Glyphosate: 
Herbicide Interactions, 
Microbial Activity, and 
Community Shifts

Scott A. Senseman
Department of Soil and Crop Sciences
Texas A&M University
College Station, TX 77843-2474
Glyphosate use in the U.S.

- Agricultural glyphosate use (Roundup) increased nearly 50% between 1995 and 1997.
- Glyphosate is applied to more than 31% of corn, 71% of cotton, and 88% of soybean.

Glyphosate characteristics

- Chemical name: N-phosphonomethyl glycine
- Non-selective herbicide
- Enolpyruvyl Shikimate-3-Phosphate Synthase (EPSPS) - Key enzyme responsible for the aromatic amino acids
- Rapidly degraded by soil microbes
- Highly adsorptive to soil

![Glyphosate acid structure](image-url)
Effect of glyphosate on soil microbial activity and biomass

R. L. Haney
Corresponding author. Department of Soil & Crop Sciences, Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843-2474; rhaney@acs.tamu.edu

S. A. Senseman
F. M. Hons
D. A. Zuberer
Department of Soil & Crop Sciences, Texas Agricultural Experiment Station, Texas A&M University, College Station, TX 77843-2474

Herbicides applied to soils potentially affect soil microbial activity. Quantity and frequency of glyphosate application have escalated with the advent of glyphosate-tolerant crops. The objective of this study was to determine the effect of increasing glyphosate application rate on soil microbial biomass and activity. The soil used was Weswood silt loam. The isopropylamine salt of glyphosate was added at rates of 47, 94, 140, and 234 μg ai g⁻¹ soil based on an assumed 2-mm glyphosate-soil interaction depth. Glyphosate significantly stimulated soil microbial activity as measured by C and N mineralization but did not affect soil microbial biomass. Cumulative C mineralization, as well as mineralization rate, increased with increasing glyphosate rate. Strong linear relationships between mineralized C and N and the amount of C and N added as glyphosate ($r^2 = 0.995, 0.996$) and slopes approximating one indicated that glyphosate was the direct cause of the enhanced microbial activity. An increase in C mineralization rate occurred the first day following glyphosate addition and continued for 14 d. Glyphosate appeared to be directly and rapidly degraded by microbes, even at high application rates, without adversely affecting microbial activity.
Carbon Mineralization

- Gas tight 1-L glass containers
- Ten mL 1 M KOH
- Humidity maintained by vial of water
- Traps replaced at regular intervals

Anderson, 1982
Figure 1. Effect of glyphosate rate on (a) carbon mineralization and (b) nitrogen mineralization from soil after 56 d of incubation. The 1, 2, 3, and 5× represent glyphosate addition rates of 47, 94, 140, and 234 µg ai g⁻¹ soil, respectively. Error bars indicate one standard deviation.
Figure 5. Relationship of carbon and nitrogen mineralized from soil in 56 d following glyphosate addition. Error bars indicate one standard deviation.
**Figure 4.** Relationship of carbon added from glyphosate and carbon mineralized from soil in 56 d following glyphosate addition. Controls have been subtracted. Error bars indicate one standard deviation.
Bioremediation and Biodegradation

Effect of Roundup Ultra on Microbial Activity and Biomass from Selected Soils

R. L. Haney, S. A. Senseman,* and F. M. Hons

ABSTRACT
Herbicides applied to soils potentially affect soil microbial activity. The quantity and frequency of Roundup Ultra [RU; N-(phosphonomethyl)glycine; Monsanto, St. Louis, MO] applications have escalated with the advent of Roundup-tolerant crops. The objective of this study was to determine the effect of Roundup Ultra on soil microbial biomass and activity across a range of soils varying in fertility. The isopropylamine salt of glyphosate was applied in the form of RU at a rate of 234 mg active ingredient kg⁻¹ soil based on an assumed 2-mm glyphosate–soil interaction depth. Roundup Ultra significantly stimulated soil microbial activity as measured by C and N mineralization, as well as soil microbial biomass. Cumulative C mineralization as well as mineralization rate increased above background levels for all soils tested with addition of RU. There were strong linear relationships between C and N mineralized, as well as between soil microbial C and N (r² = 0.96 and 0.95, respectively). The slopes of the relationships with RU addition approximated three. Since the isopropylamine salt of glyphosate has a C to N ratio of 3:1, the data strongly suggest that RU was the direct cause of the enhanced microbial activity. An increase in the C mineralization rate occurred the first day following RU addition and continued for 14 d. Roundup Ultra appeared to be rapidly degraded by soil microbes regardless of soil type or organic matter content, even at high application rates, without adversely affecting microbial activity.

includes surfactant and other inert products) has on indigenous microbial populations and activities across a range of soils varying in fertility.

Although glyphosate is not intentionally soil applied, a significant concentration of material may reach the soil surface during broadcasted preplant or early-season applications. The amount of herbicide available to soil microorganisms depends on various factors, including available nutrients, pH, temperature, and moisture, though they differ in importance depending on the pesticide involved (Weber et al., 1993). Soil water and temperature directly affect many biological processes, including plant metabolism and microbial degradation, and thereby influence bioactivity and persistence of the chemicals (Weber et al., 1993).

Dick and Quinn (1995) investigated 26 bacterial strains from sites without prior addition of glyphosate and found that all 26 could metabolize glyphosate via the initial cleavage of its carbon–phosphorus bond. Since glyphosate contains carbon, nitrogen, and phosphorus, all of which are essential nutrients for soil microorganisms, it should be readily mineralizable.

In our previous work, we found that increasing rates
<table>
<thead>
<tr>
<th>Location</th>
<th>Series</th>
<th>Texture</th>
<th>USDA soil classification</th>
<th>Soil pH (water)</th>
<th>Clay content</th>
<th>Organic C g kg⁻¹</th>
<th>Depth of sampling cm</th>
<th>Land management</th>
</tr>
</thead>
<tbody>
<tr>
<td>Waynesboro, GA</td>
<td>Lakeland</td>
<td>S</td>
<td>thermic, coated Typic Quartzipsammament</td>
<td>4.7</td>
<td>10.1</td>
<td>5.9</td>
<td>30–60</td>
<td>cropped (cotton, soybean)</td>
</tr>
<tr>
<td>Watkinsville, GA</td>
<td>Cecil</td>
<td>SL</td>
<td>fine, kaolinitic, thermic Typic Kanhapludult</td>
<td>4.8</td>
<td>12.8</td>
<td>15.0</td>
<td>0–15</td>
<td>pasture (bermudagrass)</td>
</tr>
<tr>
<td>Amarillo, TX</td>
<td>Pullman</td>
<td>SCL</td>
<td>fine, mixed, superactive, thermic Torrertic Paleustoll</td>
<td>5.7</td>
<td>28.7</td>
<td>11.6</td>
<td>0–7.5</td>
<td>cropped (sorghum, wheat)</td>
</tr>
<tr>
<td>Overton, TX</td>
<td>Bowie</td>
<td>fSL</td>
<td>fine-loamy, siliceous, semiactive, thermic Plinthic Paleudult</td>
<td>6.3</td>
<td>6.0</td>
<td>4.1</td>
<td>0–7.5</td>
<td>pasture (bermudagrass)</td>
</tr>
<tr>
<td>Stephenville, TX</td>
<td>Windthorst</td>
<td>fSL</td>
<td>fine, mixed, thermic Udic Paleustalf</td>
<td>6.4</td>
<td>13.0</td>
<td>18.3</td>
<td>0–7.5</td>
<td>pasture (bermudagrass)</td>
</tr>
<tr>
<td>Watkinsville, GA</td>
<td>Pacolet</td>
<td>SCL</td>
<td>fine, kaolinitic, thermic Typic Kanhapludult</td>
<td>6.6</td>
<td>26.9</td>
<td>52.3</td>
<td>0–5</td>
<td>pasture (tall fescue)</td>
</tr>
<tr>
<td>Malone, TX</td>
<td>Houston Black</td>
<td>C</td>
<td>very-fine, smectitic, thermic Oxyaquic Hapludert</td>
<td>7.8</td>
<td>45.0</td>
<td>13.7</td>
<td>0–10</td>
<td>cropped (sorghum, wheat)</td>
</tr>
<tr>
<td>College Station, TX</td>
<td>Weswood</td>
<td>SiCL</td>
<td>fine-silty, mixed, superactive, thermic Udifluventic Ustochoerpt</td>
<td>8.0</td>
<td>28.4</td>
<td>23.7</td>
<td>0–7.5</td>
<td>pasture (bermudagrass)</td>
</tr>
<tr>
<td>Weslaco, TX</td>
<td>Hildalgo</td>
<td>SCL</td>
<td>fine-loamy, mixed, hyperthermic Typic Calciustoll</td>
<td>8.2</td>
<td>22.3</td>
<td>9.8</td>
<td>0–7.5</td>
<td>cropped (cotton, corn)</td>
</tr>
</tbody>
</table>

† S, sand; SL, sandy loam; SCL, sandy clay loam; fSL, fine sandy loam; C, clay; SiCL, silty clay loam.
Fig. 2. Effect of Roundup Ultra on daily carbon mineralization between 1 and 7 d from different soils with (A) 0.2 to 0.5% organic C, (B) 0.6 to 0.8% organic C, and (C) 0.9 to 2.6% organic C. Error bars indicate one standard deviation.
Fig. 5. Mineralization for (A) carbon and (B) nitrogen, after 50 d of incubation for nine soils. Error bars indicate one standard deviation.
Fig. 1. Carbon dioxide evolution from soil after applications of Roundup Ultra (product; applied at $3 \times$ field rate) and analytical-grade glyphosate, and a control.
R. L. Haney · S. A. Senseman · L. J. Krutz · F. M. Hons

**Soil carbon and nitrogen mineralization as affected by atrazine and glyphosate**

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**Abstract** Atrazine alone and atrazine plus glyphosate were added to soil to determine their effect on soil microbial activity as measured by C and N mineralization (Cmin, Nmin) and soil extractable atrazine without the use of radiolabelled isotopes. Atrazine alone was added to soils as a formulated product (Aatrex 4L) at a field rate of 2× (94 mg kg⁻¹), 4× (188 mg kg⁻¹), and 6× (282 mg kg⁻¹) with an assumed soil penetration depth of 58 mm. Glyphosate, as Roundup Ultra, was added along with atrazine to soil in equal amounts bringing the total

**Introduction**

Atrazine and glyphosate are effective and widely used herbicides. Tank mixing atrazine and glyphosate is used for immediate burndown and residual weed control especially in corn (*Zea mays* L.) and sorghum (*Sorghum bicolor* L. Moensch) production. Glyphosate is a non-selective, foliar-applied herbicide used to control weeds preplant or post-emergence in tolerant crops or by using shielded sprayers in non-tolerant crops. Glyphosate’s
Fig. 1 Soil C mineralization over time as affected by herbicide addition (2×, 4×, 6× rate of herbicide additions, A atrazine, A + G atrazine plus glyphosate). The “0” line on the y-axis represents the control with no herbicide addition, which has been subtracted from each data point. The LSD of 3.5 represents the least significant difference between treatments at a given time interval.
Fig. 3  C and N added as atrazine only and atrazine plus glyphosate versus C mineralized in 56 days (a), and N mineralized in 56 days (b)
Fig. 6 Extractable atrazine during 56 days of incubation. Error bars indicate one standard deviation (Atra-Gly atrazine-glyphosate)
Short Communication

Effect of Roundup Ultra on atrazine degradation in soil

L. J. Krutz¹, S. A. Senseman¹ and R. L. Haney²

¹ Department of Soil and Crop Sciences, Texas Agricultural Experiment Station, Texas A&M University, College Station, TX, 77843-2474, USA
² United States Department of Agriculture, Southern Plains Area, Grassland, Soil and Water Research Laboratory, Temple, TX, USA

Email: krutz1@msn.com
Phone: +1-979-8455384
Fax: +1-979-8450458

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Abstract Tank mixing pesticides and the use of pre-packaged mixtures have become common agricultural practices. However, pesticide degradation in multi-pesticide systems is rarely evaluated. The objective of this laboratory study was to determine the effect of Roundup Ultra on atrazine degradation in soil. Based on a 2-mm glyphosate-soil interaction depth, the isopropylamine salt of glyphosate was added to Aatrex-amended and non-amended soil at rates of 0, 1 (43 mg ai kg⁻¹), 2, 3, 4, and 5×. Treatments were incubated for 4, 8, 12, 16, 20, 24, 28, and 32 days. Atrazine degradation was significantly different among treatments at 8 days. In the 0× treatment (Aatrex only), 87% of the atrazine was degraded. During the same 8-day period, atrazine degradation in the 1, 2, 3, 4, and 5× treatments was 77%, 69%, 60%, 61%, and 52%, respectively. Atrazine degradation approached 97% for all treatments after 12 days and statistical differences were no longer observed. Atrazine degradation was inversely correlated with Roundup Ultra rate and microbial activity at 8 (r²=0.97) and 12 days (r²=0.92). These results indicate that Roundup Ultra stimulated microbial activity while simultaneously inhibiting atrazine degradation.
Fig. 1 Effect of Roundup Ultra rate on atrazine degradation from soil during 32 d of incubation. Bars indicate LSD (0.05) among treatments at a single sampling time.
Soil Microbial Activity Is Affected by Roundup WeatherMax and Pesticides Applied to Cotton (Gossypium hirsutum)

Sarah H. Lancaster,* ‡ Richard L. Haney,‡ Scott A. Senseman,† Frank M. Hons,‡ and James M. Chandler‡

Texas Agricultural Experiment Station, Texas A&M University, 2474 TAMU College Station, Texas 77843-2474, and Agricultural Research Service, U.S. Department of Agriculture, 808 East Blackland Road, Temple, Texas 76502
Table 3. Pesticide Treatments Applied to Pasture and Cultivated Field Soil\textsuperscript{a}

<table>
<thead>
<tr>
<th>pesticide\textsuperscript{b}</th>
<th>interaction depth (mm)</th>
<th>concentration\textsuperscript{c,d} ((\mu)g active ingredient/kg soil)</th>
</tr>
</thead>
<tbody>
<tr>
<td>fluometuron</td>
<td>37.5</td>
<td>4.1</td>
</tr>
<tr>
<td>trifluralin</td>
<td>37.5</td>
<td>2.0</td>
</tr>
<tr>
<td>aldicarb</td>
<td>37.5</td>
<td>10.2</td>
</tr>
<tr>
<td>mefenoxam + PCNB</td>
<td>37.5</td>
<td>8.3 + 0.42</td>
</tr>
<tr>
<td>glyphosate</td>
<td>1.5</td>
<td>152.7</td>
</tr>
<tr>
<td>glyphosate + fluometuron</td>
<td>1.5 + 37.5</td>
<td>152.7 + 4.1</td>
</tr>
<tr>
<td>glyphosate + trifluralin</td>
<td>1.5 + 37.5</td>
<td>152.7 + 2.0</td>
</tr>
<tr>
<td>glyphosate + aldicarb</td>
<td>1.5 + 37.5</td>
<td>152.7 + 10.2</td>
</tr>
<tr>
<td>glyphosate + mefenoxam + PCNB</td>
<td>1.5 + 37.5</td>
<td>152.7 + 8.3 + 0.42</td>
</tr>
<tr>
<td>untreated</td>
<td></td>
<td>0</td>
</tr>
</tbody>
</table>

\textsuperscript{a} Soil was Weswood clay loam collected from a bermuda grass pasture and a fallow field previously planted with cotton. \textsuperscript{b} Pesticides were applied as formulated products: glyphosate, Roundup WeatherMax; fluometuron, Cotoran 4L; trifluralin, Treflan HFP; aldicarb, Temik 15G; and mefenoxam + pentachloronitrobenzene (PCNB), Ridomil Gold PC GR. \textsuperscript{c} Rate of application of formulated product: Cotoran 4L, 4.67 L/ha; Treflan HFP, 2.34 L/ha; Temik 15G, 5.62 kg/ha; Ridomil Gold OC GR, 11.23 kg/ha; and Roundup WeatherMax, 7.01 L/ha. \textsuperscript{d} Calculations of concentration assume that the mass of a 15 cm furrow slice is 2200000 kg.
Figure 2. Nitrogen mineralized during 30 days of incubation in all pesticide treatments with and without glyphosate. Bars represent the least significant difference ($\alpha = 0.10$) of 5.6 ($P = 0.0114$).
Figure 3. Cumulative C and N mineralized in 30 days after application of pesticides (A, aldicarb; F, fluometuron; M, mfenoxam + pentachloronitrobenzene (PCNB); T, trifluralin; and U, untreated) alone or with glyphosate (G).
Microbial Degradation of Fluometuron Is Influenced by Roundup WeatherMAX

SARAH H. LANCASTER,*-† RICHARD L. HANEY,‡ SCOTT A. SENSEMAN,† CHARLES M. KENERLEY,§ AND FRANK M. HONS†

Texas AgriLife Research, Department of Soil and Crop Sciences, Texas A&M University, 2474 TAMU, College Station, Texas 77843-2474, Department of Plant Pathology and Microbiology, Texas A&M University, 413C L. F. Peterson Building, College Station, Texas 77843, and Agricultural Research Service, U.S. Department of Agriculture, 808 East Blackland Road, Temple, Texas 76502

Laboratory experiments were conducted to describe the influence of glyphosate and fluometuron on soil microbial activity and to determine the effect of glyphosate on fluometuron degradation in soil and by Rhizoctonia solani. Soil and liquid medium were amended with formulated fluometuron alone or with two rates of formulated glyphosate. The soil carbon mineralization was measured hourly for 33 days. The fluometuron remaining in the soil was quantified following 3, 6, 10, 15, 20, 30, and 40 days of incubation. The fluometuron remaining in medium and fungal biomass was measured after 1, 3, 6, 10, 15, and 20 days of incubation. The addition of glyphosate with fluometuron increased C-mineralization and increased the rate of fluometuron degradation relative to fluometuron applied alone. However, more fluometuron remained in the media and less fungal biomass was produced when glyphosate was included.

KEYWORDS: Cotton; degradation; glyphosate; Gossypium hirsutum; microbial activity; fluometuron; Rhizoctonia solani
Figure 2. Fitted equations representing cumulative C mineralization 793 h (33 days) following the addition of 2.25 kg ai/ha fluometuron (—), 2.25 kg fluometuron + 1.25 kg ae/ha glyphosate (— — —), 2.25 kg fluometuron + 2.5 kg ae/ha glyphosate (— —), or no herbicide (— — —). Equation parameters are listed in Table 2.
Figure 3. First-order rate plots for degradation of fluometuron applied alone (2.25 kg ai/ha; black diamond) and with 1.25 kg ae/ha glyphosate (gray square) or 2.5 kg ae/ha glyphosate (gray circle). Fitted equations are as follows: $y = -0.03x + 0.13$, fluometuron alone; $y = -0.03x + 0.17$, fluometuron + 1× rate of glyphosate; and $y = -0.03x - 0.001$, 2× rate of glyphosate.
Table 4. First-Order Rate Constant (k), Half-Life (t1/2), and Coefficient of Determination (R^2) of Fluometuron in Soils Treated with Fluometuron Alone or with Glyphosate\textsuperscript{a}

<table>
<thead>
<tr>
<th>treatment</th>
<th>k (days\textsuperscript{-1})</th>
<th>t\textsubscript{1/2} (days)</th>
<th>R\textsuperscript{2}</th>
</tr>
</thead>
<tbody>
<tr>
<td>fluometuron</td>
<td>0.025 a</td>
<td>28.6 a</td>
<td>0.81</td>
</tr>
<tr>
<td>fluometuron + 1\times glyphosate</td>
<td>0.026 a</td>
<td>26.9 ab</td>
<td>0.71</td>
</tr>
<tr>
<td>fluometuron + 2\times glyphosate</td>
<td>0.033 b</td>
<td>21.2 b</td>
<td>0.92</td>
</tr>
</tbody>
</table>

\textsuperscript{a} Values within a column followed by different letters are significantly different at P ≤ 0.05 according to Tukey’s multiple pairwise comparisons. \textsuperscript{b} Fluometuron was applied at a concentration of 2.25 kg ai ha\textsuperscript{-1} (3.5 μg ai g soil\textsuperscript{-1}) and glyphosate was applied at concentrations of 1.25 and 2.5 kg ae ha\textsuperscript{-1} (49.7 and 99.5 μg ae g soil\textsuperscript{-1}).
Figure 4. *R. solani* biomass accumulated following 20 days of growth in minimal media containing 20 g glucose, 1 g NH₄NO₃, 0.9 g K₂HPO₄, 0.2 g KCl, 0.2 g MgSO₄·7H₂O, 0.002 g FeSO₄·7H₂O, 0.002 g ZnSO₄·H₂O, and 0.002 g MnCl₂ per L amended with 11.7 µg mL⁻¹ fluometuron alone and with 146 or 292 µg glyphosate mL⁻¹. Bars labeled with similar letters are similar according to pairwise t tests (α = 0.05).
Effects of repeated glyphosate applications on soil microbial community composition and the mineralization of glyphosate

Sarah H. Lancaster*, Emily Hollister, Scott A. Senseman, Terry J. Gentry

Department of Soil and Crop Sciences, Texas A&M University System, College Station, TX 77843, USA

*Correspondence to: Sarah Lancaster, current address: 368 Agriculture Hall, Oklahoma State University, Stillwater, OK 74078; tel: 405-744-3525; fax: 405-744-5269; email: sarah.lancaster@okstate.edu.
Figure 2. Mineralization of glyphosate in soil: 1 application —■--; 2 applications —□--; 3 applications —●--; 4 applications —○--; 5 applications —▲—. Lines fitted to first-order kinetics model: $Y = a(1-e^{kt})$. 

$^{14}$CO$_2$ evolved (% applied)
NMS analysis of FAMEs extracted from soil 3, 7, and 14 DAA

Numbers in parentheses represent the percent variance represent by each axis. 1 glyphosate application, □; 5 glyphosate applications, ▲.
Conclusions from research with glyphosate

- Glyphosate is rapidly and directly degraded by soil microbes even at high concentrations regardless of soil characteristics without adversely affecting microbial activity.
- Roundup Ultra stimulated microbial activity while appearing to inhibit atrazine degradation.
Conclusions from research with glyphosate

• Addition of Roundup WeatherMax reduced C mineralization in soils treated with fluometuron, aldicarb, or mefenoxam+PCNB formulations. Therefore, may alter soil microbial response to other pesticides.

• The addition of glyphosate with fluometuron increased C-mineralization and increased the rate of fluometuron degradation relative to fluometuron applied alone. However, less fungal biomass was produced when glyphosate was included.
Conclusions from research with glyphosate

• Changes in dissipation or distribution of glyphosate following repeated applications of glyphosate may be related to shifts in soil microbial communities.
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