Habitat fragmentation has been speculated as one of the main anthropogenic causes for the decline in amphibians (1). The destruction of wetlands has been dramatic in Illinois, where almost 90% of native wetlands have been destroyed for agriculture and urban development (2). Agricultural land use can not only increase habitat fragmentation of wetland and forest habitats, but can also increase amphibian exposure to chemicals from agricultural runoff (3). The herbicide atrazine has been reported to produce adverse effects in amphibians. Atrazine is the second most commonly used herbicide in the United States.

**RESEARCH QUESTIONS**

- **Is atrazine present in water, sediment, and tissue samples taken from Warbler Woods Nature Preserve (WWNP)?**
- **Does atrazine exposure at WWNP cause deformities in naive wood frog larvae?**

**Methods**

- **WWNP** (81.5 ha) contains secondary oak-hickory forest, old agricultural fields with tree seedlings, and four ponds (1 ephemeral and 3 permanent ponds, labeled A, B, C, and D, respectively; Fig. 1).
- Pond A (experimental pond) is <35 m to active agricultural fields. Pond C (control pond) is buffered by early successional fields and is >200 m from agricultural fields.
- One 80 μm and one 50 μm Nitex® bolting cloth enclosure were put into ponds A and C. Wood frog larvae (n=125), at Gosner stage, were put into each enclosure (Fig. 2). Tadpoles were allowed to complete metamorphosis and frozen for further tissue analysis.
- Water and sediment samples were taken weekly from March-June, then monthly from July-October in ponds A and C. Samples were frozen until tested for atrazine concentration using a test kit from AMBRAXIS, that uses an ELISA.
- Bullfrogs (*Rana catesbeiana*) were collected from the site to determine if atrazine is bioaccumulating within amphibian tissues.
- Paired t-tests were used to detect differences between atrazine concentrations in pond A and pond C samples.

**Results**

- No limb deformities were observed from the metamorphs collected from pond A.
- All tadpoles in pond C died or were lost.
- Atrazine was detectable in water from April through September (Fig. 3). There was no difference in atrazine concentration between pond A and pond C ($t=4.07$, $P=0.688$, df=19).

**Discussion**

- As little as 0.1 ppb of atrazine can be an endocrine disruptor that can produce feminization in male amphibians, resulting in hermaphrodites, gonadal deformities, and castration (4).
- Although the atrazine concentrations in WWNP water samples were over 0.1 ppb most of the year, they were highest during the time period when wood frogs were metamorphosing (Fig. 4).
  - A previous study showed limb deformities were more common in wood frogs (*R. sylvatica*) that lived in areas that were exposed to agricultural runoff (5).
  - No physical deformities were observed in *R. sylvatica* juveniles.
- Protected areas are still subjected to chemical pollution from agricultural runoff, making it increasingly difficult for managers to maintain amphibian populations.

**Future Directions**

- Sediment, Bullfrog tissues, and wood frog tissues will be analyzed for atrazine.
- Parasite species composition will be assessed from collected bullfrogs.
- Smallmouth salamanders (*Ambystoma texanum*) have accounted for all but one limb deformity seen at WWNP (Fig’s. 5 & 6). Therefore, we will repeat this experiment in 2007, using smallmouth salamander larvae.

**References**


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