Conducting a Bioassay for Herbicide Residues

by Joseph C. Neal

What is a bioassay?

A bioassay is a technique for determining if herbicide (or other chemical) residues are present in soil or water in high enough concentrations to adversely affect plant growth. This is a simple and direct method to determine if it is safe to seed or plant into areas previously treated with herbicides or into soil with an unknown history of herbicide use.

In its simplest form, a bioassay uses susceptible plants to identify if the herbicide is present in concentrations high enough to inhibit germination and/or growth. However, scientists sometimes use sensitive bioassays to estimate herbicide concentrations in soil and water, and identify unknown herbicide residues from the symptoms of injury.

When is a bioassay warranted?

When newly seeded or established plants show seemingly unexplained symptoms of injury, stress, or decline. Also, when seeding or planting into areas previously treated with residual herbicides, such as those applied for crabgrass control in turf. Top soil from abandoned farmland can often contain herbicide residues, particularly atrazine, which can injure many plants. Additionally, if you suspect that another product may have been contaminated with a herbicide, both the product and treated soil can be tested.

Table 1. Some recommended bioassay species for herbicides and the expected injury symptoms.

<table>
<thead>
<tr>
<th>Herbicides</th>
<th>Recommended Test Species</th>
<th>Expected Symptoms</th>
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</thead>
<tbody>
<tr>
<td>Acetanalides (Dual®, Lasso®, Pennant*)</td>
<td>Oat, ryegrass</td>
<td>Stunting, malformed leaves. White (not yellow) leaves.</td>
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<tr>
<td>Amitrol</td>
<td>Oat, cucumber, tomato</td>
<td>Stunting, swollen and shortened roots.</td>
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<tr>
<td>Dinitroanilines (Balan®, Treflan®, pendimethalin, others)</td>
<td>Oat, ryegrass, cucumber</td>
<td>Swollen roots, stunted plants. Reduced emergence. If plants emerge, roots are swollen and stunted.</td>
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<tr>
<td>Isoxaben (Gallery®)</td>
<td>Cucumber</td>
<td></td>
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<td></td>
<td>Mustard, chinese cabbage</td>
<td></td>
</tr>
<tr>
<td>Oxadiazon (Ronstar®)</td>
<td>Oat, ryegrass, tomato</td>
<td>Stunted shoot growth, roots less affected. Foliage necrotic where contacted by herbicide treated soil.</td>
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<tr>
<td>Sulfonylureas and imidazolinones (Glean®, Oust®, Lesco TFC®, Pursuit®, Arsenal®, others)</td>
<td>Tomato, cucumber, spinach</td>
<td>Stunting and general yellowing of the new growth.</td>
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<tr>
<td>Triazines (Atrazine, simazine, others)</td>
<td>Oats</td>
<td>Stunting, yellow leaves.</td>
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<tr>
<td></td>
<td>Cucumber, tomato</td>
<td>Stunting, interveinal yellowing of new leaves (starting with about the third true leaf).</td>
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<tr>
<td>Synthetic auxins (Banvel®, MCPP®, 2,4-D®, Turflon®, Picloram®, others)</td>
<td>Cucumber, tomato</td>
<td>Malformed, twisted shoot growth.</td>
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</table>
How to conduct a bioassay.

1. Collect representative soil samples.
   a. Sample from areas suspected of having herbicide residues as well as areas which are known to be free of herbicides. You will use the herbicide-free soil for comparison.
   b. Take separate samples from high spots, low spots, and different soils. Also sample areas where sprayer overlap could have resulted in an over-dose.
   c. Take soil cores. Remove the thatch and keep only the upper two inches of soil. Most residual herbicides will be bound in the upper two inches of soil. On sandy soils, sample to four inches.
   d. Take several samples from an area and combine them. You need enough soil to fill several pots in which you will grow the bioassay plants (I suggest 3- to 4-inch pots).

2. Select the bioassay species.

   In general, the best bioassay species is the one you intend to grow. However, crop plants and turfgrasses sometimes do not grow well indoors in pots, nor do they respond rapidly or decisively enough to be reliable bioassay species. Therefore, it is often advisable to select particular species known to perform well in bioassays such as ryegrass, oats, cucumber, and tomato. Table 1 provides a list of recommended bioassay species for different herbicide residues.

3. Seed and grow for about three weeks.

   Seed the bioassay species in both "clean" and "contaminated" soil. Place the pots in a greenhouse or on a sunny window sill and keep them watered (do not waterlog). Watch the plants for about three weeks.

4. Evaluate plant growth.
   a. Oats in "clean" soil should be about four inches tall when you evaluate the plants. Cucumbers and other broadleaf indicator plants should have three true leaves (not counting the seed leaves).
   b. Examine overall growth, as well as the leaves, and roots. Look for stunting, yellowing (or other discoloration), abnormal leaf or stem growth, and root swelling or stunting.

What to do if herbicide residues are present?

There are basically three options:

1. Leave the soil fallow (or stockpile top soil) for one growing season before planting (in turfgrass areas this is generally not feasible);

2. Plant another species which is tolerant of the herbicide, such as selecting a different turfgrass species or installing a woody ground cover bed; or

3. Incorporate (rototill) activated carbon into the soil to a depth of six inches. The recommended amount to detoxify herbicide residues is 100 lb. activated carbon per acre for every pound of herbicide active ingredient (AI) per acre suspected to be present. After incorporating activated carbon, run the bioassay again to confirm detoxification.

If option three is chosen, be aware that activated carbon does not detoxify all herbicide residues. You may, therefore, wish to run a small test in pots to determine whether the activated carbon will effectively detoxify the herbicide residues. Mix 1/2 ounce (dry measure) of activated carbon in 1 quart of water. Add 1 fluid ounce of this to each 4 inch pot of soil. [This will approximate an application of 600 lb. activated carbon per acre.] Dump the soil in a bag and mix well; then return the soil to the pot and run the bioassay. If the plants grow well, proceed with the application of activated carbon to the field. If the plants are still stunted, contact your local Cooperative Extension office for assistance.

A bioassay is a simple, inexpensive, and accurate way to determine if herbicide residues are present in high enough concentrations to affect seedling emergence or plant growth. By conducting a bioassay on new top soil or in new seedings previously treated with a herbicide, you may avoid wasted time and seed, thus saving you time and money in the long run.

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