triazine herbicides was the whole plot (1 yr, 2 yr, 3 yr, 4 yr), crop production system the subplot (continuous corn or continuous soybean), and rhizosphere proximity the sub-subplot (bulk or rhizosphere). For cumulative 14C-atrazine mineralization, a split-split-plot design with four replications of each treatment was employed in which exclusion interval was the whole plot, crop production system the subplot, rhizosphere proximity the sub-subplot, and sampling time the sub-sub-subplot (0, 2, 4, 7, 11, 14, 18, 23, 28, 32, and 35 d after application of ring-labeled 14C-atrazine). Least squares means were calculated and mean separation (P ≤ 0.05) was produced using PDMIX800 in SAS, which is a macro for converting mean separation output to letter groupings (Saxton 1998).

When interactions with time main effects including s-triazine exclusion interval or sampling time occurred, PROC NLIN was used to generate fitted values for selected models. Four models were required to describe observed results. Equation 1 is a first-order kinetics model that was used to describe four relationships: (1) atrazine degrader populations as a function of crop production system and s-triazine exclusion interval; (2) atrazine degrader populations as a function of rhizosphere proximity and s-triazine exclusion interval; (3) lag phase estimates during Phase I; and (4) the time required for 50% of ring-labeled 14C-atrazine to be mineralized (DT₅₀) during phase I. The form of the first-order kinetics model is depicted in Equation 1.

\[ Y = Ae^{-kt} \]  

where Y is the response variable, A is the maximum, t is the time variable, and k is the rate constant.

Equation 2, the Gompertz growth model, was used to describe two relationships: (1) cumulative mineralization of ring-labeled 14C-atrazine as a function of s-triazine exclusion interval and sampling time, and (2) cumulative mineralization of ring-labeled 14C-atrazine as a function of rhizosphere proximity and sampling time. The general form of Equation 2 is depicted as follows:

\[ y = a \times \exp \{- \exp \left[-\frac{(t - t_0)}{k}\right]\} \]  

where y is the cumulative mineralization of ring-labeled 14C-atrazine (%); a is the plateau representing the maximum mineralization (%); t₀ is the abscissa of the inflection point representing the lag phase (d); k is the inverse of the Gompertz mineralization rate constant (d); and t is time (d).

A linear model, Equation 3, was used to describe four relationships: (1) Gompertz predicted maximum amount of atrazine mineralized during Phase II; (2) Gompertz predicted mineralization rate constant during phase II; (3) Gompertz predicted lag phase during Phase II, and DT₅₀ during Phase II. The form of Equation 3 is as follows:

\[ Y = mx + b \]
An exponential growth model was used to describe changes in Gompertz mineralization rate constant during phase I, Equation 4:

$$Y = A \times [1 - \exp(-k \times t)]$$

where $A$ is the maximum rate constant (d$^{-1}$), $k$ is the first order rate constant, and $t$ is time (yr).

**Results**

**General Soil Biochemical Properties.** Soil C, N, and C : N ratios were affected by an $s$-triazine exclusion interval by rhizosphere proximity interaction, $P \leq 0.0059$ (Figure 1). Within year rhizosphere C, N, and C : N ratio were either greater than or equal to those of bulk soil. Soil C, N, and C : N ratios within rhizosphere and bulk soil were similar over years, and no definitive trend was evident. General microbial activity responded positively to elevated rhizosphere C and concentrations as FDA levels within year were generally higher in rhizosphere than bulk soil, regardless of crop (Figure 2).

**Atrazine Degrader Populations.** The atrazine degrader population declined as $s$-triazine exclusion interval increased, but the trend was different within production system ($P = 0.0043$) and rhizosphere proximity ($P = 0.0021$). Independent of production system or proximity to the rhizosphere, the decline in degrader populations was described by first-order kinetics (Figure 3). Within production system, initial degrader populations were 1.5-fold higher in soybean than corn 1 yr after the exclusion of $s$-triazine herbicides, but the rate in which the degrader population declined over the next 3 yr was 2.8-fold faster in soybean than corn. Regardless of crop, however, degrader populations were initially 1.9-fold higher and declined 2.8-fold slower in rhizosphere compared to bulk soil.

**Atrazine Degrader Activity.** Atrazine degrader activity as estimated by mineralization of ring-labeled $^{14}$C-atrazine varied depending on an $s$-triazine exclusion interval by sampling time interaction ($P < 0.0001$), a rhizosphere proximity by sampling time interaction ($P = 0.0005$), and a three-way interaction among $s$-triazine exclusion interval, crop production system and rhizosphere proximity ($P < 0.0001$). For the sampling time by $s$-triazine exclusion interval and rhizosphere proximity interactions, the Gompertz growth model was used to describe mineralization kinetics (Figure 4). Pooled over crop and rhizosphere proximity, linear regression indicates that the lag phase increases 1.5 d yr$^{-1}$, cumulative mineralization declines 5% yr$^{-1}$ and the mineralization rate constant decreases 0.14 d$^{-2}$ yr as the $s$-triazine exclusion interval increases (Figure 5). Pooled over crop and $s$-triazine exclusion interval, predicted cumulative mineralization was 8% higher in bulk than rhizosphere soil, with no difference in the lag phase or mineralization rate constant between soils (Figure 4). When pooled over sampling time, the three-way interaction among $s$-triazine exclusion interval, cropping system, and rhizosphere proximity indicated differences primarily in degrader activity between corn and soybean rhizosphere soils. For example, cumulative mineralization pooled over sampling time within $s$-triazine exclusion interval was equivalent between corn and soybean bulk soil 3 of 4 yr; conversely, mineralization was greater in the rhizosphere of corn than soybean 3 of 4 yr (Table 2).

**Comparing Mineralization Kinetics between Phase I and Phase II.** Changes in the lag phase, mineralization rate
constant, maximum amount mineralized, and the DT$_{50}$ for bulk soil collected under continuous corn production were described by either first order or linear models (Figures 6 and 7). During the adaptation period, the lag phase declined exponentially at a rate of $2.99 \text{ yr}^{-1}$. Conversely, as the $s$-triazine omission period increased, the lag phase increased linearly at $1.4 \text{ d yr}^{-1}$. The mineralization rate constant for Phase I increased exponentially, $k = 1.20 \text{ yr}^{-1}$, while the rate constant for Phase II declined linearly (Figure 6). Through Phase I the maximum amount mineralized increased linearly at $4.3\% \text{ yr}^{-1}$ ($P = 0.0433$), but during Phase II, the maximum amount mineralization decreased linearly at $4\% \text{ yr}^{-1}$ ($P = 0.0988$). Atrazine DT$_{50}$ decreased exponentially during Phase I ($k = 1.22 \text{ yr}^{-1}$), while the value increased linearly during Phase II, $2.2 \text{ d yr}^{-1}$ (Figure 7).

Discussion

The duration of enhanced degradation is defined as the time required for the mineralization kinetics of a pesticide in an adapted soil to return to those observed prior to adaptation. In this study it was postulated that excluding the application of $s$-triazine herbicides from adapted fields or shifting from corn to a soybean production system would expedite the return of atrazine mineralization kinetics to nonadapted levels. Regardless of the production system evaluated or proximity to the rhizosphere, atrazine mineralization kinetics 4 yr after the last $s$-triazine application still exceeded that observed prior to the soil having received its first $s$-triazine application (Figure 4). The duration of enhanced atrazine degradation, therefore, is in excess of 4 yr regardless of production system or rhizosphere proximity.

In this study, the duration of enhanced atrazine degradation was greater than that reported for thiocarbamate herbicides and organophosphorus insecticides, i.e., ≤ 2 yr, but was within the range reported for carbamate insecticides, i.e., ≤ 5 yr (Anderson and Lafuerza 1992; Arbeli and Fuentes 2007; Roeth 1986; Suett et al. 1993). As was the case with the carbamates, the duration of enhanced atrazine degradation clearly exceeded 4 yr, making it impractical to determine an absolute duration of enhanced degradation; that is, the experiment would need to be conducted for decades. Thus, a predicted value for the duration of enhanced atrazine degradation was calculated.
The duration of enhanced atrazine degradation was calculated by modeling changes in the DT$_{50}$ values during Phase I and Phase II. Specifically, the DT$_{50}$ prior to adaption was divided by the rate at which the DT$_{50}$ value increased during the s-triazine omission period; that is, 85 d divided by 2.2 $\pm$ 0.6 d yr$^{-1}$. Note that ± 0.6 d yr$^{-1}$ is the ± 95% confidence interval for the rate constant. Given that the initial estimate for DT$_{50}$ prior to adaptation is reasonable and that changes in the DT$_{50}$ values remain linear as s-triazine omission continues into the future, then s-triazine herbicides must be omitted 39 ± 14.1 yr before DT$_{50}$ returns to pre-adapted levels (Figure 7). The projected duration of enhanced atrazine degradation indicates that the microbial population or the genes coding for enhanced atrazine degradation, i.e., $atztetD$ or $trzNDF$, are stable for decades in continuous corn and soybean production systems, even when the selective agent is omitted (Krutz et al. 2010).

The stability of enhanced atrazine degradation in corn and soybean systems is surprising given the exponential decline in degrader populations over the 4-yr exclusion interval. Excluding s-triazine applications from adapted fields reduces degrader populations exponentially over time, but the reduction in degrader populations has only a minimal effect on their activity. For example, pooled over crop and rhizosphere proximity, atrazine degrader populations declined 20-fold. Concurrently, the mineralization lag phase, maximum mineralized and mineralization rate constant changed 3.3-fold, 1.2-fold, and 1.7-fold, respectively. It appears that...
when s-triazines are not applied to adapted fields that degrader populations quickly decline to a low but static level of bacteria that are nonculturable using our MPN procedure but are able to rapidly respond to an additional s-triazine application (Rhine et al. 2003). Moreover, our predicted duration of enhanced degradation indicates that a small, static atrazine degrader population can respond to an s-triazine application for decades after this class of chemistry has been omitted from the weed control program.

In summary, results from this study indicate that once a field develops a microbial population that is able to rapidly mineralize s-triazine herbicides, the duration of enhanced degradation exceeds any practical herbicide or crop rotation program, i.e. 4 yr. Switching to a weed control program or crop production system that does not include the use of s-triazine herbicide or corn will not significantly increase atrazine persistence for at least 4 yr. Although enhanced s-triazine degradation is widespread and can reduce residual weed control, neither yield loss nor complete weed control failure in adapted soils has been reported to date (Krutz et al.

Table 2. Cumulative $^{14}$CO$_2$ evolved pooled over sampling time for rhizosphere and bulk soil collected from continuous corn and continuous soybean plots located in Stoneville, MS. LSD$_{0.05}$(Year $\times$ Crop $\times$ Soil) = 2.8.

<table>
<thead>
<tr>
<th>Year</th>
<th>Corn Rhizosphere</th>
<th>Corn Bulk</th>
<th>Soybean Rhizosphere</th>
<th>Soybean Bulk</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>70.6</td>
<td>74.9</td>
<td>73.1</td>
<td>65.1</td>
</tr>
<tr>
<td>2</td>
<td>66.2</td>
<td>65.5</td>
<td>59.6</td>
<td>61.9</td>
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<td>3</td>
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<td>55.5</td>
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<tr>
<td>4</td>
<td>43.2</td>
<td>44.4</td>
<td>46.9</td>
<td>43.3</td>
</tr>
</tbody>
</table>

Figure 6. Changes in Gompertz fitted parameters including maximum atrazine mineralization, lag phase, and mineralization rate constant over Phase I and Phase II of the enhanced s-triazine degradation study: Phase I is the adaptation period where s-triazine herbicides were applied five consecutive years, while Phase II is the period in which s-triazine applications were omitted four consecutive years. Symbols represent the mean of four bulk soil replicates collected from continuous corn plots, and error bars denote one standard deviation. The Gompertz model is described by the function $y = a \times \exp\left[-\exp\left(-(t - t_0)/k\right)\right]$, where $a$ is the plateau representing the maximum mineralization (%); $t_0$ is the abscissa of the inflection point representing the lag phase (d); $k$ is the inverse of the mineralization rate constant (d); and $t$ is time (d). Fitted values are as follows:

Phase I, maximum mineralized: $y = 4.3086x + 46.7379$

Phase I, rate constant: $y = 0.6336 \times (1 - \exp^{-1.1978})$

Phase II, maximum mineralized: $y = -3.9959x + 82.3850$

Phase II, rate constant: $y = -0.1120x + 0.8850$

Phase I, lag phase: $y = 64.9959 \times \exp^{-2.9912x}$

Phase II, lag phase: $y = 1.3600x + 0.5000$
Phase I. 5-triazine applied five consecutive years

Phase II. 5-triazines omitted four consecutive years

Figure 7. Changes in the time required for 50% mineralization of ring-labeled 14C-atrazine (DT50) as estimated by the best fit of the Gompertz model to mineralization data from bulk soil collected from continuous corn during Phase I and Phase II of the enhanced 5-triazine degradation study: Phase I is the adaptation period where 5-triazine herbicides were applied five consecutive years, while Phase II is the period in which 5-triazine applications were omitted four consecutive years. Symbols represent the mean of four replicates and error bars denote one standard deviation. The Gompertz model is described by the function \( y = a \times \exp\left(\exp\left(-\frac{(t-t_0)/k}{2}\right)\right) \), where \( a \) is the plateau representing the maximum mineralization (%); \( t_0 \) is the abcissa of the inflection point representing the lag phase (d); \( k \) is the inverse of the mineralization rate constant (d); and \( t \) is time (d). Fitted values are as follows:

- Phase I, Atrazine DT50: \( y = 89.7061 \times \exp^{-1.2226} \)
- Phase II, Atrazine DT50: \( y = 2.1700x + 0.5000 \)

2007, 2009, 2010). The continued use of 5-triazine herbicides at adapted locations is still warranted, but to decrease the risk of reduced residual weed control, these chemistries should be applied POST in combination with other modes of action.

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Mention of trade names or commercial products in this publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

Literature Cited


Arbeli, Z. and C. L. Fuentes. 2007. Accelerated biodegradation of pesticides: an overview of the phenomenon, its basis and possible solutions; and a discussion on the tropical dimension. Crop Prot. 26:1733–1746.


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