

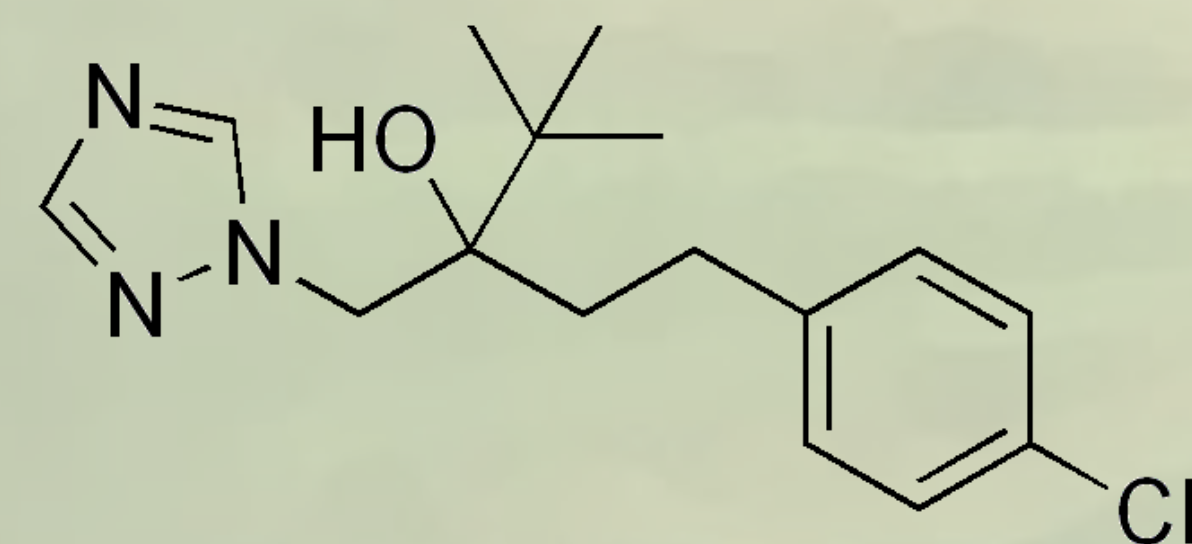
Does the agricultural fungicide Tebuconazole inhibit the growth of the amphibian chytrid fungus?

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Introduction

- *Batrachochytrium dendrobatidis* (Bd), or the amphibian chytrid fungus is a pathogen that affects the development of keratinized skin in amphibians and is responsible for decreasing amphibian populations globally (Daszak et al, 2003).
- Bd is a local concern due to its associations with amphibian population declines in Oregon including Cascades Frog (*Rana cascadae*), Red-legged Frog (*Rana aurora*), and Oregon Spotted Frog (*Rana pretiosa*) (Pearl et al, 2007).
- Common use of pesticides have negatively impacted amphibian communities and they may increase amphibian susceptibility to chytrid infection (Belden et al, 2010).
- Tebuconazole is an agricultural fungicide in the same chemical family as Itraconazole, a drug used to treat fungal infections in animals, including the amphibian chytrid fungus amphibians.
- This experiment aims to test the effects of this pesticide, which is commonly found in amphibian habitats, on the growth of Bd.

We hypothesized that Tebuconazole would inhibit the growth of the amphibian chytrid fungus in culture.



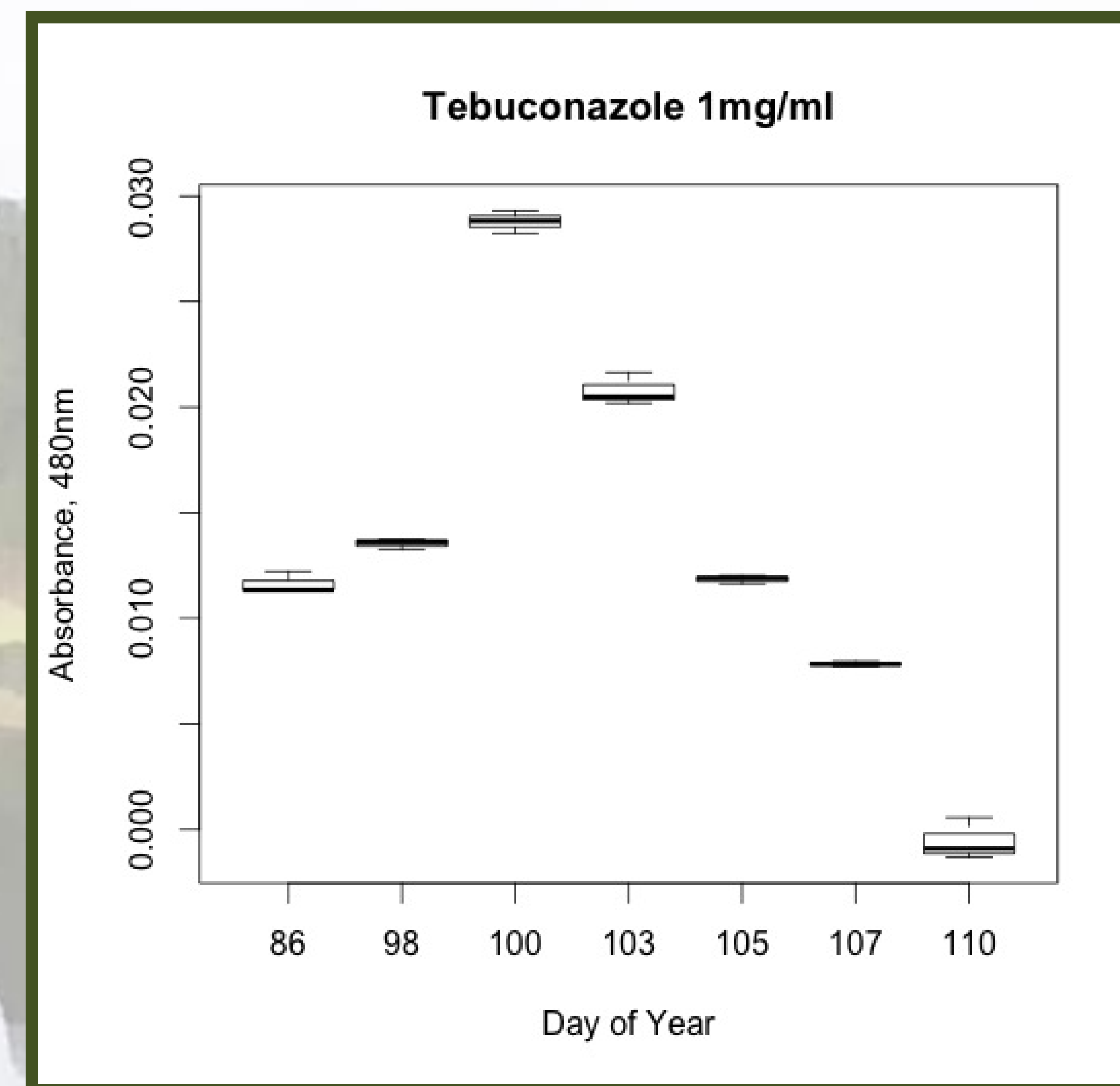
Tebuconazole

Methods

- Bd response to Tebuconazole was measured in a 1% tryptone broth using a spectrophotometer (480nm) to measure the increase in light absorbance as the fungal density increases.
- The experimental design was completely random and there were five replicates of 1mg/ml of Tebuconazole in triethylene glycol added to 5 ml pyrex tubes filled with sterile tryptone broth.
- Absorbance was measured prior to inoculations. On day 98, the tubes were inoculated with approximately 1,000 Bd zoospores (measured with a hemocytometer) harvested from 6 day old plates and the Tebuconazole treatment then incubated at 22°C in a 12 hour light/dark cycle.
- Bd growth was measured three times per week and bacterial contamination was measured at the end of the experiment by placing a sample from the treatment tubes in an agar plate.
- Data were summarized using R 2.15.3

Results

- We observed a rapid increase in fungal density in the first two days of the experiment, followed by a steady decrease in the density.
- The treatment tubes were plated and revealed no bacterial contamination, therefore we conclude the changes we observed were from increases and decreases in Bd density.

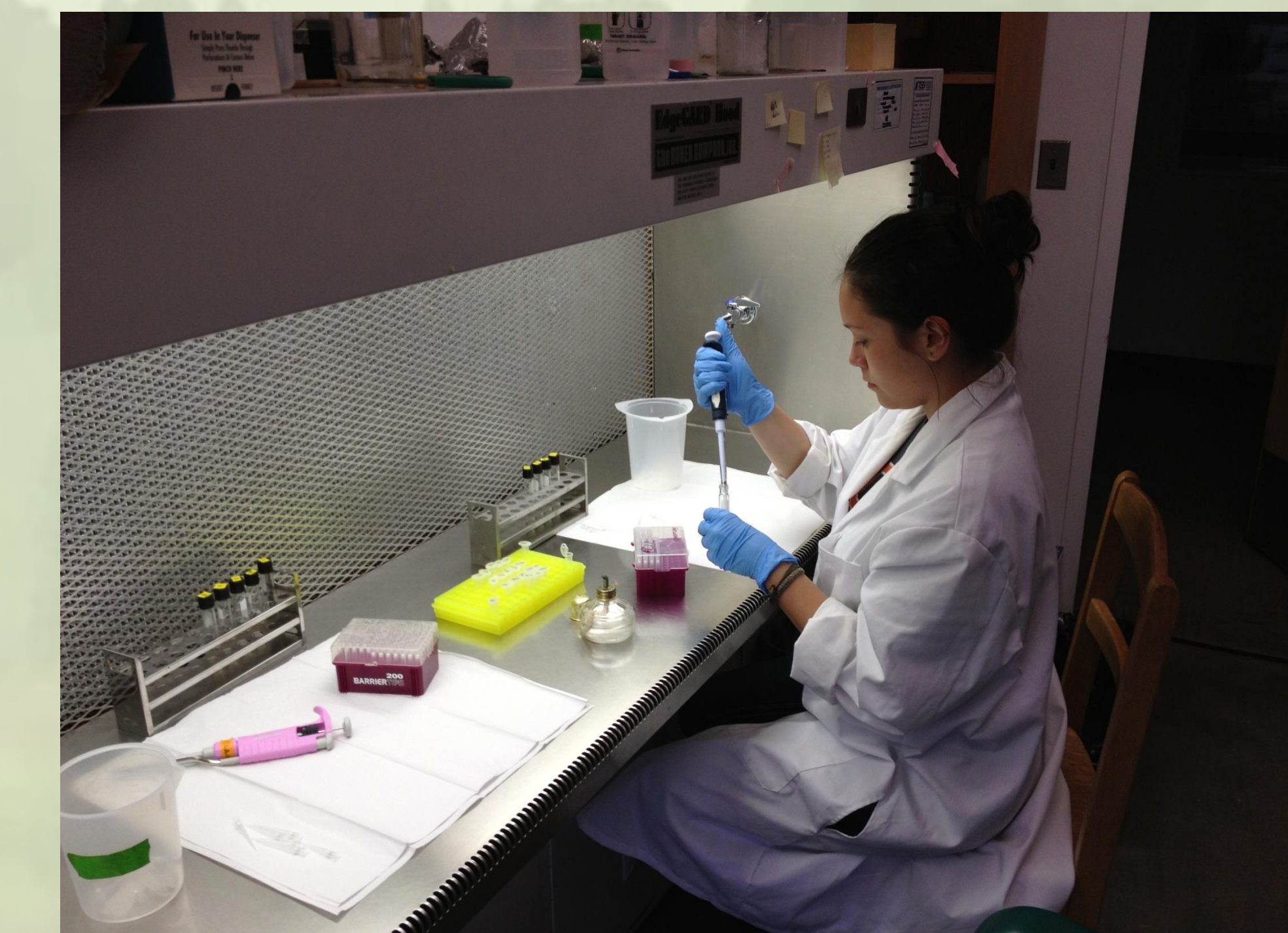


Conclusion

- Amphibian chytrid fungus growth decreased in response to a high concentration of Tebuconazole.
- Exposure time appears to be an important factor in the response.
- Initial increase in growth from day 98 to day 100 may have been a stress response that triggered a rapid increase in production.
- Fungicides present in amphibian habitats may have direct and indirect effects on disease-host dynamics in aquatic systems.

Future Directions

- We will expand this study to include additional current use fungicides detected in amphibian habitats.
- We will assess more environmentally relevant concentrations of the fungicides.
- Fungi are a vital component of the aquatic environment, and the impacts of non-point source pollution from fungicides to the aquatic community and food webs should be further investigated.



Literature Cited

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