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EFFECTS OF PH, TEMPERATURE, POND SIZE AND LOCATION ON NUMBER OF WOOD FROG (*RANA SYLVATICA*) EGGS IN VERNAL PONDS

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ABSTRACT

We examined whether number of wood frog (*Rana sylvatica*) egg clusters is associated with various environmental factors in four vernal ponds. Number of egg clusters appeared to be positively correlated with pond size, and negatively correlated with distance to the closest edge of a pond. Our small sample sizes limited the rigor of the conclusions that we could make.

INTRODUCTION

The wood frog *Rana sylvantica* is a small, two-inch frog that lives in the moist wetlands of northern North America. In Pennsylvania, it is not an amphibian of ponds and streams, but one that spends its time in woodlands and vegetative wetlands. In the early spring, after thawing from its winter dormancy, the wood frog hops its way to temporary wetlands (vernal ponds) for breeding. After mating and egg lying, the wood frogs leave the vernal pools to spend the rest of the year in woodlands. Clutches of a few hundred eggs are deposited in communal clusters. The gelatin covering, the size of the cluster and exposure to the sun all help the eggs to be warmer than the surrounding water and therefore they develop quickly. By the beginning to middle of April small black wood frog tadpoles are abundant in vernal ponds. By June the tadpoles will have developed legs and be absorbing their tail in preparation for leaving the pool. Wood frogs show characteristics of r-selected species due to the fact that they need to have a high growth rate because the ponds are temporary. Another characteristic that makes them an r-selected species is that that produce many offspring (MacLeod, 1993).

Vernal ponds are temporary bodies of fresh water, which are present for at least two months during the spring. They generally dry up in the summer months. Vernal ponds are typically small and they are fishless because ponds that dry up cannot support fish populations. Due to this fact, there will be less predators for the tadpoles. Most vernal ponds occur in woodland areas, but they can also be found in meadows, sand flats and river flood plains. Some species, such as the wood frog, are totally dependent on vernal pond habitats for one or more stages of their life cycle. They are referred to as obligate species, which means that they are obligated to complete their life cycle in ponds. Other species found in vernal ponds, but not totally dependent on them, are called facultative species. Some examples of these species are the red spotted newt, and the American toad. Vernal ponds may be protected. To receive protection they must meet the following standards: have qualities of confined basin depressions, hold water for at least two months of the year, lack fish populations, provide an important wildlife habitat, and they must be

RESULTS

Table 1 shows the data that we collected on the first day. Each pond is designated by a number. The number of egg clusters as well as the pond dimensions are close approximations. These data were used for our statistical analyses. Scatter plots of number of egg clusters vs pond area, distance to the pond edge, pH and water temeparture are shown in Figs. 1-4. Results of a Chi-square test are shown in Table 2.

Table 1. Number of wood frog egg clusters in relation to various environmental features in four vernal ponds.

	# Egg Clusters	Dimensions (m)	Area (m^2)	pH	Temperature (°C)	Distance from edge (m)
1	200	60 x 45	2700	4.15	10.4	18
2	40	20 x 15	300	4.39	7.4	3.5
3	50	40 x 30	1200	5.88	8.8	0.1
4	0	20 x 13	260	6.25	11	N/A

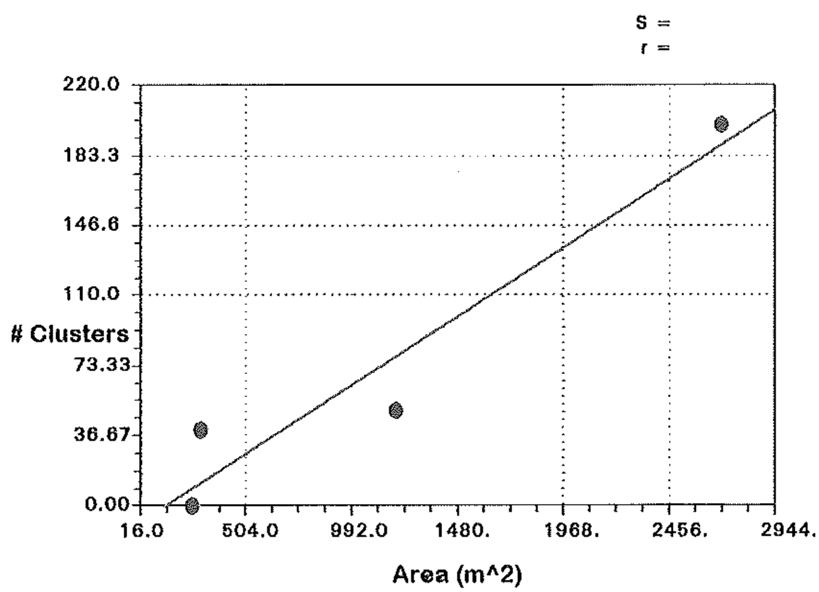

Figure 1. Number of wood frog egg clusters in relation to pond area.

certified by the National Heritage and Endangered Species Program of the division of fisheries and wild life. (Anonymous, 1996).

Our working hypothesis states that wood frog egg clusters should most numerous in large vernal ponds having a neutral pH (~ 6-7) and cool temperatures (~10° C). Our null hypothesis states that the size of the pond, pH, and temperature will have no effect on the number of wood frog egg clusters present.

There are several reasons why it is important to study the occurrence of wood frog eggs in these ponds. In the past three years many amphibian populations, including various salamanders, the red-spotted newt, the spring peeper, leopard frog, wood frog, bullfrog, greenfrog, mink frog, and gray tree frog, have been declining. The major reasons suggested for this decline are habitat destruction, chemical pollution, increased UV-B radiation, parasites, and predation. This decline may have significant ecosystem implications. A quote from ABC's Nightline on June 20, 1997 states that " If frogs begin showing signs of distress, it could be only a matter of time before other species are affected, including humans." Because of the unique life-history characteristics of the wood frog, and their close association with soil, water and yearly breeding activity, they are well suited as an indicator taxon of changes in the environmental conditions in forest environments. This research will help to provide information on the optimal living conditions of the wood frog in order to preserve their habitat.

FIELD SITE

We began this research in mid to late March, and started by locating vernal ponds alongside Petersburg Pike near Huntingdon, PA. Unfortunately we found only four in the area, which is not at all what we expected. Three of these ponds were located approximately 20-30 feet apart, and the fourth was about a half a mile down the road. The first pond, which was also the largest, was about 20 meters from the road. The other two in that area were situated behind the first one. The fourth pond was about 5 meters back into the woods and was visible from the road. All of these ponds were surrounded by thick vegetation.

METHODS AND MATERIALS

We took data on two dates, one in March, the other in April, 1998. Our measuring and testing involved one or two people in waders who entered the water, if necessary, to locate and count the egg clusters. We measured both the length and width (m) of the pond with a tape measure. We used a meter stick to measure the depth (cm) around the egg clusters at several different places depending on the amount of eggs present. We recorded the water temperature at the egg cluster site with a thermometer. Using this temperature we proceeded to test the pH at the egg cluster location. This was done by calibrating the pH meter, followed by testing the pH of the water by leaving the probe in for approximately five minutes. The location of the egg clusters in relation to the edge of the vernal pond was also measured with a tape measure or yardstick, depending how far into the pond they were.

The identification of only four ponds limited our results. The statistical procedures were restricted due to this small sample. Though we collected data on two different days, we only used the data from the first day in order to avoid the effect of additional factors. Between the two data collections, not only was it very warm and sunny out, but also it rained a good deal that morning. From the first set of data, we graphed number of clusters vs pH, water temperature, pond area and distance from the pond using a graphing program called "CurveExpert 1.2." We graphed the points for four different sets of data and found a linear best-fit line through the points (Figures 1-4). We then found the Standard Error, "S", and the correlation coefficient, "r" for each graph. From these graphs, possible correlations between these factors were explored. We also conducted a Chi- square test on the approximate number of clusters in each pond to determine the significance of our null hypothesis.

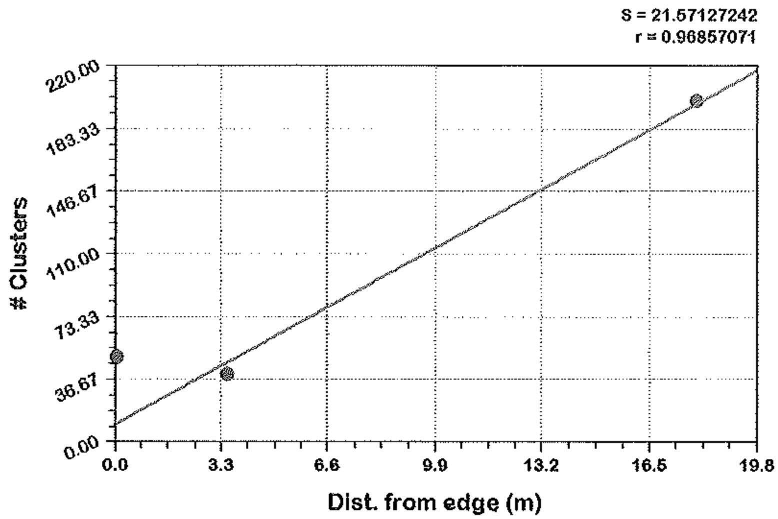

Figure 2. Number of wood frog egg clusters in relation to distance to closest edge of the pond (m). Only three ponds are shown because one pond did not contain eggs.

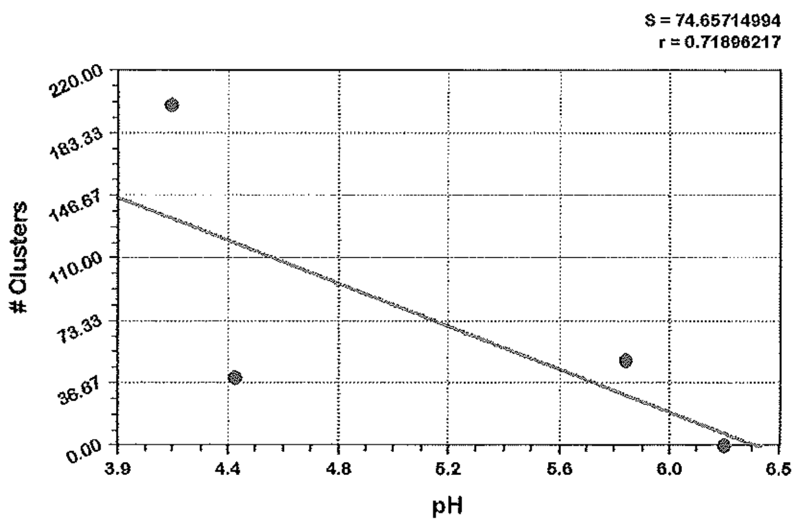

Figure 3. Number of wood frog egg clusters in relation to pH in four ponds.

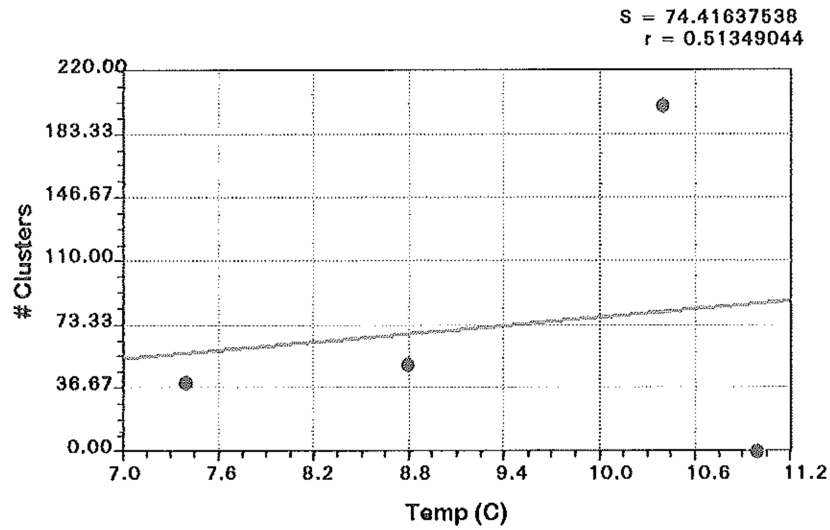

Figure 4. Number of wood frog egg clusters in relation to water temperature in four ponds.

Table 2. Chi square data for number of egg clusters.

Data for chi square test:	
DF=3	χ^2 = 318.2759
critical value: 7.815	

DISCUSSION

Several questions that we have raised are: Does pH, temperature, and location affect the abundance and presence of wood frog eggs? Does pollution in or around the ponds have any effect on the eggs? Does the presence of the gelatin coverings around the eggs affect their tolerance to extreme temperatures?

Horne and Dunson (1995a) found that low pH was, in fact, toxic to wood frog embryos after prolonged exposure with high aluminum concentrations. Metals such as copper, zinc, aluminum, and lead, along with low pH, were found negatively impact amphibian communities. In some instances, low pH was found to even prevent hatching of the eggs (Horne and Dunson, 1995a). In another study, wood frog eggs were found to take longer to develop in tanks of high dissolved organic carbon and low pH (Horne and Dunson, 1995a).

In most cases studied, low pH along with metal concentrations were found to be detrimental to the development and survival rates of amphibian eggs. Low pH commonly causes a fatal curling defect in developing amphibians (Horne and Dunson, 1995b).

Our results, though limited, gave us an idea of the possible relationships between pH, temperature and pond size/structure and the number of wood frog eggs present. Our correlation analysis showed a possible relationship between the number of egg clusters and the area of the pond, as well as the distance of the eggs from the edge (Figs. 1 and 2). The correlation coefficients were high for both regressions (0.969 and 0.960, respectively). However, number of egg clusters showed no obvious relation to pH or temperature (Figs 3 and 4). This could be due our small sample size (since we only took data from four ponds). We only had an estimated count of the egg clusters due to the unexpected hatching of the eggs. Though the eggs were still visible, they were difficult to count. This inaccurate count could also have affected any correlation that may actually exist between these factors.

The chi-square test that we conducted on the number of egg clusters in the four ponds showed that the number of wood frog eggs in the ponds must be due to some factor, thus making our null hypothesis improbable.

When we first began this project, we did not realize that there were so few ponds in the area. Had we found more ponds, our data may have been more accurate and we may have been able to conduct more statistical tests. The tests that we conducted may also have been affected by the small amount of data that we had available. For further experimentation we could have collected more data from each pond, such as pH and temperature readings from different areas of each pond. This may have increased the accuracy of our data and thus the accuracy of our statistical tests.

We also could have increased the time over which we conducted the experiment. Had we collected data more often, we may have been able to observe any changes and how those changes affected the eggs. This may have given us more insight into possible reasons for the differences in the egg abundance. The abundance may also be due to the combination of several factors, which we did not take into consideration. Further tests could be conducted if there was more data available.

Other environmental factors potentially affecting numbers of wood frog egg clusters in vernal ponds should be investigated. For example, metal content in the water or the soil around the ponds may be important as observed by Horne and Dunson (1995a, b).

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