

Herbicides

And Amphibian Decline

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What Are Herbicides?

- Herbicides are used to control unwanted vegetation by farmers, foresters, managers, and everyday people
- In 2007 Herbicides accounted for 40% of pesticide use in the US and the world- 2,096 million lbs worldwide, 531 million US (EPA 2007)
- Herbicides of some sort are found in 57% of US streams (Biello 2008)
- Many categories and modes of action

How do They Harm Amphibians?

- Kill algae food source?
- Absorption and direct effects
- Increase desiccation later in life
- Interfere with the endocrine system
- Increase chance of infections
- Lead to increased parasites in the habitat
- Other indirect effects
- It can be hard to quantify the effects of herbicides in the wild, and there are multiple indirect effects. Much of the research is done in controlled settings.

What are Some Common Herbicides?

- **Atrazine** and **Glyphosphate** (Roundup, Touchdown Total of Monsanto Corp., many others) are the most commonly used herbicides in the US and probably worldwide (Long 2005, Rohr 2004)
 - Use of herbicides is on the rise- genetically modified Roundup resistant crops are leading to increased Roundup use and superweeds
- **2,4-D** (the active ingredient in Agent Orange) is also widely used, and DOW Chemical is awaiting approval to sell 2,4-D resistant crops

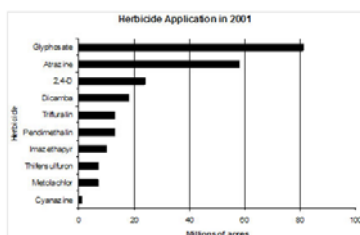


Figure 1. Agricultural use of herbicides in 2001 in millions of acres. Reproduced from www.ers.usda.gov/Briefing/AgChemicals/pestmanagement.htm. (NHTS disclaimer)

Roundup

- Contains protein disrupter Glyphosate and Polyethoxylated tallow amine (POEA), designed to increase efficiency of herbicides by penetrating plant cells
 - 2005 study by Relyea showed high levels of mortality due to the combination of the active ingredient and surfactant

Roundup

- Previous studies suggested that soil can break down the toxic components of Roundup, but this takes too long (half life 7-10 days), and soil type made little difference in Relyea's experiment
- The EPA classified Glyphosate as having a slight to moderate effect on amphibians, and little effect on other vertebrates, thus people considered it safe
- But this testing was only on *Glyphosate*, and few species were used. Turns out that *Roundup* is highly lethal to amphibians! (Relyea 2005)

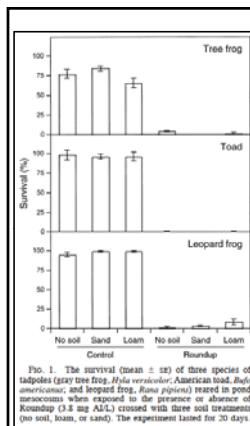


FIG. 1. The survival (mean \pm SE) of three species of tadpoles (gray tree frog, *Hyla versicolor*; American toad, *Bufo americanus*; and leopard frog, *Rana pipiens*) reared in pond mesocosms when exposed to the presence or absence of Roundup (3.8 mg AI/L) crossed with three soil treatments (no soil, loam, or sand). The experiment lasted for 20 days.

Roundup

Tadpoles- exposed to 3.8 mg AI/L for 20 days

- Gray tree frog survival reduced from 75% to 2%
- American toad survival reduced from 97% to 0%
- Leopard frog survival reduced from 98% to 4%
- **Mortality among species over 20 days 98%**

(Relyea 2005)

Roundup

Juvenile frogs- exposed to 1.6 mg AI/L for 24 hours

- Wood frog survival reduced from 96% to 32%
- Gray tree frog survival reduced from 100% to 18%
- Fowlers toad survival reduced from 100% to 14%

– **Mortality across species after one day 79%**

(Relyea 2005)

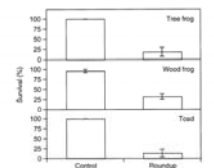


FIG. 2. The survival (mean \pm SE) of three species of juvenile frogs and toads (gray tree frog, *Hyla versicolor*; wood frog, *Rana sylvatica*; and Fowler's toad, *Bufo woodhousei*) after 24 hours of exposure to a direct application of Roundup in laboratory tanks (1.6 mg AI/L).

Roundup

- Roundup probably does not cause mortality by killing algae- deaths occurred quickly, in other studies algae increased because there were too few tadpoles to consume it, others still died when fed fish flakes (Relyea 2005)
- The surfactant that makes it more toxic to weeds also helps it to penetrate animal cells
 - Commercial testing focuses on active ingredients

Roundup

- This experiment shows that the active ingredient and surfactant in Roundup is highly lethal to a variety of frogs at the tadpole and juvenile stage (Relyea 2005)



Roundup

- LC50 for larval salamanders at 96 hours was 2.7 to 3.2 mg AI/L at 96 hours for a variety of species (Reyla 2009)



Roundup Use on the Rise

- “Twenty-one weed species around the world are now resistant to Glyphosate, up from zero in 1996 -- the year Monsanto started marketing its genetically engineered Roundup Ready crops.” (Mercola.com 2011)
 - Genetically modified crops have caused a 7% rise in pesticide use
- Tests with mammalian species show more ways Glyphosphate is a problem... potentially to other vertebrates (Ho 2012)
 - Yet the European Union is considering letting 100-150X more Glyphosphate into the environment

Atrazine

- Systematic herbicide that disrupts photosynthesis
- **Many** problems with amphibians!
- *Lithobates pipiens* exposed to 0, 0.1, 25 ppb
 - Levels often seen above this, even in rainwater
- Developed eggs in testes, low sperm count, etc. Endocrine disruption
- Transect run from Utah to Iowa, same problem was seen
- (Hayes et al., 2002)



Atrazine and Other Stressors

- Streamside salamanders (*Ambystoma barbouri*) exposed at various levels from egg to metamorphosis with limited and unlimited food and in presence or absence of a dry down (Rohr et. Al, 2004)



Atrazine

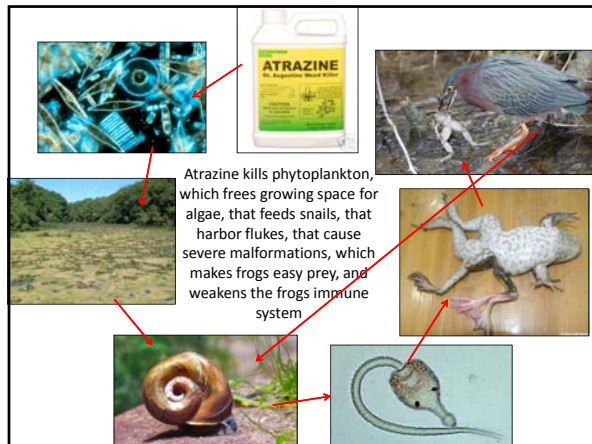
- The higher concentrations of Atrazine decreased hatching success and survival
- Larvae exposed to high concentrations of Atrazine were not affected any more by dry down, those exposed to the lower levels were
 - Why?
- Atrazine decreased metamorph size, especially when coupled with low food. It also decreased the length of the larval stage.
 - Atrazine increased larval activity, but did not affect feeding rate

Atrazine

- So, at least some larvae survived, even at high exposures *and* multiple stressors, even if they are smaller. Good, right?
- Maybe it doesn't matter...

Atrazine

- Atrazine exposure of streamside salamanders in early life was shown to cause an increase in the chance of desiccation months later (Rohr 2004)
- Alters gender
- Atrazine is persistent in the environment, may take months to a year to break down
- Highly mobile, it has been found above the Arctic Circle (Rohr and McKoy 2010)
- There is one animal that benefits from Atrazine- the snail that is a host of trematodes (Rohr 2008)



Are Herbicides All Bad?

- In class you have heard that herbicides are used in conservation management
- Herbicides can be used to control unwanted vegetation
 - Some plants are dangerous to frogs!
 - Invasive plants leech toxic chemicals into the water
- But the positive aspects of herbicides do not outweigh the negative ones, especially with improper use



Why Herbicides are the Most Important Factor!

- They increase trematodes and endocrine disruption
 - Pesticide chemicals in general and the producing factories can contribute to global warming, acid rain, and ozone depletion
 - By making agriculture easier they encourage fragmentation and urbanization
 - Also by expanding agriculture they encourage more fertilizers and insecticides to be used
 - Roads are built to make transportation of herbicides and farm equipment easier
 - Generally weakens amphibians to pathogens and other infectious agents - and there are a lot of these to consider (Chytrid, Rana, Red-leg diseases, alveolates, *Saprolegnia*)
- Many of our anthropogenic changes to the environment are only possible because of agriculture- and herbicides greatly enhanced and expanded agriculture

Summary

- Atrazine and Glyphosate are two commonly used herbicides worldwide, and use is on the rise
- The POEA surfactant included with Roundup is largely responsible for deaths
- Atrazine deforms male frogs, increases chance of infections and number of parasites, increases desiccation, is highly mobile and persistent
- Herbicides are a contributing factor to other causes of decline
- And much is unknown...
 - Caecilians?
 - Bio-magnification from algae and dead amphibians?
 - Effects in lab rats?
 - 2,4-D GMOs?
- By proper herbicide use problems will be reduced, but alternate chemicals are still needed!

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