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The Effects of Phosphate on Larval Western Barred Tiger Salamanders (*Ambystoma mavortium*)



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Funded by UCARE (Undergraduate Creative Activities and Research Experiences Program)

Research Questions

This study seeks examine the effects of elevated phosphate levels on the larval stage of the Western Barred Tiger Salamander (*Ambystoma mavortium*).

Hypothesis: Additional phosphate in the water will cause the larval salamanders to have a slower metamorphosis rate than the control group with no additional phosphates. This hypothesis is based off of research that showed that additional nitrogen in the water caused a slower growth rate in the Western Barred Tiger Salamander (*Ambystoma mavortium*) (Griffis-Kyle, 2007).

Introduction

Amphibians play an important role as an indicator species. An indicator species is one that is affected by changes in the environment before other species. Amphibians absorb most everything from their environment through their skin. This makes amphibians especially susceptible to changes in the environment, especially chemical ones. They are good indicator of the general health of an ecosystem, and allow scientists to begin to pinpoint potential threats to other species as well as amphibians.

As an amphibian the salamander has an aquatic gill-breathing stage that is followed by a terrestrial lung-breathing stage. The process from which the salamander changes from the larval gill breathing stage to the terrestrial stage is called metamorphosis. When this particular salamander is completely morphed into the terrestrial stage its' gills will no longer be visible.

This salamander is found all across the state of Nebraska, but in the past 10-12 years it has declined in Cass, Douglas, Gage, Jefferson, Johnson, Lancaster, Nemaha, Otoe, Pawnee, Richardson, and Washington counties (Ferraro, 2016). A fertilizer, monoammonium phosphate, has been shown to have an increased usage in this area (Damme, 2018).



Photo on the left is a larval salamander. Photo on the right is a terrestrial salamander.



Methods

Collection

- Bessey National Forest, Thomas County NE

Tank Setup

- 20 liter mark on tanks

Tank Adjustments

- Recorded: food, temperature, observations
- Baseline phosphate levels obtained before adding phosphate
- .05 ppm phosphate EPA limit for a stagnant lake (Mullins, 2009)

Adding Phosphates

- First addition on November 5, 2019
- Levels tested again after adding; were not consistent
- New test kit
- Phosphate added to get all treatment to 10 ppm
- 2 weeks later added phosphate to get all treatment to 20 ppm

Testing Hardness/pH

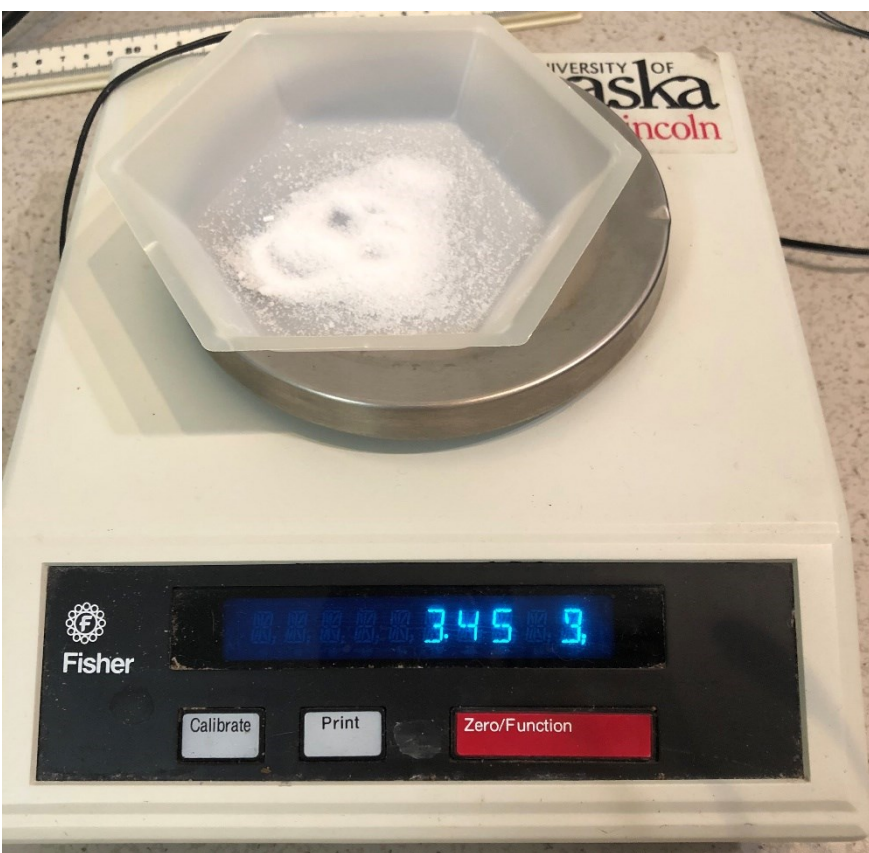
- Charcoal and ammonia added phosphate 2-3 ppm
- December water not added unless below filter line
- Replaced 5 liters and brought phosphate level down
- Dates of full metamorphosis were recorded



Top: Shows the set up of the tanks. Right: The water temperature is being measured. An ice pack was used to help prevent morphing before adding phosphate.



The sodium phosphate was weighed (right) before being mixed with water. The vortex genie (left) was used to mix the water and dissolve the sodium phosphate.



Results

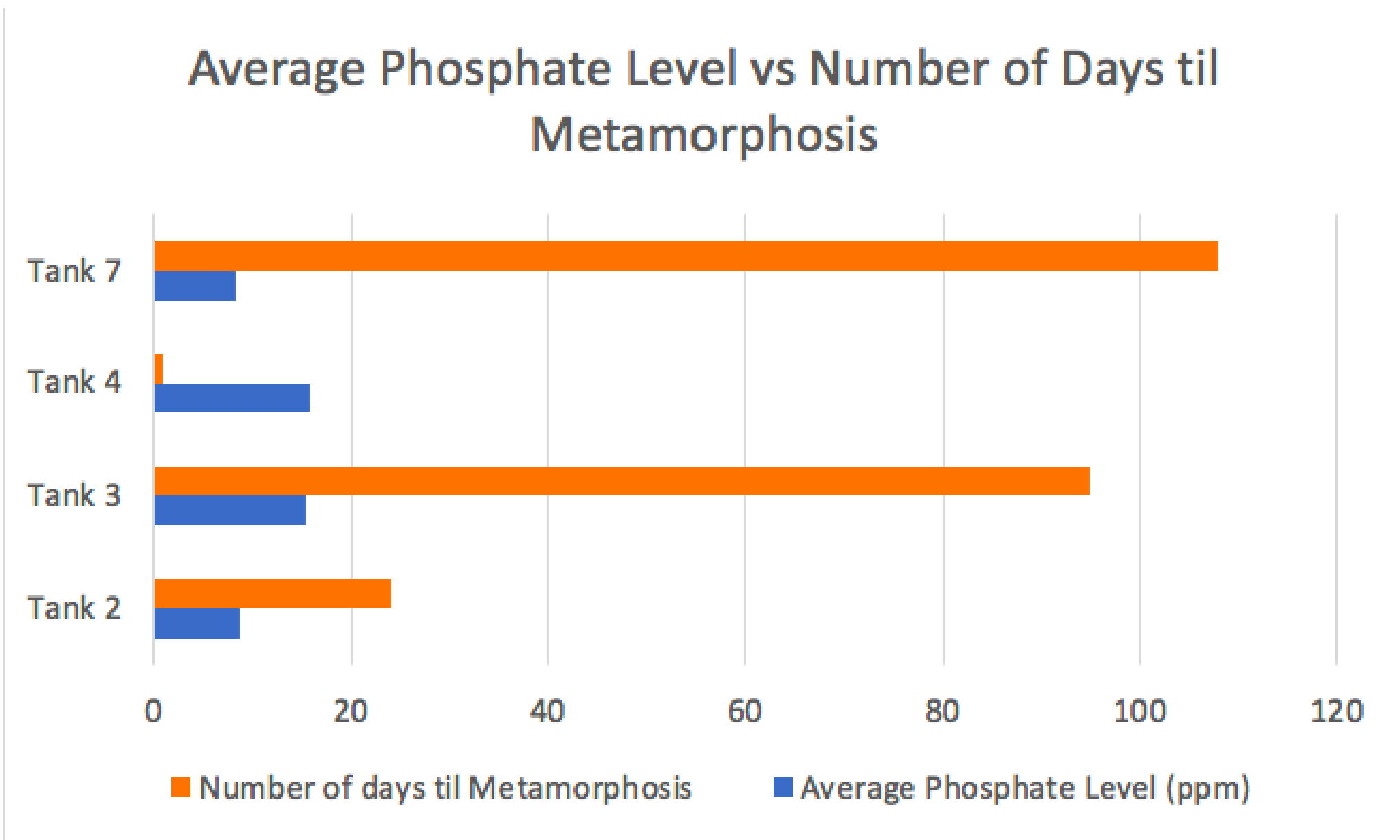
R value for significance is + or -0.7

Correlation Coefficients Related to Morphing		
Tank #	PO ₄ Level	Temperature
1	0	0
2	0.3651484	-0.6288942
3	0.9433701	0.2779605
4	0	-0.612262
5	0	0
6	0	0
7	0.6770032	-0.0136182

Table 4: Correlations comparing the phosphate levels to when the salamanders morphed and the temperature to the morphing data for each individual tank.

Correlation Between Morphed Control and Treatment		
PO4 Control	PO4 Treatment	PO4 Correlation
0.3651484	0.9433701	-1
0.6770032	0	
Temp control	Temp Treatment	Temp Correlation
-0.6288942	0.2779605	-1
0.0136182	-0.612262	

Table 2: This data took the correlation from Table 1 and found the correlation coefficient between the control group and the treatment group for both the phosphate level and temperature.



Graph 1: This graph shows the days till metamorphosis plotted against the average level of phosphate. Tank 4 morphed on the first day of measuring phosphate with the accurate test kit.

Conclusion

The hypothesis was rejected. The dates the larvae morphed alternated between treatment and control. The treatment metamorphosis was not slowed. Temperature was found to not be significantly correlated to morphing. Further studies are needed to determine the effects of phosphate on larval salamanders. There is some data suggesting increased phosphate levels may influence metamorphosis of the larvae. However, contrary to the hypothesis this data suggests that the increase in phosphate may have caused metamorphosis to occur. Despite this only one salamander suggests this data which is not significant enough to say that the phosphate levels influenced metamorphosis.

References

- Griffis-Kyle, K. L., 2007, Sublethal effects of nitrite on eastern tiger salamander (*Ambystoma tigrinum tigrinum*) and wood frog (*Rana sylvatica*) embryos and larvae: implications for field populations. *Aquatic Ecology*, 41(1), 119-127.
Ferraro, Dennis M., 2016, Tiger salamanders disappearing in region. *Faculty Publications in the Biological Sciences*. 395. digitalcommons.unl.edu/bioscifacpub/395
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Mullins, Gregory Lee, 2009, Phosphorus, *Agriculture & the Environment*. Virginia State University