Characterization of Vernal Pond Amphibian Populations in the Ottawa National Forest Prior to Clear Cutting and Selective Thinning

Christopher German and Jennifer Slavick

Abstract

Four vernal ponds in the Ottawa National Forest serve as the experimental regiments while three ponds on University of Notre Dame Environmental Research Center (UNDERC) property serve as a control regiment. Each pond was sampled during three separate sampling periods throughout the summer: May 30 – June 2, 2002, June 25 – 26, 2002, and July 15 – 16, 2002. Significant correlation and ties between air temperature and larval amphibian density, maximum vernal pond depth and larval amphibian density, conductivity and larval amphibian density, and larval amphibian density and species richness have been illustrated through this year’s data. Consequently, as air temperature increases density decreases, as maximum pond depth increases density increases, as conductivity increases density decreases, and as density increases species richness increases. Such correlating and significant environmental factors that did not appear to be relevant in last year’s study may now provide a series of benchmarks to which change can be compared.

Introduction

Amphibian populations play a vital role in both aquatic and terrestrial environments due to their diversity and abundance, however, they are declining at alarming rates worldwide (7). Concern is building as to why amphibians are dying off and what this may mean to human populations (6, 9, 12). Since they reside in two separate environments, have highly permeable unprotected skin, and eggs that lack hard outer shells and an amniotic sack, amphibians have become valuable indicators of early stage environmental change (1, 2). With such physical attributes, amphibians are highly sensitive to minor changes in climate, ultraviolet radiation (UV-B), predation, pollutants and toxins, diseases, and environmental modifications (1, 2, 4, 9, 10, 12).

The Ottawa National Forest, located in northern Wisconsin and Michigan’s Upper Peninsula, is planning on clear cutting and selectively thinning sections of forest. In accordance with the National Forest Management Act of 1976, however, national forests are required to maintain viable numbers of all vertebrate species within their land (11). The effects of clear cutting upon vertebrate populations in the Ottawa National Forest
Increased predation and higher susceptibility to disease. Increased frequency, and reduced immunosuppression, all of which lead to premature death.

Radiation also leads to physiological depletions such as eye damage, increased cancerous physical development (1, 2, 4). It should be noted that increased exposure to UV-B radiation increases in mortality, and altering both rate and morphology of reproduction increases in mortality, lowering the rate of directly correlates with amphibian population decline through lowering the rate of increased sunlight exposure also leads to an increase in UV-B radiation, which.

Number of species A will in mal the area in regard to species B. Reproduction prior to the destruction of pond but species B does not a disproportionate number of species A will in mal the area in regard to species B. In addition, if a species A successfully reproduces prior to the destruction of pond, but species B does not, a disproportionate increase earlier than the time of larval metamorphosis, the young amphibian offspring will have for amphibian populations that annually use a pond as a breeding ground. If the pond ponds on the property due to increased sunlight exposure would present large problems a dramatic effect upon the environment. Premature seasonal evaporation of many vertebral Removal of vegetation and canopy cover surrounding the vertebral ponds may have

 liefing populations and environments can be compared. 2002 in conjunction with data collected in 2001 will create a baseline against which post-
Effects upon aquatic environments aside, clear cutting and thinning will create pronounced changes in the vegetation surrounding the vernal ponds. While nearly all amphibians are born and develop in aquatic environments and return to breed, a significant portion migrate to the surrounding terrestrial environments during their non-breeding season (9, 11). Less surrounding hardwood and shrub vegetation may lead to a decrease in habitat diversity, cover, and organic matter and an increase in predation. In addition a decrease in vertebrate populations, which may have symbiotic relationships with the amphibian populations, would also be expected.

It is unknown as of yet what the actual results of clear cutting and thinning around vernal lakes in the Ottawa National Forest will be, the above hypotheses are based upon past studies. While this study will eventually cover the environmental changes following vegetation removal, for now the emphasis lies on collecting data regarding amphibian population diversity, population numbers, and natural rates of population growth and decline.

Materials & Methods

Vernal Pond Characterization

Four ponds in the Ottawa National Forest and three ponds at University of Notre Dame Environmental Research Center (UNDERC) were used for amphibian population sampling and characterized through multiple tests and measurements once at the beginning and end of the research period. Initially the circumference, length, width, and maximum depth of each pond was measured to create a numbered grid sectioning the perimeter of each pond into one meter square plots zero and one meter from the shoreline. Wooden stakes were placed 10 meters around the perimeter of each pond for

Throughout the summer, perimeter grids were updated during every sampling. Sampling was conducted through a coin flip. Due to variable pond elevation, "A" and "B" were determined through a coin flip. Plot number sampled was determined using a random number table and assigned "A" or "B". The plots were assigned "A" and those plots one to two meters from the shore were meter plots. Each pond was given a number and those plots zero to one meter from the shore were counted. At least twenty percent of the shoreline area of each pond was sampled. Each larval amphibian sample around each pond was taken.

Measurements for each of these lists were taken at five randomly selected points around each pond. These included conductivity meter, and pH with a Hanna Instruments pH meter. The range was recorded. Dissolved oxygen meter, conductivity with a Hanna Instruments HI 9033 multi- meter were measured with a yellow 55 dissolved oxygen meter. Conductivity with a Hanna Instruments HI 9033 multi- meter were taken using a standard alcohol thermometer. Dissolved oxygen (DO) was measured with a Garmin e2 global positioning system (GPS). Amphibian diet and water temperatures were taken. Locations of each coastal pond were taken in UTM coordinates through the use of Global Positioning System (GPS) and by use of GPS coordinates. The

commercial vegetation, and species of macrophyte and surrounding vegetation were also observed. Observations regarding weather, distance from shoreline to forest edge, percent margin of color, water turbidity, primary substrates, and the presence of fish. Additional reference points during sampling, qualitative observations at each pond were water
Once sample plots were determined, a 31-gallon modified Rubbermaid bin with the bottom removed, measuring 0.876 m long, 0.508 m wide and 0.425 m deep, was placed into the plot and held securely to the bottom of the pond, preventing any amphibian larvae from escaping. The bin was swept thoroughly with a small fish net and the contents placed into a bucket for analysis. This process was completed three times per plot and all larval amphibians captured were taken back to the lab in labeled Ziploc containers filled with pond water. If adult amphibians were captured, species and quantity were noted and promptly returned to the pond following sampling.

**Larval Amphibian Analysis**

All captured larval amphibians were returned to the lab and placed into labeled tanks for quantitative analysis. Upon identification using the USGS Patuxent Wildlife Research Center dichotomous key (http://www.pwrc.usgs.gov/tadpole/), each larval amphibian was measured using General calipers. For Anurans, tail length, torso length, and total length was recorded. For Caudata, total length was recorded. Following identification and measurement larval amphibians were returned to their vernal pond of origin.

**Drift Fence Pilot Experimentation**

UNDERC numbered vernal pond two was used for a drift fence experimentation trial lasting forty-eight hours. 175 meters of drift fencing were used with bucket traps placed flush with the fence every 20 meters. Traps were checked every 12 hours and all catches recorded.
Bar charts were created using Microsoft Excel, regression scatter plots graphs were created using SYSTAT, and Pearson correlations were conducted using SPSS. To determine larval amphibian density, the total number of individuals captured at each vernal pond was divided by the number of plots sampled. Standard deviation was calculated for each density determined. Pond size (m²) was determined using the radius formula, \( A = \pi a^2 b \), where \( a \) and \( b \) are length and width radii.

Results

Species richness in all of the vernal ponds, both control (UNDERC) and experimental (Ottawa National Forest), varied greatly among sampling times and ponds. The greatest species richness was found in ND-N2 during the 6/25/02 - 6/26/02 sampling period, which was also the most species rich pond over the course of all sampling periods. Except for V.P. 2, low species richness relative to ND-N2 was found in the other ponds. Richness tended to be highest during the second...
sampling period and lowest during the first sampling period, indicating that mid-June is the most common spawning time. Compared to data collected summer 2001, overall species richness increased by two species in ND-N2, one species in ND-N3, two species in V.P. 2, one species in V.P. 3, and decreased by one species in ND-N1. There were three collection periods summer 2002 and two collection periods summer 2001.

Early in the summer it appears that *Rana sylvatica* is the most abundant species with a large number of individuals unidentified anurans and caudata.

ND-N2 has the largest amphibian community while ND-N3 and V.P. 2 each follow with roughly one-half that population size. During the second sampling period the number of *Rana sylvatica* captured increase from 5 to 25, with 8 individuals in ND-N2 and 17 individuals in V.P. 2.

*Hyla versicolor* is the second most abundant
number of individuals sampled, the total diversity over last years in individuals captured, cumulative total of 42 and V'P. 2 having a cumulative total of 69 with ND-2 having a throughout the summer, amphibian populations had the most abundant ND-2 and V'P. capured throughout the summer were Rana spholidia.

N3, and V'P. 2 and ND-2, V'P. 2, V'P. 5 respectively. In addition, 8 of 10 adults. Both Rana spholidia and Pseudacris triseriata have populations in 3 ponds; ND-2, ND-2, abundant species but instead replaced by Pseudacris triseriata (4 individuals) (fig. 4).

By the Final sampling Period Rana spholidia (4 individuals) is no longer the most V'P. 2 and V'P. 5. Pseudacris triseriata is the most widespread species with populations in ND-1, ND-2, species during the second sampling period with numerous individuals in ND-2.
captured decreased dramatically, even with 3 sampling periods as opposed to 2.

Approximate values for individuals caught over 3 sampling periods this summer are 1 for ND-N1, 69 for ND-N2, 8 for ND-N3, 42 for V.P. 2, 16 for V.P. 3, 5 for V.P. 5, and 2 for V.P. 6. Approximate values for individuals caught over two sampling periods last summer are 30 for ND-N1, 105 for ND-N2, 132 for ND-N3, 170 for V.P. 2, 0 for V.P. 3, 53 for V.P. 5, and 39 for V.P. 6. Larval amphibian density for the first sampling period was the highest in ND-N2 with 1.0 individuals/m² (Fig. 5). V.P. 2 and ND-N3 have roughly half this density.

Density of nearly all the pools with the exception of V.P. 6 and ND-N3 increased during the second sampling period (Fig. 6). ND-N2 once again had the highest density with 2.8 individuals/m² and was followed by V.P. 2 and V.P. 3 with 1.6 and 1.1 individuals/m² respectively. A drop in density at all vernal pools with the exception of ND-N3 characterized the third sampling period (Fig. 7). V.P. 2 had the highest density with 1.4 individuals/m² with ND-N2 following at 0.8 individuals/m² and ND-N3 and V.P. 5 following with 0.06 and 0.1 respectively.

Density of larval amphibians this decreased by a very large margin compared to sampling data from last summer, in which ND-N3 had an approximate density of 14 individuals/m² and V.P. 2 had an approximate
(Table 15). Correlation between species richness and pond size strongly indicates that as $d = 0.072$, and larval amphibian density and maximum depth ($p$-value = 0.712) the most relevant correlations were species richness and pond size ($p$-value = 0.712).

Table 1. During the first sampling period the period showed some relevant relationships (Table 1). During the first sampling period the amphibians and larval amphibian populations during these sampling periods and larval amphibian populations were collected only in ND-NZ during the first sampling period.

The least abundant, dense, and widespread species, Podarcis muralis, was the most abundant, two Canadian species collected this year, Amphiura lacera, was the most abundant species. These species are collected this year, Amphiura lacera. Of the morphologically similar species, were collected this year, Amphiura lacera. A result is likely that all outside their range (5), however a large number of Amphiura lacera is No Amphiura jeffersoni were collected this summer and the INDEC area is Amphiura jeffersoni the second most abundant and dense species. Amphiura jeffersoni was the most abundant and dense species and summer, instead Amphiura strigata was the most abundant and dense species and A.p. 5' during the second sampling period. No Podarcis muralia were collected last year and most widespread of all species, covering four Vernon ponds, ND-1, ND-NZ, A.p. 2, and Podarcis muralia was the most likely due to melanophores. In addition, Podarcis muralia was the highest density of all species for the first two sampling periods and replaced by Podarcis muralia during the final sample periods. Podarcis muralia was the most abundant and had the highest density of all species. Caetes was approximately 1.5 individuals/m² in ND-1 during the first sampling period. Sampled last summer, the lowest density of a pool in which larval amphibians were density of 15 individuals/m² for the first of the two sampling periods. Of all ponds
pond size increases so does species richness. While the correlation between pond size and richness is not significant, more replication might drop the significance to below 0.05. Strong correlation between larval amphibian density and maximum depth indicates that as maximum depth increases so does amphibian density. Further significant correlations between water temperature and air temperature \( (F\text{-value} = 0.841, p = 0.018) \), dissolved oxygen and air temperature \( (F\text{-value} = 0.862, p = 0.012) \), pH and air temperature \( (F\text{-value} = 0.743, p = 0.056) \), and pH and pond size \( (F\text{-value} = 0.767, p = 0.044) \) are also found amongst first sampling period data.

During the third sampling period correlations occur between conductivity and species richness \( (F\text{-value} = -0.861, p = 0.061) \), air temperature and larval amphibian density \( (F\text{-value} = -0.845, p = 0.072) \), and species richness and larval amphibian density \( (F\text{-value} = 0.903, p = 0.036) \) (Table 1b). Thus as conductivity increases species richness decreases, as air temperature increases larval amphibian density decreases, and as larval amphibian density increases species richness increases. In addition, as maximum depth increases pH increases \( (F\text{-value} = 0.989, p = 0.011) \) and as water temperature increases pH increases \( (F\text{-value} = 0.927, p = 0.024) \). While correlations between species richness and dissolved oxygen \( (F\text{-value} = 0.777, p = 0.122) \) and between water temperature and maximum depth \( (F\text{-value} = 0.888, p = 0.122) \) appear during the third sampling period, both correlations are not significant.

Corresponding to significance in correlation are regression plots of various environmental data compared to larval amphibian density. During the first collection period strong regression between maximum depth and density \( (r^2 = 0.5075) \) reinforces the statement that as maximum depth increases larval amphibian density increases (Fig. 11f).
While correlation between pond size and species richness was very close to significant (Table 1a), regression between the two ($r^2 = 0.0803$) indicates otherwise (Fig. 8). As for
the other regressions of environmental factors with larval amphibian density, none of the
$r^2$ values are large (Fig. 10). During the third sampling period all three significant
correlations involving species richness and larval amphibian density had strong
regression plots. Species richness compared to larval amphibian density show not only
the strongest correlation effecting density but the strongest regression as well ($r^2 =
0.8148$), backing the claim that as larval amphibian density increases species richness
increases.

Both air temperature compared to larval amphibian diversity ($r^2 = 0.7141$) and
conductivity compared to species richness ($r^2 = 0.7417$) (Fig. 9) also show strong
regressions that back negative correlation, meaning that as
air temperature increases larval amphibian density
decreases, and as conductivity increases species richness
decreases. Conductivity
compared to density ($r^2
= 0.4531$) and pH
compared to density ($r^2 = 0.3784$) have notable
regression, indicating possible connection. Strength
of correlation between pH and density is fairly weak
($F$-value = -0.053) (Table 1b); therefore a trend is

![Figure 8 - Pond Size (m²) vs. Species Richness.](image)

![Figure 9 - Conductivity (uS/cm) vs. Species Richness.](image)
Figure 11 - Total larval amphibian density (individuals/m²) per plot at each vernal pond during the third sampling period (July 15 - July 16, 2002) as compared to (a) dissolved oxygen, (b) water temperature, (c) air temperature, (d) conductivity, (e) pH, (f) maximum depth, (g) species richness, and (h) pond size.
Larval amphibians collected (Fig. 10c).

Collection period, indicating that as dissolved oxygen increases so does the number of larval amphibians collected ($r = 0.4973$, $F = 0.033$, $P = 0.13$). A noteworthy regression between dissolved oxygen and larval amphibians is compared to not significant ($r = 0.4507$, and as species richness increases the total number of larval amphibians increases ($r = 0.6348$, $P = 0.01$). As conductivity increases the total number of larval amphibians decreases ($r = 0.4483$, $P = 0.05$). These results are strengthened during the third sampling period.

Larval amphibians are strongly correlated with each environmental factor. Correlations seen in correlation and regressions of each environmental factor (Fig. 11).

Verbal pond (Fig. 12).

That is maximum depth increases as does the total number of amphibians found at a site (Fig. 10b). Supports earlier relationship between maximum depth and density, indicating strong relationship between maximum depth and larval amphibian density (Fig. 10, Fig. 11). During the first sampling period significant regression shows relationships between larval amphibian density and number of larval amphibians (Fig. 12).

Regression plots of environmental factors and number of larval amphibians (Fig. 13).

As for regressions of other environmental factors with larval amphibian density, none were substantial (Fig. 11). While this year's data shows regressions and significance, next year's data shows no such links.

As for regressions of environmental factors with larval amphibian density, none were substantial (Fig. 11). While this year's data shows regressions and significance, next year's data shows no such links.
Figure 12 – Total Number of larval amphibians captured at each vernal pond during the first collection period (May 30 – June 2, 2002) as compared to (a) water temperature, (b) air temperature, (c) dissolved oxygen, (d) conductivity, (e) pH, (f) maximum depth, (g) species richness, and (h) pond size.
Last year's data shows no ties between regression plots of environmental factors to number of larval amphibians collected.

Average tadpole size, average tadpole body size, average tadpole tail size, and the range of tadpole size increased with each sampling period, showing growth in each species through the summer (Table 2). Compared to last year's data, *Rana sylvatica* tend to be smaller in the early/mid summer (first and second sampling periods respectively) (16.90 – 26.20 mm and 29.90 – 50.90 mm vs. 25 – 62 mm) though larger by the end of the summer (third sampling period) (53.00 – 61.70 mm vs. 43 – 60 mm). *Pseudacris crucifer* also tends to be smaller as compared to last year's data (17.80 mm vs. 19 – 31 mm). While no *Hyla versicolor* were found in the mid-summer last year, they were most

<table>
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<th>Collection Date</th>
<th>Average Tadpole Size</th>
<th>Average Body Size</th>
<th>Average Tail Size</th>
<th>Range of Tadpole Size</th>
<th>Number Collected</th>
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<td>19.55</td>
<td>37.88</td>
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Densities fluctuate most likely due to metamorphosis of species at different
juvenile stages fully developed.

Development niche that will allow for the greatest resources availability when the
instead of late summer metamorphosis it is a new species to the area, trying to find a
later summer. Such small population numbers of Pseudacris irisinga may indicate that
into the third sampling period, Ambystoma maculatum appears to be developing into the
metamorphosis is most likely responsible for the decrease of most populations some
summer with exceptions being Ambystoma maculatum and Pseudacris irisinga. While
Amphibian populations were abundant during the second collection period mid-
summer, with exceptions being Ambystoma maculatum and Pseudacris irisinga. We
when we cannot know until more data is collected in the coming years.

reduced a result of increasing biological diversity in the Okawa National Forest due to fluctuation
above age population size and density as a whole, through richness increased. Whether this is a
Amphibian populations from summer 2001 to summer 2002 but decreased in both
Discussion

whether over the forty-two hour sampling period.

The drift fence plot experiment obtained only one result in hab 5 along fence 2, a
summer through provided size data this summer (Table 2).

Ambystoma maculatum, Pseudacris irisinga, and Rana clamitans were not captured last
found in summer 2001 (28.90 - 29.30 mm vs. 15 - 37 mm) Ambystoma lateral.

Hyla versicolor found in late summer 2002 did tend to be smaller than those
abundant during the mid-summer this year with a range of 11.40 - 27.00 mm.
ponds at certain times. Larger species such as *Rana clamitans* take a longer period of time between larval and adult stages, indicating more permanent standing water in a pond as would be necessary for development. Mid-June, however, is likely the major spawning time due to high vernal pond productivity during this period of the summer.

An expected effect of decreased canopy cover with clear-cutting and thinning is increased sunlight exposure to the surface of the vernal ponds. Increased sunlight exposure not only allows more UV-B radiation to penetrate the pond, which is already known to be detrimental to amphibian populations (1,2,5), but increases ambient air temperature, which decreases larval amphibian densities according to our data. In addition, as ambient air temperature increases more insect larvae metamorphose from the vernal ponds in which they are developing, leading to a decrease in available food supply available to larval amphibians in those vernal ponds.

Maximum depth of vernal ponds proves to be a large factor in larval amphibian density; as depth increases so does amphibian density. Furthermore, as density increases amphibian richness increases. The maximum depth of a pond is in large part determined by rate of evaporation, therefore, if a pond is exposed to higher light levels and ambient air temperature the rate of desiccation will increase, resulting in lower larval amphibian density and richness.

Five of the seven ponds sampled had less than fifty-percent emergent vegetation cover throughout the entire summer. A decrease in emergent vegetation may be a reason for a decrease in amphibian populations over the past year, however no detailed information regarding changes in vegetation abundance is available.
it much more difficult for Ensared amphibians to escape.

An increase in trap size and the placement of a funnel on the top of the traps would make
that the fences worked beautifully, though the traps were far too shallow to be effective.
whole at these vermal ponds. The drift fence plot experiment was successful in the fact
which or added to allow a better glimpse of larval amphibian ecology amongst the larger

To make the study more robust in the coming years, a number of things can be

Toxins, diseases, or environmental modifications (1, 2, 4, 9, 10, 12).

be in good health, lacking any indications of over exposure to UV-B, pollutions and

The larval and adult amphibians captured throughout the entire study appeared to

The characteristics of a breeching event (9, 11).

sink, providing habitat for amphibians migrating from nearby vermal ponds but lacking

Accordingly, it is likely that V. p. 6 is not an amphibian source but instead an amphibian

during the second sampling period. V. p. 6 provided no larval amphibian whatsoever
they did not dry up until mid-July. While V. p. 2 provided a number of larval amphibians

V. p. 3 and V. p. 6 were the only two ponds to thoroughly desiccate and even then

necessary.

Further investigation into effects of vegetation upon larval amphibian populations is

overcrowding could become a problem and mass population deaths could result.

decreased and emergent vegetation possibly increase food sources available to tadpoles.

increase larval amphibian populations. On the downside, as predation pressure is

beneficial for larval amphibians to hide amongst, thus reducing predation pressure and

An increase in emergent vegetation cover could possibly create more diverse
To give more correlations and trends as the summer progresses, environmental data should be taken during every sampling period, thus seasonal environmental variance can be taken into account at a finer level. In addition, light levels and evaporation rate should be measured at each pond, giving detail as to the amount of UV-B exposure and desiccation rate. A more detailed vegetation survey would be largely useful to evaluate as well, since it is a large source of both food and habitat and may very well be largely responsible for variances in larval amphibian density and richness from pond to pond. In terms of data analysis, having detailed raw data available from previous years, especially in electronic format, would be invaluable for tracking change over time.

Important ties between larval amphibian density and richness were found through this year’s study, and as time progresses larger seasonal variations will become more apparent, which is important in tracking environmental change with the introduction of clear cutting and selectively thinning.

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Appendix 6

Sources


Appendix


