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PURPOSE

The purpose of the JOURNAL OF ECOLOGICAL RESEARCH is to print original ecological research reports written by undergraduate students enrolled in ecology and environmental science courses at Juniata College. Some of the research included here has been carried out at the Raystown Field Station. Through participation in this journal, it is hoped that students will begin to gain an appreciation of the publication process in science. In addition, the journal serves as an archives of past student research projects that current students can potentially use as background for their own projects. Topics on all aspects of the science of ecology are welcome.



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Cover illustration: Mountain Laurel

(drawing from USDA-NRCS PLANTS Database / Britton, N.L., and A. Brown. 1913. *Illustrated Flora of the Northern States and Canada*. Vol. 2: 684)

***NOTOPHTHALMUS VIRIDESCENS* AS A POTENTIAL PREDATOR OF *AMBYSTOMA MACULATUM* IMPLICATIONS FOR COEXISTENCE IN VERNAL PONDS**

Mary Coursey, Catherine Hoyt and Desiree Wagner

ABSTRACT

Larvae of the marbled salamander (*Ambystoma opacum*) are known predators of spotted salamander larvae (*Ambystoma maculatum*) in ponds where these species coexist. In Pennsylvania, vernal ponds are also home to the red-spotted newt (*Notophthalmus viridescens*), which has a variable diet that may include *A. maculatum* larvae. In this study, we assessed the red-spotted newt as a potential predator of the spotted salamander. Red-spotted newts showed a significant prey preference for spotted salamander larvae over the amphipod, a similarly-sized prey alternative. We also evaluated the implications of these findings for the coexistence of *A. opacum*, *N. viridescens*, and *A. maculatum* in vernal ponds.

Key words: *Ambystoma maculatum*, *coexistence*, *predation*, *prey preference*, *vernal ponds*

INTRODUCTION

Though much ecological research has demonstrated that a predator has the ability to reduce or eliminate prey populations in the wild, the coexistence of predator and prey is not atypical. In laboratory studies, several factors are cited as facilitating the coexistence of predator and prey, three of which include (1) “immigration” or the supplementation of the prey population from a source outside the system, (2) prey shelters that allow escape from predators, and (3) the reduction of contact between predator and prey (Stenhouse 1985). In the field, other factors that prolong the persistence of a prey population may include dispersal, increased longevity, repeated reproduction, availability of alternative prey, and fluctuation in predator densities (Stenhouse 1985).

Some salamanders (e.g., marbled salamanders, *Ambystoma opacum*) prey on other salamander species (e.g., spotted salamanders, *Ambystoma maculatum*). Alternative prey (e.g., anuran larvae) and prey refuges have been shown to increase survival

of *A. maculatum* larvae when coexisting with predatory *A. opacum* larvae in the laboratory (Stenhouse 1985). In vernal ponds where spotted and marbled salamanders coexist, extreme variability in *A. opacum* population densities between ponds and between years may occur, suggesting that *A. maculatum* populations might persist by fluctuating in a responsive manner (Stenhouse 1985).

In this study we tested whether red-spotted newts (*Notophthalmus viridescens*) prey on *A. maculatum* larvae. These two species coexist in vernal ponds in Pennsylvania. We specifically tested whether *N. viridescens* shows a food preference for either *A. maculatum* larvae or similarly sized amphipod crustaceans (*Gammarus minus*).

MATERIALS AND METHODS

Field site

This study was conducted in April 2005 at a vernal pond along Petersburg Pike near Huntingdon, PA. The pond is located in an old growth forest with moderate disturbance from logging and light traffic

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from members of a hunting club who own the land. Many species were found in this pond besides salamanders, including wood frogs (*Rana sylvatica*), spring peepers (*Hyla crucifer*), several bird species (e.g., ducks, geese, and belted kingfishers *Ceryle alcyon*), and various insect larvae.

Experimental design

Spotted salamander egg masses were collected from the vernal pond and brought back to the laboratory. Water collected from the pond was used in all subsequent experiments. The egg masses were incubated at room temperature and aerated to speed development. Ten red-spotted newts were also collected from the vernal pond. The prey preference of the red-spotted newt was tested by placing one newt with five spotted salamander larvae and five amphipods in one-liter buckets filled with pond water. Ten independent trials were conducted, along with a control where 10 salamander larvae and 10 amphipods were placed in a bucket. The trials were placed in the refrigerator for 24 hours. The number of

amphipods and larvae still present was recorded, and the condition of remaining organisms was noted.

Statistical analysis

A chi-squared goodness-of-fit analysis (Minitab 14) was used to determine whether *N. viridescens* significantly preferred one prey item over another.

RESULTS

Table 1 shows the number of prey remaining in each individual prey preference trial after a 24-hour period. Significantly fewer *A. maculatum* larvae than amphipods remained after 24 hours (Table 2). No *N. viridescens* mortality was observed. In trial 2 where three larvae were found, all three larvae were dead. Likewise, in trial 7 where four larvae were found, three were dead. In trial 1, the newt shed its skin and one amphipod was dead. Trial 8 also had a dead amphipod. All control amphipods and larvae were present, eliminating the possibility of missing *A. maculatum* larvae or amphipods due to predation between the two.

Table 1. Number of amphipods (*Gammarus minus*) and *Ambystoma maculatum* larvae remaining in each trial bucket after 24-h exposure to predatory *Notophthalmus viridescens*.

Trial	Amphipods	Larvae
1	5	2
2	5	3
3	3	0
4	3	0
5	5	0
6	5	4
7	5	4
8	4	0
9	3	1
10	4	0
control	10	10

Table 2. Results of a chi-squared goodness-of-fit analysis, comparing the frequency of amphipods (*Gammarus minus*) and of *Ambystoma maculatum* larvae preyed upon by *Notophthalmus viridescens*.

χ^2	df	p
11.00397	1	< 0.001

DISCUSSION

In our study, red-spotted newts significantly preferred spotted salamander larvae over the similarly-sized amphipods. Perhaps the amphipods were

regarded as “foreign” prey items because they do not exist in the natal vernal ponds inhabited by red-spotted newts and spotted salamanders. Nevertheless, eight amphipods were consumed. The trials in which the most amphipods were consumed (two) also had no

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spotted salamander larvae remaining, suggesting that the newts ate the larvae first and then consumed amphipods when given no other option. Observations of predation behaviors were not made; in future work, lab and field observations would be useful to qualitatively determine the prey preferences of the red-spotted newt.

Our results suggest that *N. viridescens* is a potential predator of spotted salamander larvae. *N. viridescens* are known to prey on spotted salamander eggs, but little observation of predation on the larval stage has been observed in the wild (Petranka, 1998). *A. opacum*, on the other hand, is known to significantly increase spotted salamander mortality (Stenhouse, 1985). This has important implications in the vernal pond we studied: how can *A. maculatum* coexist in the same pond with two potential predators, *N. viridescens* and *A. opacum*?

Newts are known to have a wide diet range, which also varies between life stages. Because of this, their effects on spotted salamander populations may be minimal. Furthermore, the presence of alternative prey (such as tadpoles and insect larvae) may further reduce predation on spotted salamander larvae. No *A. opacum* larvae were seen in the pond on any collection trip during this study, and no adults were found in the surrounding woods, despite the fact that adults were found near the pond in the previous fall. As suggested by Stenhouse (1985), inter-year differences in predator densities may allow *A. maculatum* populations to

persist. In addition, contact between predators and prey may be reduced in the vernal pond examined in this study because of abundant leaf litter and vegetation, which may act as prey refuges.

It is inherently difficult to reproduce field conditions in the laboratory, and in studies such as this involving predation and species interaction, the natural environment plays a major role in those interactions. Long-term field studies examining density fluctuations between these salamander populations and the role of the biotic physical environment in their predator-prey interactions is necessary to further our understanding of community structure and coexistence in pond habitats.

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COMPARING BODY SIZE TO WEB AREA IN *CYCLOSA CONICA*

James Serfass

ABSTRACT

I investigated the relationship between body size and web area in the spider species *Araneidae cyclosa*. I hypothesized that there would be a positive relationship between web area and body size. I found no relationship between body size and web area in any of four populations in Huntingdon County, Pennsylvania. Web area was unrelated to body mass, body length, and abdomen length. There were no significant differences in any of the measured variables among the four sample populations. However, the mean mass of spiders with stabilimentum decoration on their webs was larger than that of spiders without decorated stabilimenta ($t = 1.46$, $df = 42$, $P = 0.076$). Also, the mean web areas were significantly smaller for those with decorated stabilimenta than for other webs ($t = -2.60$, $df = 30$, $P = 0.007$).

Key words: body size, *Cyclosa conica*, stabilimentum, web

INTRODUCTION

Prey capture is the ultimate function of a spider web (Foelix, 1996). The purpose of my research was to determine if web area is a major factor in this function. I did this by comparing spider size to web area to determine if a relationship exists between the two variables. Without the influence of any other factors, web area should be proportional to prey capture, which would then affect the size of the predator (Foelix, 1996). My hypothesis was that web area would be directly proportional to spider size.

In addition, I investigated whether the presence of a stabilimentum in a web is related to spider size. A stabilimentum is formed by pieces of debris and prey items that are lined vertically through the hub of a web. There are multiple hypotheses as to the purpose of stabilimenta. These include: (1) web advertisement, (2) prey attraction, (3) camouflage, (4) strengthening of the web, and (5) stabilimenta serve no purpose (Eberhard 1990). Foelix (1996) suggests that different stabilimenta serve different functions in different types of orb webs, the chief function among *Cyclosa* species being camouflage. I hypothesized that a larger spider body mass could be the result of

more effective prey capture by webs containing a stabilimentum.

FIELD SITE

Field sites were located in Huntingdon County, Pennsylvania. Three sites (RFS1, RFS2, and RFS3), were located at the Raystown Field Station near the village of Entriken while one (PC) was located at the Juniata College Peace Chapel east of Huntingdon. The sites at the Raystown Field Station were all just north of the Grove Farm facility in a deciduous, second growth forest with significant undergrowth. Sites RFS1 and RFS3 were on a south facing slope while RFS 2 was on the top of the hill. The Peace Chapel site was in a similar type of forest and is also on a south-facing slope. All sites had an area of approximately 100 square meters. Overall, the vegetation was similar between the two sites although the Peace Chapel site had fewer small bushes and more young trees.

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METHODS AND MATERIALS

I collected samples at the Raystown Field Station on October 8, 15, and 28, 2004. The Peace Chapel data were collected on October 21, 2004. The weather was mild on October 8 and 28, with a temperature of approximately 55° F and sunny conditions. On October 15 and 21, conditions were less favorable, with rain and cooler temperatures.

I surveyed each site for webs, and upon discovering a web, I captured each spider in a small sample bottle. I used a compass to determine the orientation of each web. I took the compass bearing perpendicular to the side of the web on which the spider was resting. I also measured the height of the hub of the web above the ground with a tape measure. I used the same tape measure to find the height and width of each web. Only the radial spirals which are used for prey capture were considered; the outlying connecting and supporting threads were not measured.

Web location was designated by the type of plant growth on which the web was constructed. The four categories were: Large trees, those greater than 6 inches in diameter, medium trees, those less than 6 inches in diameter and greater than 5 meters in height, and small trees, those that had a small diameter and were three to five meters high with few branches. Bushes/young trees were less than three meters high and had mostly small branches near their tops and had few leaves. Webs were inspected for captured prey and stabilimentum decoration. Prey was clearly visible and was either wrapped or unwrapped. I considered a stabilimentum decorated if it had captured prey items lined vertically through the hub or had a thick visible line of thread crossing vertically through the hub.

To calculate web area, I used the average radius of the web and estimated the web as a circle

using the formula: $A = \pi r^2$. Lengths were measured using a Leica S4E microscope. Body length was determined by measuring from the farthest tip of the abdomen to the tip of the cephalothorax. Abdomen length was taken from the end of the abdomen to the start of the cephalothorax. Spider mass was determined using a Cahn C-31 Microbalance set to an accuracy of 1 µg for 250 mg. I weighed all specimens within 8 hours of capture.

I compared spider mass to web area using linear regression (Excel, 2003). I considered a relationship significant if $r^2 > 0.70$. I also compared body length to web area and web height to web area in the same way. I compared the sample means for spider mass and web area using a one-sample Z-test (Minitab, 2004). I also conducted a 2-sample t-test (Minitab, 2004) to compare the masses of spiders with decorated stabilimenta vs. undecorated webs as well as web area of webs with decorated stabilimenta to undecorated webs. I used a one-way ANOVA (Minitab, 2004) to compare web area among the four sample groups. I considered the differences to be significant if $P < 0.10$.

RESULTS

The mean body mass of the population was 8.031 ± 1.830 ($\pm$ SD) mg. All sample means were within this SD range. The mean web area was 114.5 ± 71.8 cm². All sample means were within this range, as well. Spider size (body mass, body length) was not related to web area (Fig. 1 and 2). However, the areas of undecorated webs (121.6 cm²) were 38.4% larger than webs with a decorated stabilimentum (74.9 cm²) ($t = -2.60$, $df = 30$, $P = 0.007$). Also, the masses of spiders with webs having a decorated stabilimentum (8.81 mg.) were 11.1% larger than those whose webs were without a stabilimentum (7.83 mg.) ($t = 1.46$, $df = 42$, $P = 0.076$).

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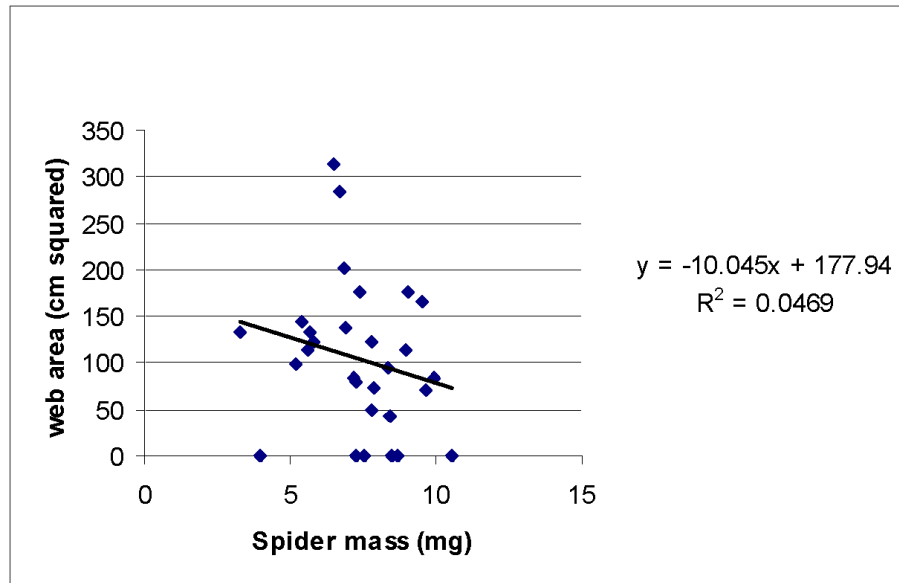


Figure 1. Spider body mass (mg) vs. web area (cm²).

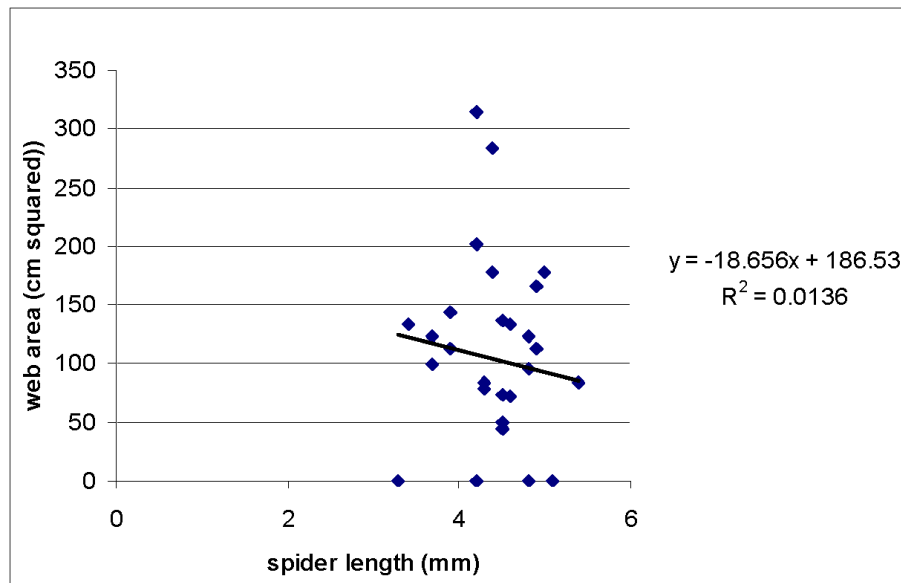


Figure 2. Spider body length (mm) vs. web area (cm²).

DISCUSSION

I found no correlation between spider size (body mass, body length) and web area (Figs. 1 and 2). This finding indicates that factors other than web area influence body size in *Cyclosa conica*. These factors may include web location, competition, and the quality of the web itself. Also, spiders often relocate their webs or change web area based on prey capture (Nakata, 1999). The web on which the spider was

captured may not have contained the same characteristics as the spider's previous webs.

Also, the inclusion of incomplete webs in the data set was a factor that affected the linear regression of body size to web area. I collected five webs at the RFS2 site that did not contain sufficient spiral threads for the web area to be calculated. Therefore, I counted the areas as zeroes. These zeroes could have affected the correlation slightly. However, even if the zeroes were excluded, the data would not produce a

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significant r^2 value when comparing web area to any of the variables in body size.

My findings concerning the decorated stabilimenta are supported by Tso (1998) who found web diameter to be 19% smaller for spiders with decorated stabilimenta and daily insect interception rates 150% higher in those webs. These findings support my results that spiders with decorated stabilimenta have a larger mass probably because more effective prey capture would lead to greater food availability.

ACKNOWLEDGEMENTS

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EFFECT OF BOAT LAUNCHES ON BENTHIC MACROINVERTEBRATES IN A LENTIC SYSTEM

Lauren E. Forster

ABSTRACT

Benthic macroinvertebrates are often used as indicator species when analyzing the effects of possible pollution sources (Nelson, 2000). To test the impact of boat-launch related pollution, I sampled benthic macroinvertebrate populations at six sites on Pennsylvania's Raystown Lake during the Fall of 2004. I used the Biological Monitoring Working Party (BMWP) Score (Chapman, 1996) to assess the pollution tolerance of macroinvertebrate families. The pollution sensitive families Coenagrionidae ($P = 0.012$) and Libellulidae ($P \leq 0.001$) were significantly less abundant in harbors with boat launches. The pollution tolerant families Asellidae ($P \leq 0.001$) and Gammaridae ($P = 0.031$) were significantly more abundant in harbors with boat launches. Diversity and abundance of the other four families sampled were not significantly different between the harbors. This study supports the hypothesis that boat launches affect certain pollution sensitive families.

Key words: Boat launch, Hester-Dendy sampler, lentic, macroinvertebrates, pollution.

INTRODUCTION

Benthic macroinvertebrates are important indicator species of aquatic ecosystem health (Nelson, 2000); their abundance and diversity can be used to assess the effects of possible pollution sources (Wilhm et. al., 1978). Although there have been numerous studies examining the effect of human disturbances on macroinvertebrate communities in lotic systems, little work has been conducted in lentic systems. The lentic environment of Raystown Lake, PA serves as a flood control reservoir and is an important part of the Chesapeake Bay watershed. The lake is also highly used for recreation, including fishing, water sports and boating. The 30-mile long, 8,300-acre lake and its boat launches are used by over 1.5 million visitors a year (USACE, 2004), which can cause a variety of anthropogenic impacts. For example, water contamination by incompletely combusted fuel is known to occur in areas of increased boat traffic such as launches (Secco et. al., 2004). Because Raystown

Lake has been designated as a threatened lake by the state of Pennsylvania (U.S. House of Representatives 1999), it is vital that the health of the lake be monitored closely. The purpose of this study is to examine the effect of boat launches on macroinvertebrate community composition in the lake. My hypothesis is that the presence of boat launches, and the associated boat traffic on Raystown Lake has negatively affected pollution sensitive families of macroinvertebrates.

FIELD SITE

I conducted this study at six harbor sites on Raystown Lake, Huntingdon County, Pennsylvania (Fig. 1). Three harbors containing boat launches and docking areas were chosen as sampling sites. These three harbors were then compared with three harbors without boat launches. The non-launch harbors were relatively similar in size, bathymetry and proximity to each of the boat launch harbors. The boat launches are managed by the U.S. Army Corps of

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Engineers. Harbor 1 is more heavily utilized as a seasonal docking area for boats. The other two are

used primarily for launching and the temporary docking of boats.

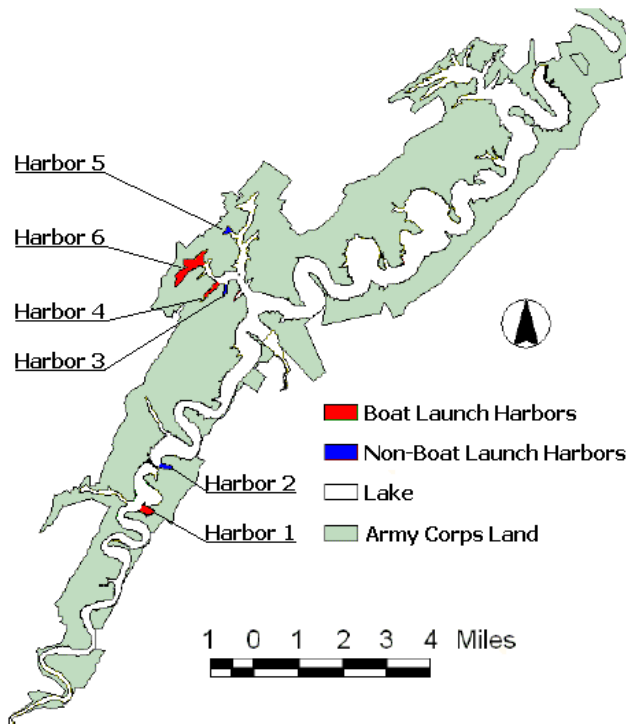


Figure 1. Map of sampling sites on Raystown Lake, Pennsylvania.

METHODS & MATERIALS

I collected benthic macroinvertebrates using Hester-Dendy artificial substrate samplers (Brower et al., 1998, p. 118) at each of 6 harbor sampling sites on Raystown Lake. The samplers each had a total surface area of about 0.12m² and were anchored to bricks (Figure 2) using bailing twine. Three samplers were placed randomly at 1-2.5 m depth in each harbor and were exposed from September 8 to November 3,

2004. I retrieved the samplers from the sites after a period of about 44-46 days. Upon retrieval, the samplers were placed in a plastic bag with lake water. I also conducted water quality measurements at each harbor sampling site; including water temperature, pH, and conductivity, and Secchi disk readings (Table 2). I measured the pH using a HACH EC10 meter (HACH 1996). Conductivity and temperature readings were obtained using an Oakton Acorn series meter (Oakton 1998).

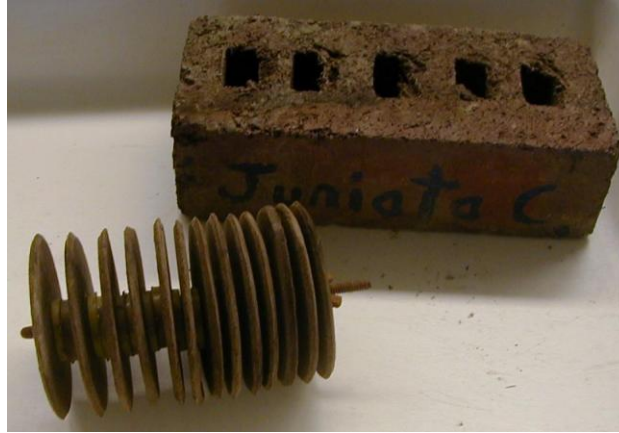


Figure 2. A Hester-Dendy macroinvertebrate sampler with brick anchor.

After collection, the samplers were disassembled and rinsed in a shallow tray to ensure that all the macroinvertebrates were separated and counted. I then preserved the invertebrates in sample bottles containing a 30% ethanol solution. The macroinvertebrate taxa were identified to the family level. I determined family diversity using the Shannon diversity index (Brower et. al., 1998, pp. 180-185). I then compared the abundance per sampler area of each family between harbors with and without boat launches using a Mann-Whitney U Test (Carver 2004). I considered the differences to be significant if $P \leq 0.05$. I used the Biological Monitoring Working Party Scores (Chapman 1996) for each family to assess macroinvertebrate pollution tolerance. The families were scored on a scale of 1 to 10, with 10 being the most sensitive. For the physiochemical data, I used the Welch's t-test to determine if there were any differences between the two harbor types.

RESULTS

Overall, 774 benthic macroinvertebrates from eight families were collected on the artificial samplers at the six sampling sites. The abundances for the two harbor types are given in Table 1. The families Coenagrionidae ($P = 0.012$) and Libellulidae ($P \leq 0.001$) were significantly less abundant in harbors with boat launches. The families Asellidae ($P \leq 0.001$) and Gammaridae ($P = 0.031$) were significantly more abundant in harbors with boat launches. I found no significant difference between any of the other four families of macroinvertebrates. Shannon diversity indices did not differ between harbors with and without boat launches. Libellulidae were absent from the boat-launch harbors, while Asellidae were absent from non-boat launch harbors. Aside from these two families, the other six taxa were all found in both boat-launch and non-boat-launch harbors. The conductivity in the boat-launch harbors was significantly higher than that of the non-boat launch harbors ($t = 2.02$, $df = 11$, $P = 0.034$) (Table 2). There were no significant differences in the pH, temperature or Secchi depths between the two harbor types.

Table 1. Numbers of benthic macroinvertebrates collected from Raystown Lake, Fall 2004.

Order: Family	Common Name	Boat Launch	No Launch	BMWP
Ephemeroptera:Heptageniidae	Mayfly	48	53	10
Odonata:Coenagrionidae	Narrow winged Damselfly	11	33	8
Odonata:Lestidae	Spread winged Damselfly	5	9	8
Odonata:Libellulidae	Skimmer Dragonfly	0	2	8
Diptera:Chironomidae	Midge	289	242	2
Trichoptera:Polycentropodidae	Caddisfly	3	19	7
Isopoda:Asellidae	Sowbug	15	0	3
Amphipoda:Gammaridae	Scud, Sideswimmer	38	7	6

Table 2. Mean physiochemical conditions at each harbor site on Raystown Lake, Fall 2004.

Harbor	Temperature (°C)	pH	Conductivity (µS)	Secchi depth (m)
1-BL*	14.1±1.0	6.5±0.5	230±24.3	1.4
2	13.7±0.0	5.7±0.0	205±0.0	1.4
3	13.4±0.5	6.2±0.7	148±10.2	1.4
4-BL*	13.3±0.4	5.9±0.1	200±26.2	1.5
5	14.3±0.4	7.3±0.4	163±3.8	1.3
6-BL*	14.8±0.1	7.0±0.2	156±0.6	1.3

*BL indicates the presence of a boat launch

DISCUSSION

My results generally support the hypothesis that boat launches affect benthic macroinvertebrate populations. The families Asellidae and Gammaridae appear to be more tolerant of boat launches while Coenagrionidae and Libellulidae are less tolerant of launches. The BMWP Scores (Table 1) provide support for this conclusion by ranking Coenagrionidae and Libellulidae as more pollution-sensitive families than Asellidae and Gammaridae. Nelson and Roline (2003) found similar abundance patterns in their study, which indicated that some families are more tolerant of pollution, like mine runoff, than others. Pinel-Alloul (1996) found that water conductivity is one of the most significant variables effecting macroinvertebrate communities. The increase in conductivity in harbors with boat launches could be due to anthropogenic sources (Topalian et. al., 1998). The Secchi depth readings were conducted on different days, but indicate murkier conditions than normal, which could be attributed to the large flood event that raised the lake level 15ft. during the sampling interval.

Although Shannon diversity did not differ between the harbors, the types of families did. The higher abundance of the pollution tolerant families in boat-launch harbors may be due, in part, to the availability of resources not utilized by other more sensitive families. Non-boat launch harbors have a higher abundance of pollution sensitive families, which may out compete some of the more tolerant families. Pollution sensitivity and perhaps competition play an important role in determining which families are more abundant in the harbor types.

The abundance of the most pollution sensitive of the families, Heptageniidae, was not significantly different between the two harbor types. I expected that there would be an abundance difference for this family of mayflies, because the BMWP gives it a sensitive score of 2. However, with the removal of an outlier in the abundance data, a significant difference ($P = 0.018$) was observed for Heptageniidae. This outlier could have been caused by a variety of factors (e.g., the substrate where the

sampler was placed was rockier than any of the other sites: cf. Pinel-Alloul, et. al., 1996).

A large flood event took place in the first few weeks of sampler exposure, during which the level of the lake rose over 15 feet above normal. This increased the placement depth of the samplers to over 16 feet for about a week and caused an increase in debris around some sampling sites. This flood event could have caused the loss of three uncounted samplers and may have had other unknown effects on my results.

Because limited data are available regarding macroinvertebrate abundance in lentic systems, I compared my results to those conducted in lotic environments. Wilhm et. al. (1978), who used similar sampling methods, found 36 families of macroinvertebrates (using 42 samplers). I found eight families using 15 samplers. The number of families collected in these two studies is similar relative to the number of samplers used.

My study suggests that boat launches affect macroinvertebrate populations. Because benthic macroinvertebrates are indicator species of aquatic ecosystem health, my results suggest that other aspects of the aquatic system (e.g., fish populations) may have been affected, as well.

Further investigation is needed to identify the specific biotic and abiotic factors of boat launches that influence macroinvertebrate abundance. Alternative sampling methods would also provide more data since artificial substrate samplers attract the colonization of some species and discourage others, which introduces a species bias (Brower et. al., 1998). Experimentation during peak seasons of recreation at boat launches may also provide additional insight.

ACKNOWLEDGEMENTS

I thank Dr. Charles Yohn for his guidance with this project, as well as my research assistants Melissa Wilson, Karen Stringer, & Jessica Perry. I also thank the Juniata College Environmental Science and Studies Department and the Raystown Field Station.

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SEASONAL DIET OF STRIPED BASS (*MORONE SAXATILIS*) IN RAYSTOWN LAKE

Bradley Watkin

ABSTRACT

Striped bass (*Morone saxatilis*) show seasonal dietary changes in saline environments (Nelson et al., 2003). The goal of this study was to determine if the landlocked freshwater striped bass in Raystown Lake of central Pennsylvania also show seasonal diet variation. The diets of bass captured during the Spring and Fall were compared based on stomach contents. I found significant differences in prey composition ($\chi^2 = 10.07$, $df = 2$, $P = 0.007$) and median weight (Mann-Whitney $U = 2.93$, $P = 0.047$) of stomach contents. This suggests that seasonal diet change does occur in the striped bass of Raystown Lake.

Key words: Raystown Lake, seasonal diet, striped bass (*Morone saxatilis*)

INTRODUCTION

Located in the Ridge and Valley Region of central Pennsylvania, Raystown Lake is the largest lake solely within the state (Fig. 1.). Its deep, clear water offers superb opportunities for fishing striped bass (*Morone saxatilis*); and is one of America's top 25 fishing destinations as ranked by Field & Stream magazine (Bestul et al., 2003). Adult striped bass feed predominantly on members of the herring family (Clupeidae), such as gizzard shad (*Dorosoma cepedianum*) and alewife (*Alosa pseudoharengus*) (Schultz et al., 2003). Raystown Lake has a large population of these bait fish (Grace 2003). Seasonal dietary changes have been documented in ocean dwelling striped bass (Nelson et al., 2003). The goal of my study was to examine the seasonal diet variation in the landlocked striped bass of Raystown Lake.

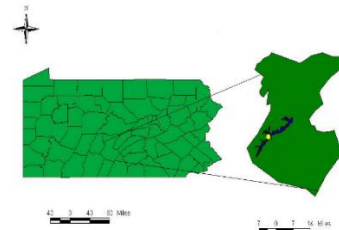


Figure. 1. Location of Raystown Lake in Huntingdon County, Pennsylvania.

METHODS

Striped bass stomachs were obtained from Raystown Striper Club tournaments held at Raystown Lake during Spring 2002 and 2003 and Fall 2003. Captured bass were brought to a check station where the stomachs were removed. I separately bagged and labeled each stomach, preserving them in 70% ethanol and then refrigerated them to avoid post-capture digestion. Each stomach was later dissected and its contents identified using a key to freshwater

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fishes (Eddy 1978). Prey items were grouped by species, blotted dry of excess liquid and then weighed (following Nelson et al., 2003).

To compare the number of prey items between Spring and Fall, I used a Chi Square test (Minitab 2000); and to compare median stomach-content weight, I used a Mann-Whitney U test (Minitab 2000). I considered differences to be significant if $P < 0.05$.

RESULTS

I found a significant difference in the percent occurrence of alewife, gizzard shad and other prey items in the stomach contents of striped bass between the Spring and Fall (Table 1, Fig. 2), ($\chi^2 = 10.07$, $df = 2$, $P = 0.007$; for more details refer to Watkin, 2003). I also found a significant difference in median stomach-content weight of striped bass between the Spring [$16.6 \pm 18.02\text{g}$ ($\pm$ SD)] and Fall ($8.6 \pm 13.45\text{g}$) (Figs. 2 and 3.), (Mann-Whitney $U = 2.93$, $P = 0.047$).

Table 1. Total number of prey items in the stomachs of striped bass (*Morone saxatilis*) in Raystown Lake, Huntingdon County, Pennsylvania. Collected in Spring 2002, 2003 and Fall 2003.

# of Stomachs	Spring N= 52	Fall N= 13
Alewife (<i>Alosa pseudoharengus</i>)	146	17
Gizzard Shad (<i>Dorosoma cepedianum</i>)	11	3
Crayfish (<i>Orconectes virilis</i>)	0	2
Rainbow Trout (<i>Oncorhynchus mykiss</i>)	2	0
Empty stomachs	11	4

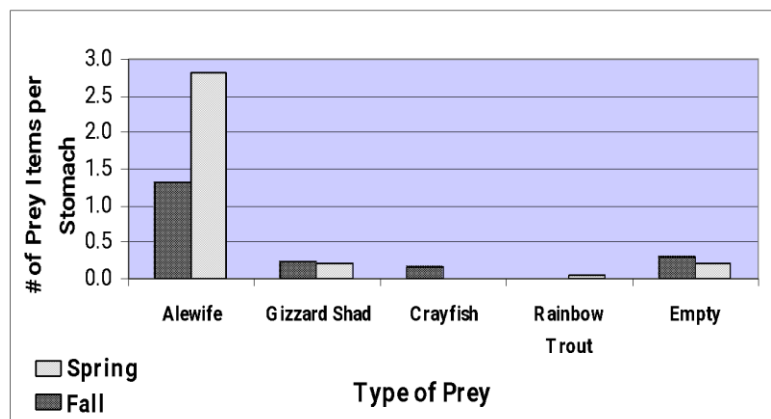


Fig. 2. Mean number of prey items per stomach of striped bass (*Morone saxatilis*) in Raystown Lake, Huntingdon County, Pennsylvania, during Spring 2002, 2003 and Fall 2003.

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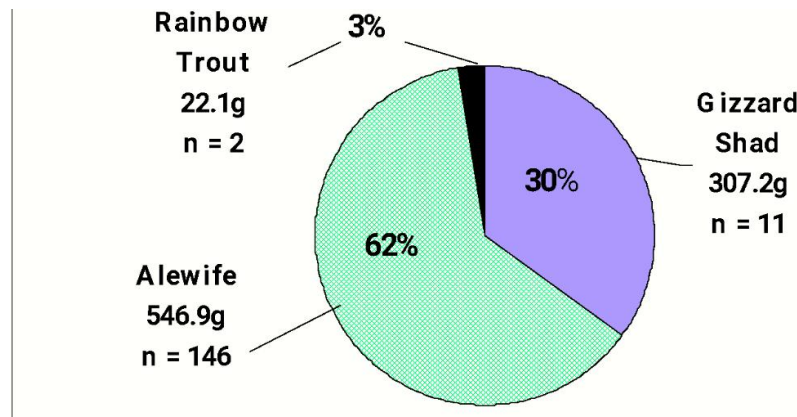


Fig. 3. Percent prey composition by weight in the stomachs of striped bass (*Morone saxatilis*) from Raystown Lake, Huntingdon County, Pennsylvania, during Spring 2002 and 2003.

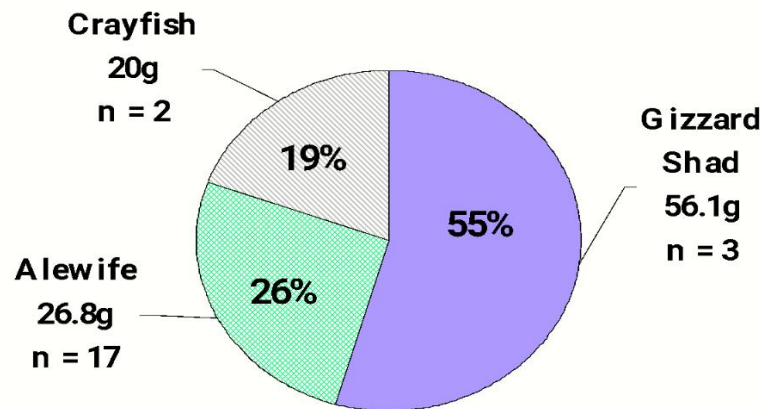


Fig. 4. Percent prey composition by weight in the stomachs of striped bass (*Morone saxatilis*) from Raystown Lake, Huntingdon County, Pennsylvania, during Fall 2003.

DISCUSSION

My results show a diet shift in striped bass between Spring and Fall. Bass in the Spring had fuller stomachs, perhaps due to higher levels of prey availability during that season (cf. Brönmark and Hansson, 1999). Spawning behavior can also account for increased feeding, because spawning occurs in the early Spring. At this time, the bass will increase their food intake to make up for the energy lost for spawning (Nelson et al., 2003). Another factor that affects stomach fullness of bass is their digestion rate. The digestion rate of fish is affected by both meal size and water temperature. Because the water is cooler in the early Spring, the rate of digestion would be slower than that of the Fall. This would also result in the bass having fuller stomachs during the Spring (Hurst and Conover, 2001). A bias

in my study is that at the tournaments, only the largest bass caught were turned in by fishermen. Larger striped bass eat larger prey items (Cyterski et al., 2002). Therefore this diet analysis may not pertain to smaller striped bass in Raystown Lake. The knowledge of striped bass seasonal feeding behavior gained from this study will hopefully aid future sport fishery management at Raystown Lake.

ACKNOWLEDGEMENTS

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OPTIMAL ENVIRONMENT FOR EGG DEVELOPMENT IN SPOTTED SALAMANDERS (*AMBYSTOMA MACULATUM*)

Stacey Galvin, Corinne Hamblet and Jessa McKillop

ABSTRACT

Unhatched Spotted Salamander (*Ambystoma maculatum*) eggs were placed in environments that varied in temperature, pH, and both temperature and pH. Temperature ranged from 2.3 to 22.9°C, whereas pH ranged from 4.65 to 7.12. After 15 days the development of the eggs was analyzed by determining the number of live, dead or unhatched eggs. A chi-square analysis showed that the different experimental environments had a significant effect on egg development.

Key words: Spotted Salamander, pH, temperature, egg development, *Ambystoma maculatum*

INTRODUCTION

The spotted salamander (*Ambystoma maculatum*) is one of the most abundant species of salamander in Pennsylvania. Adult spotted salamanders mate during late March to early April once temperatures begin to rise, and heavy rains have fallen. Temporary (vernal) ponds are critical habitats for reproduction and egg development in this species (Horne and Dunson, 1995). Females lay large, globular egg masses, containing up to 200 eggs that are attached to submerged branches. The eggs take two to seven weeks to hatch, depending on varying environmental factors (Egan, 2004).

Several studies have been done to determine the effects of both varying pH and temperature on spotted salamander reproduction and development in vernal ponds. While some studies claim that pH is more important (Clark, 1986), others have concluded that temperature is the best predictor of hatchling success (Albers, 1986). Low pH slows egg development, thereby limiting the breeding success of amphibians. Because the egg sacs surrounding the embryos are semi-permeable, they are particularly vulnerable to habitat contamination (Turtle, 2000). Furthermore, ponds are particularly susceptible to acidification due to acid rain, leaf litter, runoff and other causes (Horne, 1995; Turtle, 2000). In addition, both hatchling size and developmental rates are affected by varying temperatures. Normal limits for successful hatching

were found to be in the range of 4 or 5°C to 24 or 25°C. Drying of temporary ponds also influences larval survival rates from year to year. As vernal ponds dry towards the end of the summer season, salamander survival depends on whether the larvae had time to reach metamorphosis, which takes up to three months after egg hatching (Light, 1989).

We tested the effects of changes in pH, changes in temperature, and changes in both pH and temperature in comparison to a simulated natural habitat. Our study was conducted to determine which of these factors is most influential. We hypothesized that changes in the environment would affect the survival rate of spotted salamander eggs, with temperature causing more of a negative effect than pH.

FIELD SITE

On March 29, 2005 Spotted Salamander eggs were obtained from a vernal pond along Petersburg Pike about 1.7 miles from Cold Springs Road. The water in the vernal pond was relatively clear and not moving. The surrounding environment was full of bushes, and other ponds that did not contain any eggs.

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METHODS AND MATERIALS

Four tanks were labeled as “control,” “tank 1” (pH and temperature variant), “tank 2” (pH variant) and “tank 3” (temperature variant). Tanks 2 and 3 were filled with water from Petersburg spring. Tank 1 and the control were filled with water from the vernal pond. About 80 spotted salamander eggs were collected from the vernal pond. They were all from the same location where some vegetation was also collected. Approximately 20 eggs were placed in each tank along with a handful of vegetation, and the tanks were placed in a walk-in cooler (10° C). Tank 2 and the control tank were left in the cooler throughout the whole experiment. Tanks 1 and 3 were moved between an inner room in the cooler (~3° C) and the lab (~25° C) daily. Each day for 15 days, we measured water pH and temperature, and recorded the number

of eggs hatched in each tank (see Table 1). On days four and ten, new water from each source was collected and the tank water was changed. On the final day, we recorded the total number of eggs hatched, along with the number of dead and live larvae present (see Table 5). Minitab was used to perform an analysis of variance to test for significant differences.

RESULTS

The daily pH and temperature and their overall means for the control and treatment tanks are given in Tables 1 to 4. Table 5 displays the status of egg development in each tank at the end of the study. A chi-square analysis demonstrated that the varying experimental conditions had a significant effect on survival rate ($\chi^2 = 33.12$, $P < 0.001$).

Table 1. Daily pH and temperature in the control tank

Day	Temperature	pH
1	0.00	0
2	10.20	6.14
3	9.80	6.05
4	9.90	7.12
5	9.30	6.11
6	10.10	6.32
7	10.30	6.48
8	9.30	6.74
9	10.50	6.12
10	8.90	5.01
11	9.70	6.06
12	9.90	6.21
13	9.40	6.44
14	9.30	6.78
Average	9.043	5.827
Standard Deviation	2.642	1.745

Table 2. Daily pH and temperature in the tank with varying pH and temperature

Day	Temperature	pH
1	0.00	0
2	2.80	5.55
3	20.40	5.67
4	2.70	6.86
5	9.30	6.34
6	2.50	6.24
7	20.30	6.09
8	3.10	5.44
9	20.30	6.03
10	3.30	4.65
11	22.90	5.48
12	3.10	5.94
13	21.80	4.96
14	2.80	5.28
Average	9.664	5.324
Standard Deviation	9.107	1.637

Table 3. Daily pH and temperature in the tank with relatively high pH

Day	Temperature	pH
1	0.0	0
2	10.1	6.18
3	10.2	6.23
4	9.8	6.94
5	9.6	6.65
6	9.9	6.54
7	9.8	6.65
8	9.0	6.88
9	9.8	6.18
10	9.1	5.37
11	9.4	6.49
12	10.1	6.18
13	9.2	6.46
14	9.3	6.9
Average	8.95	5.975
Standard Deviation	2.605	1.767

Table 4. Daily pH and temperature in the tank with varying temperature

Day	Temperature	pH
1	0.00	0
2	2.80	6.04
3	21.30	6.13
4	2.60	6.96
5	8.70	6.53
6	2.20	6.32
7	20.80	6.37
8	3.20	6.13
9	21.40	5.78
10	3.30	5.31
11	22.80	6.55
12	3.40	6.31
13	22.60	5.86
14	2.80	6.49
Average	9.85	5.77
Standard Deviation	9.415	1.708

Table 5. Status of egg development at the end of the study period (15 d).

Egg Status	Control Tank	Temperature and pH Variant	pH Variant	Temperature Variant
Unhatched	16	1	12	0
Hatched	4	9	6	18
Dead	1	0	0	1

DISCUSSION

Temperature and pH had significant effects on survival rates of spotted salamander eggs (Table 5). The control tank showed the least amount of hatching compared to the other tanks. This may have been because the control tank had a constantly low temperature that inhibited egg development. The two tanks with variable temperatures showed more hatching, perhaps because periodic increases in water temperature facilitated development. The pH seemed to have relatively little effect on egg development, as indicated by the similar frequencies of hatching between the control tank and the pH variant tank, and between the temperature variant tank and the temperature and pH variant tank (Table 5). However, pH was varied relatively little in our study. Future studies should examine the effects of more extreme variation in pH, and also the effects of different constant temperatures.

Sources of error in our study could be lack of sunlight for the control and variant pH tanks, uncontrolled variation of pH in the pH variant tank,

and accidental loss of eggs while changing the water of the control tank.

Similar to our study, Horne and Dunson (1995) reported no effect of pH on spotted salamander development time, though survival rate was negatively impacted. Similarly, Clark (1986) found that at a pH of 4.51, hatchling success was 80% and only slightly higher (88%) at a pH of 6. Yet another study reported that 90% hatching success at pH levels of 7, 8, and 9, but at a pH of 5.5, mortality rates rose to 43.7% (Pough, 1976). On the other hand, a study conducted in Maryland determined that embryo survival was not correlated with pond pH ranging from 3.66 to 5.18 (Albers, 1986).

However like our study, Light and Bogart (1989) showed that *Ambystoma* developmental time is influenced by water temperature. In their study, embryos hatched earlier, but at a smaller size as temperature increased.

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TREE DIAMETER IN RELATION TO SOIL MOISTURE ALONG AN ELEVATIONAL GRADIENT

Kyle Narsavage and Laura Weber

ABSTRACT

Tree diameter was compared to percentage soil hydration along an elevational gradient on Juniata College's Property near Seven Points, Huntingdon County, Pennsylvania. It was hypothesized that larger tree diameters and higher soil moisture would be found closer to a stream at the bottom than the top of our study slope. However, no significant trends were observed. Along two transects, tree diameter did not vary significantly with distance from the stream (transect 1: $P = 0.499$; transect 2: $P = 0.762$). In addition, we detected no significant variability in soil moisture with respect to distance from the stream.

Key words: Elevation, Seven Points, soil moisture, tree diameter

INTRODUCTION

Plants depend on soil moisture for growth and survival (Salter, 1940). A lack of soil moisture has a strong effect on tree growth where trees are accustomed to damp soil conditions and not adapted to live in arid environments. Trees also have the ability in some locations to tap into the ground water table to obtain their necessary water requirements. When water tables are too deep for root systems to reach, trees are vulnerable to drought conditions because they depend solely on soil hydration supplied from precipitation and runoff (Alavi, 2001). Tree-species composition, tree-stand age, and tree density, as well as human harvesting methods are factors that contribute to determining amounts of water runoff. Trees deplete the amount of water present in the soil when they absorb it into their root system. Different transpiration rates and root depths of specific species cause different rates of depletion. Replenishment of soil-water storage is dependent on canopy cover and varies greatly between species. (Schume, 2003) The frequency of soil moisture renewal, either by rainfall or irrigation, and the ability of the soil to absorb water, is very essential to the survival of plants. Another important factor for

the survival of plants is the rate at which the water moves through the soil. The force of gravity controls this factor (Salter, 1940).

Seasonal flooding has both its benefits and drawbacks related to soil moisture. Short-term flooding has the capability of increasing soil moisture and the deposit of nutrients, whereas long-term flooding can be potentially harmful, reducing the amount of dissolved oxygen (Keeland & Sharitz, 1997). Additionally, long-term flooding can increase the resistance of the soil to water. In other words, it can lead to increased movement of water across the root cortex; however, this allows for less absorption (Keeland & Sharitz, 1997). In addition, changes in water levels can influence the evapotranspiration demand, which may affect tree growth. High levels of evapotranspiration demand not only results in stomatal closure, but also reduced tree growth (Keeland & Sharitz, 1997).

In this study we hypothesized that trees near a stream at the bottom of a forested slope would be larger because of higher soil moisture than those near the top of the slope.

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FIELD SITE

In April 2005, data were collected from a forested slope on Juniata College's property at Seven Points, located in Huntingdon, Pennsylvania.

METHODS AND MATERIALS

On April 6, 2005, data were taken along two transects extending upward from a stream at the bottom of a slope to the top. Each transect was divided into three quadrants that were 10 meters apart. Quadrant 3 was more elevated than quadrant 1, which was closer to the water source. The dimensions of a single quadrant were 10 m wide by 5 m deep. Within each quadrant, observations were taken on the density of the trees and the ground cover. Tree diameter was calculated from the circumference of the tree. Additionally, four soil samples were randomly collected from each quadrant and placed into airtight containers. The samples were weighed before and

after they were dried in a microwave oven. Percentage soil hydration was calculated as

$$[(M_t - M_d)/M_t] \times 100\%,$$

where M_t equals total soil mass, and M_d equals dry soil mass.

The data were analyzed using MINITAB 14. Since frequency histograms of tree diameter were not normal (both skewed to the right: see Figs. 1 and 2), nonparametric Kruskal Wallis tests were used to performed on the data for both transects, individually.

RESULTS

Tree diameter did not vary significantly with elevational quadrat along either transect 1 ($H = 1.39$, $df = 2$, $P = 0.499$) or transect 2 ($H = 0.54$, $df = 2$, $P = 0.762$). Soil moisture showed no obvious relationship to distance from the stream, as well (Table 1).

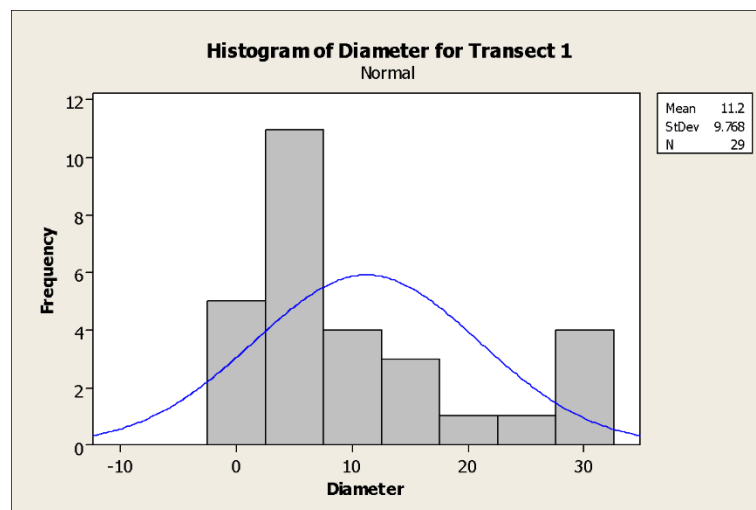


Figure 1. Frequency histogram for tree diameter for transect 1.

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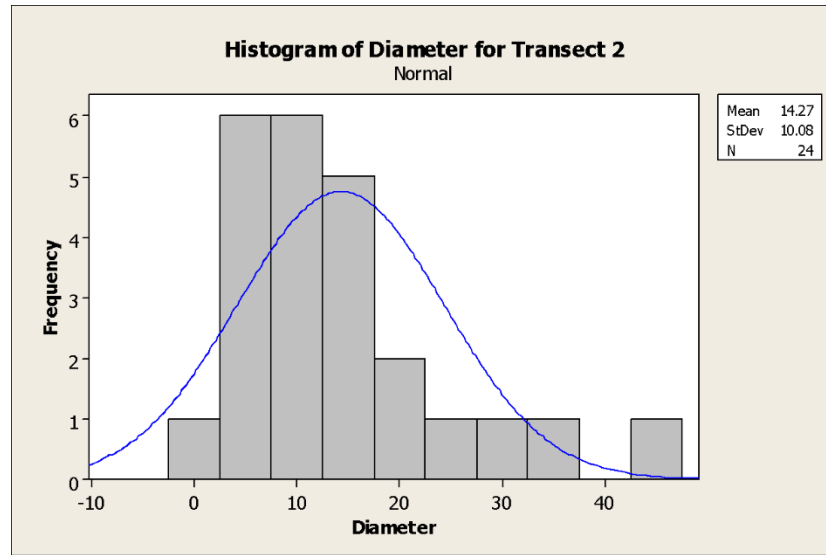


Figure 2. Frequency histogram for tree diameter for transect 2.

Table 1. Mean percentage soil hydration for each quadrant along transects 1 and 2.

Soil Sample (Transect – Quadrant)	Average Percentage of Soil Hydration
1-1	41
1-2	21
1-3	35
2-1	35
2-2	35
2-3	39

DISCUSSION

Neither tree diameter nor soil moisture showed any relationship with elevation along the study slope. There are several possible explanations for these results. First, differences may not have been observed because all of the data were collected on only one day. Second, consideration was not given to several possible confounding factors including size and shape of quadrats, variation in tree species and age, soil characteristics, precise variation in elevation, human disturbance, and the amount of sunlight that penetrates through the ground coverage.

If this study were to be repeated, the species of each tree should be identified. Also, if the quadrants were larger and spaced further apart up the mountain slope, more variation may have been detected. In addition, the results may have differed, if transects were studied in locations other than Seven Points, especially those with a greater elevational

range. Furthermore, longer-term sampling of soil moisture may pick up trends not noticed in this study.

ACKNOWLEDGMENTS

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MOUNTAIN LAUREL ABUNDANCE ON A NORTH VERSUS A SOUTH FACING SLOPE

Michael Chatt and Eric Walberg

ABSTRACT

Mountain Laurel (*Kalmia latifolia*) is an important plant in the ecology of the Appalachian region, especially on late successional slopes. It can be a threat to the biodiversity of rocky slope habitats, and has become problematic to conservation in areas where management has reduced the disturbances that would otherwise control them. Knowledge of the factors affecting their abundance is therefore important. Our study examined the relative abundance of *K. latifolia* on north versus south facing slopes to determine the reaction of this species to associated microclimatic differences. We surveyed two transects, each spanning a north and a south facing slope in Central Pennsylvania. *K. latifolia* was significantly more abundant on south versus north facing slopes.

Key words: Altitude, abundance, microclimate, mountain laurel (*Kalmia latifolia*)

INTRODUCTION

Environmental conditions are known to vary with topographical features, creating microclimatic variation over relatively small ranges. Topographical factors known to have significant effects include altitude, slope direction, and distance from water. For example, Brannon (2002) found that south facing slopes tended to be warmer and drier than north facing slopes. Smokey shrews were found to prefer the moist habitats on northern slopes and near streams.

In contrast, Ford et al. (1999) found that mountain laurel (*Kalmia latifolia*) tended to prefer upper, xeric, south facing slopes in the southern Appalachians. However, Gill (2000) found that in the Piedmont area in Georgia these plants preferred north facing slopes that were also relatively dry. Both studies agree that laurels prefer high slopes and ridges (see also Day et al., 1974; Neary et al., 1984). While we cannot, therefore, say specifically that *Kalmia latifolia* will always prefer one slope over another, it would be useful to know which slope they prefer in central Pennsylvania. This is because, at least in the Appalachian mountains, *K. latifolia* is a very important plant: it grows in the shady under-story of pine-growth areas where it prefers the same acid soils,

and inhibits seedling germination of other plants (Brose et al., 2001). Because of this, it was predicted by Brose et al. (2001) that many areas that have remained undisturbed by fire were slowly becoming laurel fields. The reason for this is because, as a dense, low growth, dry-wood shrub, *K. latifolia* is extremely vulnerable to fire, so much so that burning is a prescribed method of mountain laurel control (Ford et al., 1999). However, because of human intervention in fire management, *K. latifolia* have in many areas begun to dominate habitats and will continue to do so unless disturbance by fire is reintroduced into those areas (Brose et al., 2001).

Our study focused on identifying a difference between laurel growth on north versus south facing slopes: we hypothesized that laurels would prefer the warmer, brighter conditions of south facing slopes over the opposite conditions present on north facing slopes. This could be used to help identify in our area problematic spots in which manual maintenance may be necessary to maintain a desired habitat type. If, however, no variation is detected, it would suggest that the microclimates do not affect mountain laurel, or were not different enough to do so.

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FIELD SITE

The first field site (transect 1) was accessed via PA Route 26, south of State College, Pennsylvania. The sampling area extends from N40° 43' 32.0", W77° 52' 49" at the bottom of the north facing ridge to N40° 43' 25.3", W77° 52' 40.0" at the top. The south facing ridge extends from N40° 43' 33.6", W77° 52' 37.0" at the bottom to N40° 43' 19.0", W77° 52' 41.8". On both sides the sample transect ranged from 507 to 629 m altitude. Both the north and south facing hillsides are comprised of boulder fields through which the sparse but significant flora protrudes, including trees. The hill is facing such that it is nearly due north and due south facing. The second field site (transect 2) began 500 m east and 100 m north of the first, and ran parallel to the first. It was of equivalent terrain to transect 1, but began at 523 m altitude and ended at 648 m altitude.

METHODS AND MATERIALS

Along each sample transect, we surveyed mountain laurels every 3 m in elevation (41 sample sites per slope or 82 samples per transect 1; and 42 samples per slope or 84 samples per transect 2), at which we counted the laurel stems protruding from the ground within a 3-m radius. Abundance of laurels was calculated as laurels per m². Density comparisons between north and south facing slopes were made by using paired t-tests.

To determine temperature differences at different altitudes, we placed max-min thermometers at each endpoint location of the first field site. Unfortunately, due to excessive warming of the leaves we covered them in by the sun, only minimum temperature data can be considered viable, and only data from the south side is of use because, on the north side, only one data point could be collected at the top, and the thermometer at the bottom was lost, and then found to be broken, yielding no results.

In an attempt to assess soil differences, we took three soil samples from each of six sites: at the four endpoints and at 580m altitude. These were tested for pH (using the LAB-AIDS Chemplate method), and examined for qualitative differences in moisture and texture, though actual moisture could not be determined because of lack of time.

RESULTS

Along transect 1, laurel abundance was significantly greater on the south facing ridge [0.714 ± 0.257 ($\pm$ SD) stems m⁻²] than on the north facing ridge (0.027 ± 0.055 stems m⁻²) ($t = 15.8$, $P < 0.001$). Similarly along transect 2, laurel abundance was significantly greater on the south facing side (0.713 ± 0.356 stems m⁻²) than on the north facing side (0.058 ± 0.104 stems m⁻²) ($t = 11.4$, $P < 0.001$).

Soil pH varied with the altitude at which the sample was taken. Values for the highest altitude endpoints were consistently 5.5-6, values for the middle altitudes were 6-6.5 on the north side and 5.5-6 on the south, and values for the lower endpoints were 5.5-6 on the north and 5-5.5 on the south. The soil in all places was found to be similar in properties and in gradient: a layer of non-decomposed and partially decomposed matter, followed by a very dark grey layer, followed by a layer of light brown, sandy, rocky substrate. Because the pH of the lower layer tended to be higher than the top layer, only values for the topsoil were reported.

Temperature data were taken only from the south facing slope, and could not be subjected to statistical analysis. The upper endpoint temperature averaged four degrees cooler than the lower endpoint temperature for all minimum temperature values, and maximum temperature values were disregarded because they were unreliable. No data could be collected for the North facing slope, therefore we have no data comparing the temperature of north and south facing slopes.

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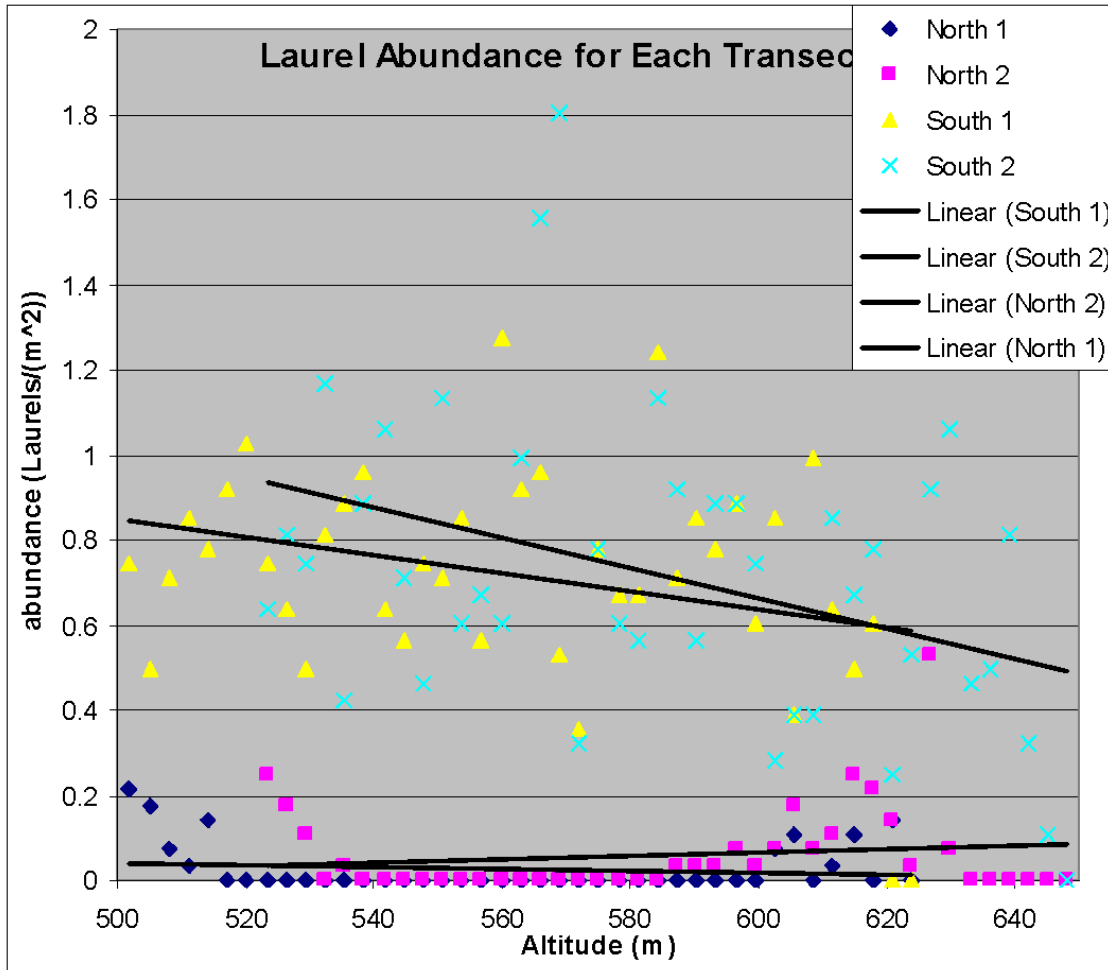


Figure 1. Laurel abundance, at each sampling altitude for all four slopes.

DISCUSSION

We found a significant difference between the abundance of Mountain Laurels on north versus south facing slopes. It is obvious that south facing slopes had significantly higher occurrence of Laurels. Because sampling was repeated in a second area, the data are reproducible, and remained extremely significant.

We were unable to take into account all the factors we wanted. As mentioned, temperature could not be used because of sampling difficulties. Our max-min thermometer on the bottom of the north slope was lost and when found, was found to be broken (and had recorded no data), and on our first trip we forgot to place a thermometer at the upper endpoint of the north facing slope. Thus we had only a single point for the north facing slope. As well, because of solar heating of the substrate, the thermometers registered much higher day-temperatures than were actually

possible, thus only minimum temperatures could be used for comparison. The sensitivity of the method for measuring pH was low and would not detect subtle differences, and had a subjective scale. Also, the attempt at soil moisture content had to be aborted because the soil did not dry out. Therefore we cannot really rule out soil chemistry as having an effect on the observed abundance. Further study is clearly needed.

Our second sample area may have been too close to our first for local variations to be overcome: both are on the same hill, half a kilometer apart. It was access that limited us to that area, so further studies should sample a variety of areas to see whether our results can be generalized. Nonetheless, our data do seem to demonstrate that laurels prefer the conditions existing on south facing slopes as opposed to those of north facing slopes, at least in the mid-Appalachian area that we investigated.

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ACKNOWLEDGEMENTS

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EVALUATION OF SPRUCE CREEK WATER QUALITY BASED ON MACROINVERTEBRATE CHARACTERISTICS

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ABSTRACT

Macroinvertebrates are indicative of the overall health of stream and river systems due to their tendency to remain stationary within an area and their ability to respond quickly to stress. These organisms were used to examine the overall quality of Spruce Creek and one of its main tributaries, Warriors Mark Run. According to previous water-chemistry data, Spruce Creek should have macroinvertebrate populations that indicate better water quality than Warrior's Mark Run. Macroinvertebrates were collected on 2 dates in February of 2005 at 2 different sites in Spruce Creek and 3 different sites in Warrior's Mark Run. Three different methods of stream classification by macroinvertebrates were used (Taxa Richness, No. EPT, PTI). Spruce Creek generally showed a significantly better water quality than Warrior's Mark Run.

Key words: Agricultural pollution, macroinvertebrates, nutrient loading, Spruce Creek, water quality

INTRODUCTION

Water quality has become a major concern across the United States, and long term monitoring of surface water quality is critical in determining if aquatic systems are being degraded. Monitoring projects throughout the country test for many chemical and physical factors in streams, but these may not necessarily relate closely to the overall health of the stream. Other factors, including short duration events, may affect organisms in ways which periodic analysis of physical and chemical characteristics cannot fully assess. An alternative approach is to use a biological population, such as benthic macroinvertebrates, to evaluate stream quality. Macroinvertebrate studies are often used to evaluate overall water quality since these organisms can integrate the impacts of short term variations over long time periods. Not only do they respond quickly to stress but they remain relatively stationary during their larval stages (Yandora, 1997). As a result, macroinvertebrates are considered to be indicator species – i.e., species that can identify the state of the habitat they currently occupy (Indicator Species, 2005).

Macroinvertebrates can be used in several ways to characterize a stream. The Global Rivers Environmental Education Network (GREEN, 2005) uses a Pollution Tolerance Index (PTI) in which a stream earns a certain number of points depending on the species present (GREEN, 2005). The United States Environmental Protection Agency (EPA) recommends a Taxa Richness method or a Number Ephemeroptera, Plecoptera, and Trichoptera (No. EPT method). The Taxa Richness method measures the total number of species present in a sample. The Number EPT is calculated from the total types of mayfly (Ephemeroptera), stonefly (Plecoptera) and caddisfly (Trichoptera) larvae within a sample (Resh et al., 1995). Taxa richness and the number EPT, when used in conjunction with one another, become an accurate estimator of stream water quality; Taxa richness takes into account the effect on the entire community, whereas Number EPT takes into account the effect on pollution sensitive species (NC DEM, 2001).

Spruce Creek is a spring-fed stream located in Central Pennsylvania and is classified as a high quality cold water fishery in the Pennsylvania Code (Drainage List, 2005). Warriors Mark Run is one of the major tributaries associated with Spruce Creek. This watershed is of special concern due to the

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exceptional trout fishery located within its waters. Spruce Creek runs through the property of a concentrated animal feeding operation (CAFO) while Warriors Mark Run is mainly surrounded by traditional agriculture. Both of these operations may be the cause of degradation of the entire system (Morse, 1996). A baseline water-chemistry database is currently being constructed (Diesel et al., in preparation), but a macroinvertebrate study has not been conducted in many years.

Water chemistry of Spruce Creek and Warriors Mark Run has provided a starting point for assessing water quality, and this study attempts to corroborate that work by using macroinvertebrate communities, and provides additional information on the health of the aquatic biota in the 2 streams. Based on previous stream chemistry work, the macroinvertebrate populations should indicate that

Spruce Creek has better water quality for sustaining aquatic biota.

METHODS

On Spruce Creek, sites 1 and 2 were chosen to represent a generally unaffected macroinvertebrate community (site SC 1, Fig. 1), and a macroinvertebrate community below the CAFO (site SC 2, Fig. 1). On Warrior's Mark Run, sites 5, 6, and 7 were chosen to represent a generally unaffected macroinvertebrate community (site WMR 7, Fig. 1), a macroinvertebrate community affected by urban land use (site WMR 6, Fig. 1), and a macroinvertebrate community affected by traditional agricultural land use (site WMR 5, Fig. 1). Sites WMR 6 and 7 are on two different branches of Warrior's Mark Run that converge above site WMR 5.

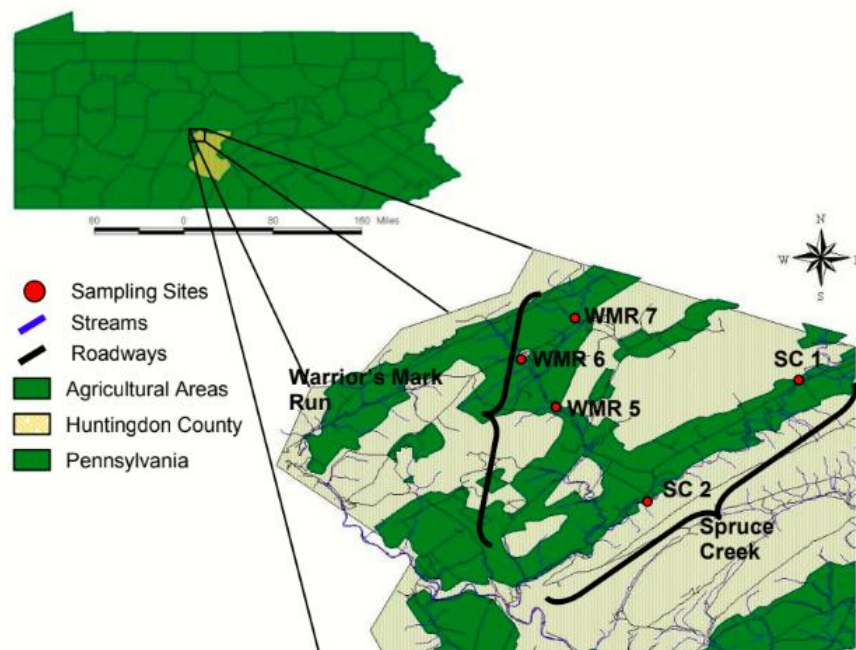


Figure 1. Sampling sites on Spruce Creek and Warrior's Mark Run

Macroinvertebrate samples were collected during 2 sampling periods in February 2005, using the single habitat approach (Plafkin et. al., 1989). This method requires that sampling occur downstream from a riffle with a cobble substrate; suitable stretches of stream were found at all 5 sampling locations. Macroinvertebrates were collected from 6 separate locations in a 10-m long reach downstream of the riffle using 500 μ m D-nets. Each of the 6 sampling periods entailed kicking for 10 seconds in a 0.5 by 0.5

m² area directly upstream from the D-net. The macroinvertebrates that were collected during this time were then placed in to Ziploc™ bags filled with stream water; these bags were kept in a cooler until they could be separated in the laboratory.

The macroinvertebrates were then separated by species for each sampling location. One or more of each of these organisms was placed in a specimen vial and preserved with a 30% ethanol solution for identification. All organisms were then identified to

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order. All data was tested for normality using a one-sample Kolmogorov-Smirnov Test and for differences between the 2 streams using a one-tailed Welch's T-test in SPSS 13. Differences were considered significant at $\alpha \leq 0.05$.

RESULTS AND DISCUSSION

A total of 33 orders of macroinvertebrates were found within Spruce Creek and Warriors Mark Run combined, and included species that ranged from being pollution sensitive to tolerant (Table 1).

Table 1. Orders and families of macroinvertebrates found within Spruce Creek and Warriors Mark Run.

<i>Ephemeroptera Ameletidae</i>	<i>Trichoptera Odontoceridae</i>	<i>Coleoptera Psephenidae</i>
<i>Ephemeroptera Baetidae</i>	<i>Trichoptera Philopotamidae</i>	<i>Coleoptera Unknown</i>
<i>Ephemeroptera Ephemerellidae</i>	<i>Trichoptera Polycentropodidae</i>	<i>Decapoda Astacidae or Cambaridae</i>
<i>Ephemeroptera Heptageniidae</i>	<i>Trichoptera Rhyacophilidae</i>	<i>Diptera Athericidae</i>
<i>Plecoptera Perlidae</i>	<i>Trichoptera Uenoidae</i>	<i>Diptera Ceratopogonidae</i>
<i>Trichoptera Brachycentridae</i>	<i>Acarina Hydrachenellae</i>	<i>Diptera Chironomidae</i>
<i>Trichoptera Cinnephilidae</i>	<i>Amphipoda Haustoriidae</i>	<i>Diptera Simuliidae</i>
<i>Trichoptera Glossasomatidae</i>	<i>Annelida Oligochaeta</i>	<i>Diptera Tipulidae</i>
<i>Trichoptera Hydropsychidae</i>	<i>Coleoptera Dryopidae</i>	<i>Megaloptera Corydalidae</i>
<i>Trichoptera Hydroptilidae</i>	<i>Coleoptera Elmidae</i>	<i>Planaria Tubellaria</i>
<i>Trichoptera Limnephilidae</i>	<i>Coleoptera Gyrinidae</i>	<i>Prosobranchia Bithyniidae</i>

Table 2. Average richness, EPT, PTI Score, water quality rating and total nitrogen concentrations for each site on Spruce Creek and Warriors Mark run.

Site	Average Richness	Average EPT	Average PTI Score	PTI WQ Rating	Average [NO ₃] [*]
Spruce Creek 1	16.0 ± 1.4	7.5 ± 0.7	17.0 ± 1.4	Good	7.4 ± 2.8
Spruce Creek 2	17.5 ± 0.7	8.0 ± 0.0	24.0 ± 0.0	Excellent	7.7 ± 3.0
Warriors Mark Run 7	15.5 ± 3.5	6.0 ± 1.4	18.5 ± 3.5	Good	8.0 ± 2.8
Warriors Mark Run 6	11.5 ± 2.1	6.5 ± 0.7	13.5 ± 3.5	Fair	8.2 ± 2.9
Warriors Mark Run 5	11.0 ± 1.4	4.0 ± 0.0	14.0 ± 1.4	Fair	10.1 ± 3.3

*Average concentrations for 1 year. n = 80 (Diesel et al., in preparation)

Average taxa richness for Spruce Creek was significantly higher than the average taxa richness for Warriors Mark Run ($P = 0.090$) (Table 2, Fig. 2). Spruce Creek showed a significantly higher

average No. EPT than Warriors Mark Run ($P = 0.088$) (Fig. 3). These methods of classification together may indicate that Spruce Creek does have better water quality than Spruce Creek.

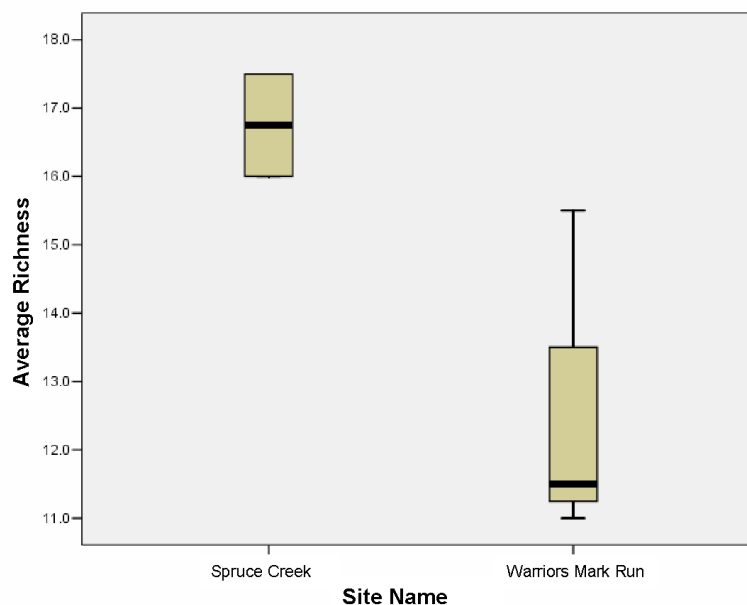


Figure 2. Average taxa richness for Spruce Creek and Warriors Mark Run.

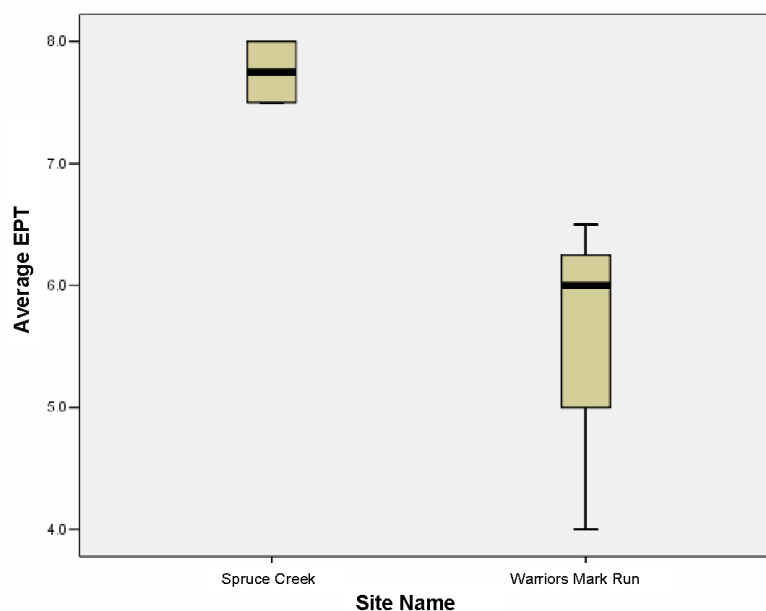


Figure 3. Average No. EPT for Spruce Creek and Warriors Mark Run.

Warriors Mark Run and Spruce Creek did not significantly differ in the average PTI score, but water quality classifications were generally higher in Spruce Creek (Table 2, Fig. 4). Spruce Creek ranged from Good to Excellent water quality while Warriors Mark Run sampling sites were classified in the Fair to Good categories. This type of classification generally has more error because of its broad definitions. For

instance, the GREEN PTI method ranks all caddisfly larvae as pollution sensitive, when in fact net-spinning caddisflies are somewhat sensitive to pollution (AMRC 2005). Net-spinning caddisflies were present in the 2 creeks, which may suggest this method of classification for Spruce Creek and Warriors Mark Run is less accurate.

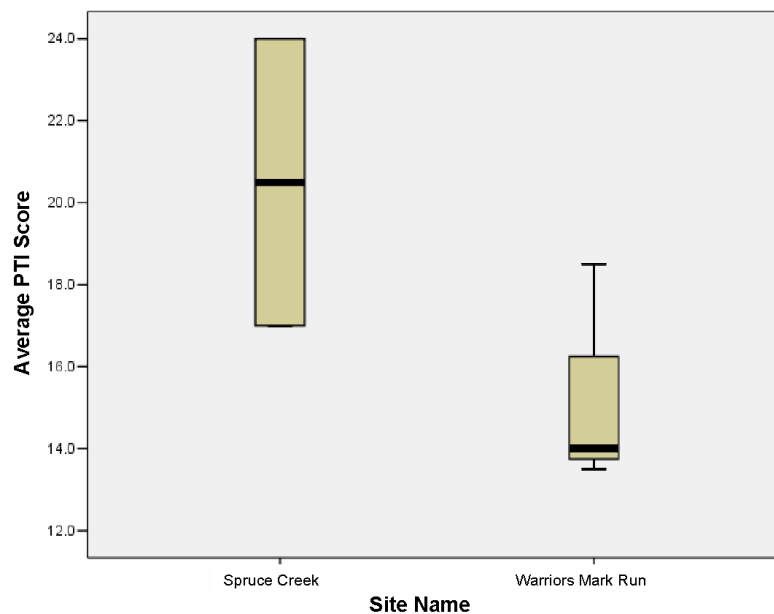


Figure 4. Average PTI score for Spruce Creek and Warriors Mark Run.

Total nitrogen concentrations in Warriors Mark Run tend to be higher than total nitrogen concentrations in Spruce Creek, indicating that Warriors Mark Run has a higher nutrient load. This

aspect of water chemistry suggests that Spruce Creek has better water quality than Warriors Mark Run (Table 2, Figure 5). The macroinvertebrate results agree with this.

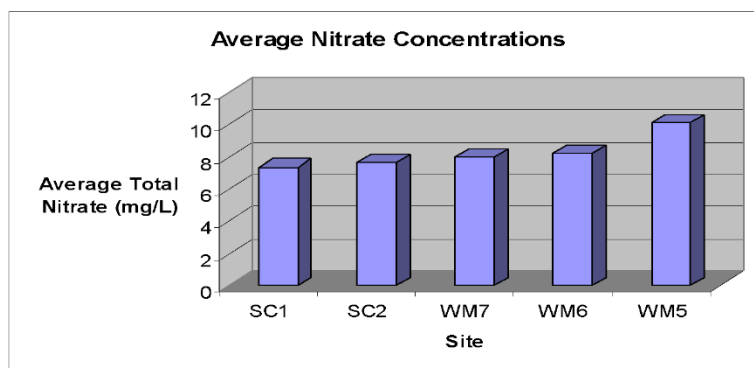


Figure 5. Total average nitrate concentrations (mg/L $\text{NO}_3^- - \text{N}$) over 1 year for Spruce Creek and Warriors Mark Run.

Water quality in Warriors Mark Run is poorer at sampling site WMR 5 compared to its upstream counterparts. Average taxa richness, EPT and PTI score all decline from site 7 to site 5 and from site 6 to site 5 (Table 2). Generally, the convergence of the 2 streams creates an increase in pollution going through

WMR 5. Total nitrate concentrations also tend to increase in this direction. This suggests that traditional agriculture is indeed affecting the water quality of Warriors Mark Run. These trends are not apparent in Spruce Creek.

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Spruce Creek shows little to no difference in water quality between SC 1 and SC 2 even though the stream flows through a CAFO from site to site. Discharge could potentially be a factor controlling dilution of nutrients, but insufficient discharge data for SC 2 will not allow for a detailed analysis.

CONCLUSIONS

The water quality of Spruce Creek and Warriors Mark Run has been previously characterized by stream chemistry data which, although providing important water-quality information, is highly variable through time. Macroinvertebrates can characterize the overall health of the watershed through time due to their stationary nature and ability to respond quickly to stress. Therefore, macroinvertebrates can provide a better overall assessment of stream quality. The macroinvertebrate populations in Spruce Creek indicate a higher water quality than that of Warriors Mark Run, which supports conclusions drawn from the water chemistry data. Used together, these methods provide a powerful tool for examining water-quality issues in this Central Pennsylvanian watershed.

ACKNOWLEDGEMENTS

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NEST-SITE PREFERENCE OF GRAY SQUIRRELS

Krystle Cox, Jeff Glaid, Piere Ondrasik and Sean Scheler

ABSTRACT

The gray squirrel (*Sciurus carolinensis*) inhabits most of the eastern coast, but is more concentrated in the northeastern states. It primarily nests in oak, hickory, and walnut trees. This study was conducted to see what type of habitat gray squirrels preferred. Three locations were examined: roadside, deep woods and streamside. A one square acre portion of land was determined using a measuring tape and tree marking ribbon. We counted nests within the acre and used a Merritt hypsometer to measure the height of the nests. We also used this tool and geometric triangulation to find the height of the trees, and used a measuring tape to find the circumference of trees in the area. It was hypothesized that the squirrels would prefer oak trees.

Key words: Oak, Merritt hypsometer, Sciurus carolinensis, tree height

INTRODUCTION

The gray squirrel *Sciurus carolinensis* is preyed upon by owls, foxes, bobcats and hawks, among other predators (Merritt, 1987). They live high up in deciduous trees where they seek cover; and they feed on nuts, buds, insects, bird eggs, baby birds, and occasionally tree sap (Linders, West, and Vander Haegen, 2004). According to Merritt (1987), their population density is 2-20/acre, their home range is about 2-7.4 acres, and they inhabit mixed coniferous deciduous forests. "Their abundance is dictated by seed crop productivity rather than specific plant communities," so they are obviously more likely to choose trees that yield the most seeds, or nuts (Merritt, 1987). Their populations tend to be especially large in oak forests.

Gray squirrels live year-round in leaf nests that they construct in the crotch of tree limbs. In the summer, nests are hard to see because they are made with green leaves amongst the green trees, but they are much easier to spot in the winter when the nest of dead leaves stands out amongst the stark bare branches. Usually nests are made of leaves and twigs; they are water-resistant; and the inside is lined with moss, grass, shredded bark and other vegetation

(Merritt, 1987). The nests have a single entrance that faces the tree trunk (Merritt, 1987). Gray squirrels live to be about five years old, and in that time they have about two litters of young per year (Moran, 2005). These crepuscular animals have very good balance and a multi-functional tail that acts as a sunshade, a rudder, a signal, and a blanket.

We tested whether gray squirrels prefer to nest in the woods, near a water source, or by a road. We hypothesized that squirrels would build more nests in the woods and close to water. In trees near the road, we hypothesized that they would build their nests higher up off of the ground. Finally, we assumed that the nests would be found in greater numbers in the thicker, taller, and sturdier trees. Our null hypothesis stated that the squirrels would not show a preference to a particular site or tree and would build at random heights. It is worthwhile to study these hypotheses because they may improve our understanding of the environmental and ecological impacts that human influences have on squirrels.

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METHODS AND MATERIALS

Between April 18-22, 2005 our group measured out a one-acre block of wooded area at each of three different sites located near or around Huntingdon, Pennsylvania. We chose diverse sites that we thought would best show the effects of the environment on squirrel nesting. The first area chosen (Site 1) was an isolated wooded area near “the cliffs” close to the Juniata College campus. This area was chosen due to its semi-remote nature and overall lack of human interference. The second sample site (Site 2) was a wooded road-side area near Juniata College’s peace chapel; it was chosen to test the effects that direct contact with human activity may have on squirrel nesting habits. Lastly, we chose an area near the Juniata River (Site 3) to see if close proximity to water affects the nesting behavior of squirrels.

In each area, within the measured one-acre lot, a count was made of all of the trees containing squirrel nests. For each nest, the circumference of the tree was measured with a flexible measuring tape at

the diameter at breast height (DBH). Tree diameter was calculated as

$$d = C/\pi,$$

where d equals diameter and C equals circumference. The heights of the nest (and tree) were measured using geometric triangulation and a Merritt hypsometer (see Fig.1; following LeBlanc, 2004).

According to the University of California, a Merritt hypsometer is a useful tool for giving tree height, which is calculated as a function of the percentage of the measurer’s distance from the tree. I.e., the reading on the hypsometer tells one how tall the tree is in relation to their distance from it. For example, if you are at a 100-foot distance, and the hypsometer yields a reading of 30, you can take 30 percent of 100 feet, which is 30 feet (.30 x 100 = 30). However, due to our study being set in densely wooded area, we worked from a distance of 50 feet. Even at this closer distance, the Merritt hypsometer will give accurate readings

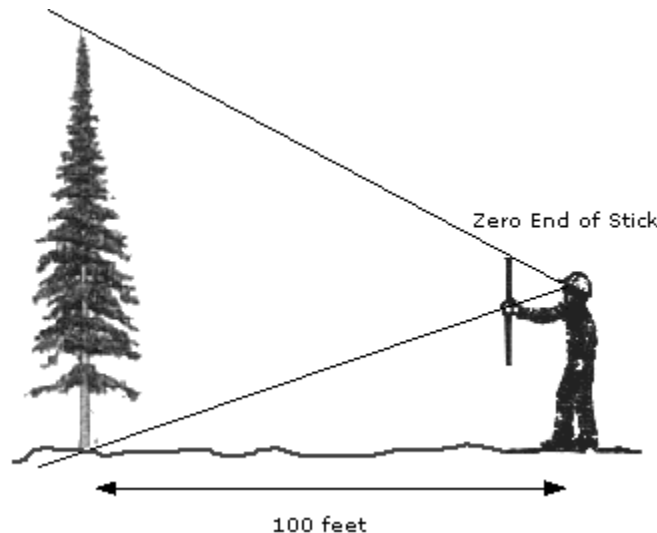


Figure 1. Use of Merritt Hypsometer (LeBlanc 2004)

At each site, tree type for each squirrel nest, tree types in general were recorded. We took measurements not only of trees with nests in them, but also of randomly selected trees (the number of which was equal to the number of nested trees in each respective area). We selected random trees by picking ones near the nests to perhaps find an association between squirrel nesting and tree type or size.

Lastly, within our one-acre plots, we marked off a 10ft. x 10ft area and counted all trees (above 10 inches in circumference only) in that block so as to obtain a rough estimate of the overall tree density at each site. This could be useful in determining whether forest density has an affect on the nesting habits of the grey squirrel.

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Minitab was used for performing chi-square tests, t-tests and regression analyses where appropriate.



Figure 2. Photographs depicting two squirrel nests (Little 1980)

RESULTS

Table 1 shows the measurements from site 1 of tree height, tree circumference, and squirrel-nest height. Table 2 shows the same measurements for site 2. The tables also show the types of trees in which the nests were found. As can be seen, there were more squirrel nests at site 1 than site 2. Tree height, tree circumference and squirrel-nest height were approximately the same for both sites; however, there were different types of trees present at each site. For

both sites combined, squirrel nests occurred more frequently at high heights than medium or low heights (Chi-square test: $\chi^2 = 9.0$ $P = 0.011$; Table 3). Squirrel-nest height is also marginally correlated with tree height ($r^2 = 0.34$, $P = 0.058$). The tables show that oak trees were the primary tree type at site 1; and maple trees were the prevalent tree type at site 2. However, a chi-square test showed no effect of tree type on nest occurrence ($P = 0.39$). Two-sample t-tests also showed no effect of tree type on nest height ($t = 0.16$, $P = 0.882$) or of tree circumference on nest height ($P = 0.39$).

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Table 1. Tree height, tree circumference, tree type, and squirrel-nest height in a one square acre plot of land at site 1

Tree Number	Nest Height (ft)	Tree Height (ft)	Tree Circumference (in)	Tree Type
1	40	40.31	28	Oak
2	38.75	39.06	26	Oak
3	38.75	54.38	36	Oak
4	38.44	60.63	47.5	Oak
5	55.63	56.88	36.25	Oak
6	50.63	59.38	82	Oak
7	61.88	67.5	31	Oak

Table 2. Tree height, tree circumference, tree type, and squirrel nest height in a one square acre plot of land at site 2

Tree Number	Nest Height (ft)	Tree Height (ft)	Tree Circumference (in)	Tree Type
1	46.25	57.81	39	Maple
2	47.5	55.31	53.5	Maple

Table 3. The observed and expected frequencies of squirrel-nest height. Tree height categories represent 20 ft. intervals

	Sections of trees			
	High	Medium	Low	Total
Observed	9	0	0	9
Expected	3	3	3	9

Tables 4 and 5 show tree height, tree circumference, and tree type of random trees from both site 1 and site 2. The same number of tree measurements was completed as done for trees with squirrel nests that were found in the one square acre piece of land at each site (seven trees for site 1 and

two trees for site 2). As can be seen, tree height and tree circumference are approximately the same for the random trees as the trees with squirrel nests ($t = 1.22$, $P = 0.241$ for tree height; $t = 1.65$, $P = 0.137$ for tree circumference).

Table 4. Tree height, tree circumference, and tree type of randomly selected trees in a one square acre plot of land at site 1

Tree Number	Tree Height (ft)	Tree Circumference (in)	Tree Type
1	55	40.5	Oak
2	40.94	30.5	Maple
3	49.38	34	Oak
4	43.13	26.5	Black cherry
5	49.38	29.5	Hickory
6	63.13	31.5	Oak
7	62.19	37	Oak

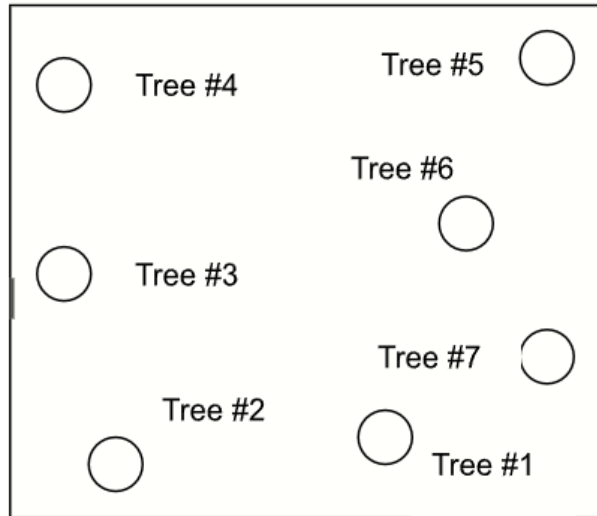
Table 5. Tree height, tree circumference, and tree type of randomly selected trees in a one square acre plot of land at site 2

Tree Number	Tree Height (ft)	Tree Circumference (in)	Tree Type
1	30	31	Maple
2	46.88	30	Maple

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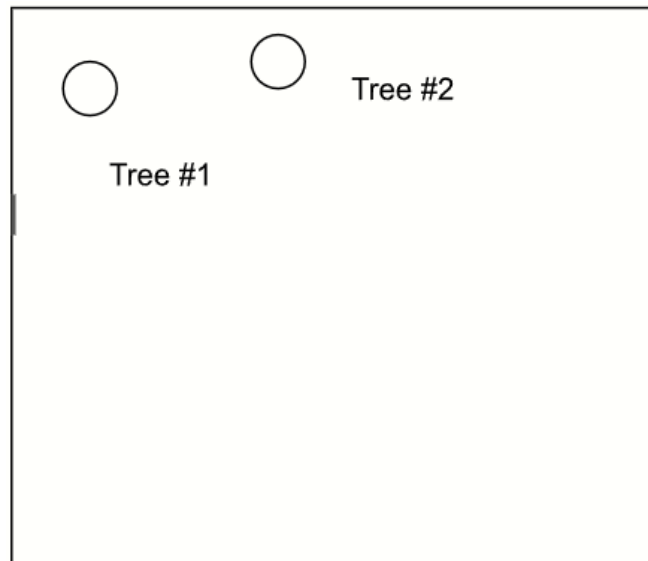
Figure 3 is a diagram showing the distribution of trees found with squirrel nests at site 1. As can be seen, the trees with the nests appear to be located along the edges of the one-acre plot of

land. This could be due to the fact that the trees in the center of the one-acre plot of land appeared to be less dense than the ones along the outer edges.



HOUSES

Figure 3. Distribution of trees with squirrel nests in the one square acre plot of land at site 1



ROAD

Figure 4. Distribution of trees with squirrel nests in the one square acre plot of land at site 2

Figure 4 is a diagram showing the distribution of trees with squirrel nests at site 2. The diagram illustrates that the trees with squirrel nests are located on the far edge of the one square acre plot of land, farthest from the road.

There were no squirrel nests found at site 3, which may be due to the fact that no oaks were seen at

this site. The only trees identified in the one-acre plot of land at site 3 were walnut, maple, and sycamore trees. A density measurement of only 17 trees per meter squared was measured. A density measurement of 26 trees per m^2 was calculated at site 1 and 21 trees per m^2 at site 2.

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DISCUSSION

Our results indicate that gray squirrels prefer to nest high in oak and maple trees. Their nest height was also positively correlated with tree height, at least marginally so. Tree height explains 34% of the variation in nest height. However, nest height was not related to tree type or circumference. Nor were trees with and without nests significantly different in height or circumference. This latter result was unexpected and may be due to the small sample sizes of this study.

Most squirrel nests were observed at site 1, which was isolated from excessive human disturbance and had a high density of trees (26 trees m⁻²). There were only two squirrel nests observed at site 2; however, the maple trees that housed the squirrel nests were taller and appeared to be more stable than the surrounding population of maple trees. This site was located along a roadway, which could have contributed to the lower squirrel population. There was a fairly high density of trees at site 2 (21 trees m⁻²); however, maple trees do not provide a sufficient source of food for the squirrels. Site 3 contained no squirrel nests among the one-square acre plot of land. At this site, the trees were much smaller and less dense (17 trees m⁻²), which could have contributed to the extremely low number of squirrel nests. The fact that the trees were small at this site may be due to flooding or pollution, leading to the underdevelopment and death of many trees. Also the railroad tracks and route 22 run parallel to the Juniata River and may contribute to pollution and disruption of the environment along the river, leading to a lower squirrel population. The trees along the river consisted of primarily sycamore trees, but a few walnut and maple trees were also found. These trees may not be stable enough to house a squirrel and its nest.

Further research is needed to determine if gray squirrels have a preference for where they build their nests. A larger plot of land (possibly 10 square acres) should be used to ensure a larger sample size of trees. Extraneous variables should also be controlled at each site. For example, future water sites relatively undisturbed by humans may be found on Juniata College property near Seven Points.

ACKNOWLEDGEMENTS

We thank Dr. Todd D. Gustafson for loaning us his tape measure, thus enabling us to measure tree circumferences and our one square acre of sampling area at each site. We also thank Dr. Glazier for being available to answer various questions we had about this research, and Eric Butler for his assistance with Minitab and our statistical testing.

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DIURNAL CHANGES IN THE ECOSYSTEM OF WARM SPRING

Selina Aurandt and Allyson Jackson

ABSTRACT

By following a previously established experimental design, data were collected to estimate the metabolism of a freshwater spring. Because plants give off oxygen only during daylight hours, while animals use oxygen continuously throughout the day, we hoped to be able to see changes in dissolved oxygen and pH over a day to night period. Data on dissolved oxygen, pH, solar radiation, water temperature and air temperature were collected over two 24-hour periods at Warm Spring in Huntingdon, Pennsylvania. We found marked fluctuations in these parameters during the diurnal period. These changes were used to estimate gross ecosystem respiration and primary productivity. Due to the unique nature of a spring, oxygen and pH showed more stability closer to the source and more variability further away.

Key words: Diurnal cycles, ecosystem metabolism, oxygen, pH, Warm Spring

INTRODUCTION

Springs occur where underground reservoirs of water seep to the surface, and are prevalent throughout central Pennsylvania. They are interesting ecosystems to study because of their unique trait of having approximately constant water temperature year round. This allows springs to support year-round vegetation and resist freezing in the winter. Our field site, Warm Spring, has been the focus of many investigations into its water chemistry and biotic diversity, but never a complete ecosystem metabolism study. By monitoring the changes that occur in the spring over a diurnal period, we hope to better understand if and how day and night cycles of plants and animals affect water chemistry.

Diurnal changes in water chemistry have been studied in many aquatic ecosystems. Odum (1956) first conceived of the experimental design that we used for this study. He later used this method in comparing 12 different Floridian ecosystems (Odum, 1957). Since that time, ecologists have used the diurnal curve method to better understand aquatic ecosystems. Wetzel and Likens (2000) refined Odum's strategy that became the blueprint for our study of Warm Spring.

We hypothesized that dissolved oxygen should increase during the day due to higher solar radiation, which causes an increase in plant photosynthesis. Photosynthesis involves the intake of carbon dioxide and release of oxygen by plants. While plants can only photosynthesize during the day, the animals in the spring are constantly absorbing oxygen through respiration. Therefore, as night progresses, oxygen content should continue to decrease until daybreak when plants begin photosynthesizing again.

We also hypothesized that pH should increase in the daylight hours due to the plants absorbing more carbon dioxide and decrease at night as animals continue to produce carbon dioxide. All of these parameters will be examined, as well as their relationship to water temperature, air temperature, and amount of solar radiation. Furthermore, from these data we will estimate the gross primary productivity and respiration of the spring ecosystem. By understanding the metabolism of Warm Spring we will better understand how the ecosystem produces and uses oxygen throughout the day.

FIELD SITE

Warm Spring, located on Cold Springs Road in Huntingdon, Pennsylvania, maintains a reasonably

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constant temperature year round. A man-made structure has been created to pool the water that comes from the ground, after which the water flows into a spring brook. The relatively unpolluted spring runs through a deciduous forest that, at the time of our study, did not yet have leaves. The spring, however, maintains vegetation in and around its water year round. The spring itself runs over a changing variety of sediments, beginning with sand and cobbles at the source, and progressing to sand, mud, and boulders further downstream. For our study, Site 1 was located at the point where the pool of water feeds into the spring. The water was shallow and littered with stones. Site 2 was located 250 feet downstream, following the curve of the spring. Its sediment consisted of sand, mud, and few boulders. This area had also collected much more detritus such as logs and dead leaves.

MATERIALS AND METHODS

To estimate the spring's metabolism, we used various chemical indicators to follow changes in pH and oxygen during a 24-hour period. Measurements were taken at two sites every three hours for two separate 24-hour periods on April 2 and 9, 2005. On both dates we measured dissolved oxygen content and water temperature using a YSI oxygen meter, and solar radiation using a Li-Cor solar radiation detector. On April 9, we refined our sampling methods to include pH using a Markson pH meter, air temperature on land using a basic thermometer, and maximum solar radiation at each site. On this date, measurements were taken in triplicate at each site in order to obtain a mean and standard deviation. Adjustments were also made between the two dates to account for Daylight Savings

Time, which began on April 3. Because of this, we started sampling at 1 A.M instead of 12 midnight, in order to have the measurements be comparable. Another difference in sampling methods between the two days was that on April 2 we walked through the spring to reach Site 2, whereas on April 9, we chose to not disturb the ecosystem and reached Site 2 by land.

RESULTS

We monitored the spring on both a cloudy and a sunny day. Fig. 1 and Fig. 2 respectively show the maximum amount of solar radiation on each day. The trend for both days shows the highest reading to occur in early afternoon, but April 9 shows a much higher reading. There is also a difference shown between the two sites on the April 9 due to the amount of tree cover blocking sunlight.

Results for changes in water temperature are found in Fig. 3 and Fig 4. Both figures show a greater change throughout the day at Site 2 than at Site 1. Site 2 tends to be consistently cooler than Site 1. On April 9, we also measured land air temperature, shown in Fig. 5. The peak air temperature occurs at the same time (1:00 P.M.) as peak water temperature on April 9.

On April 9, we also took readings of pH, which are shown in Fig. 6. Site 2 has an overall higher pH than Site 1 with a peak for both sites at 1:00 P.M.

Fig. 7 and Fig. 8 show the changes in dissolved oxygen content in the spring on each day. There is a slight increase in dissolved oxygen at 12:00 PM on April 2. However, on April 9 there was an earlier increase in dissolved oxygen at Site 2, peaking at 10:00 AM, while Site 1 shows no trend.

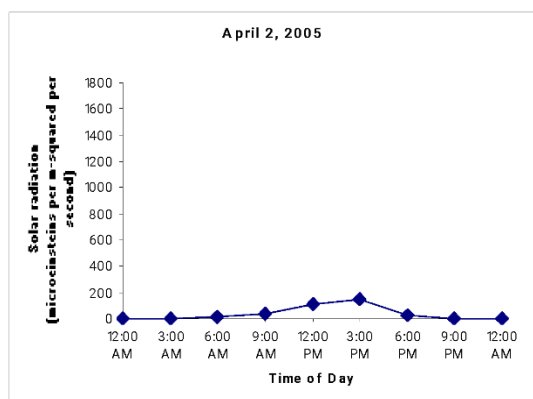


Figure 1. Change in solar radiation throughout a cloudy 24-hour period at Site 1 only. Maximum is shown at 3:00 P.M.

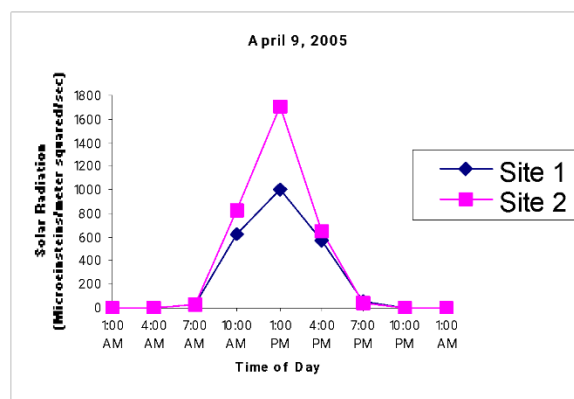


Figure 2. Change in solar radiation at both sites on a sunny day. Maximum is shown at 1:00 P.M. for both sites.

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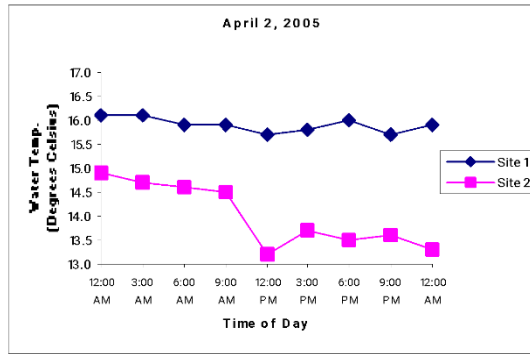


Figure 3. Change in water temperature over a 24-hour period. Trend shows overall decrease in temperature at Site 2 while Site 1 remains constant.

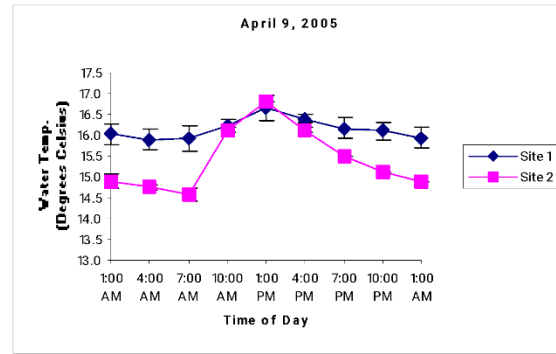


Figure 4. Change in water temperature over a 24-hour period. Both sites peak at 1:00 P.M. Standard deviation for three samples is shown by black bars.

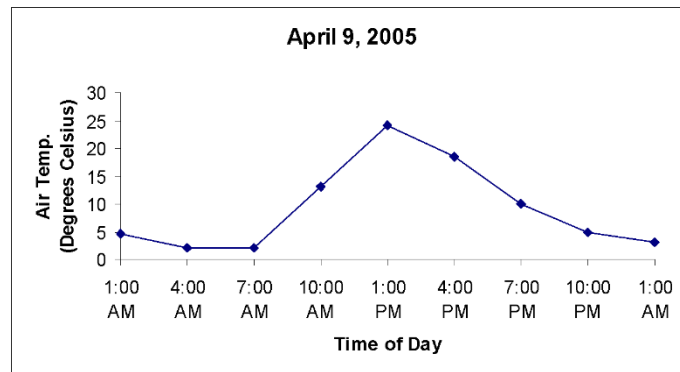


Figure 5. Land air temperature on April 9, 2005. Peak occurs at 1:00 P.M.

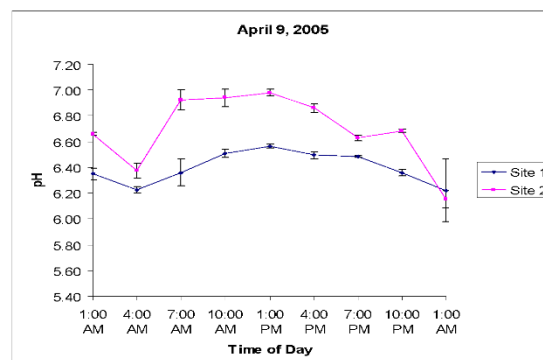


Figure 6. pH change at both sites on April 9, 2005. Standard deviation for three samples is shown by black bars.

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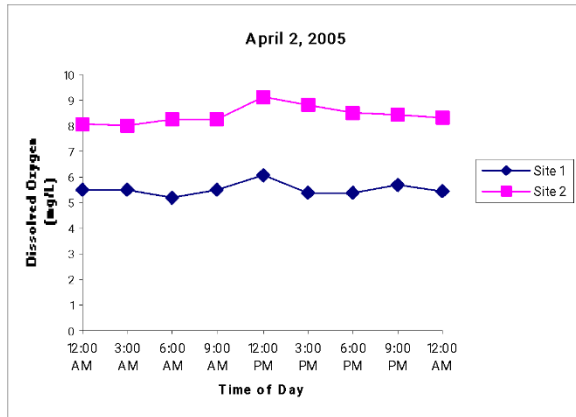


Figure 7. Changes in dissolved oxygen at both sites on April 2, 2005.

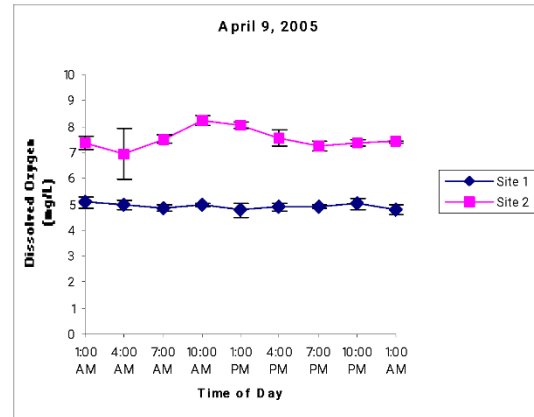


Figure 8. Changes in dissolved oxygen at both sites on April 9, 2005. Standard deviation for

We calculated percent saturation using known 100% saturation values for water at each temperature and compared them to the dissolved oxygen content (DO), using the formula:

$$(\text{DO}_{\text{sample}} / \text{DO}_{\text{theoretical at 100\% saturation}}) \times 100.$$

Standards for 100% saturation were taken from Wetzel and Likens (1979). Percent saturation of oxygen is shown in Fig. 9 and Fig. 10.

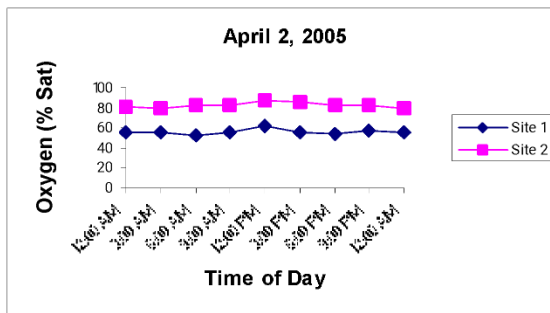


Figure 9. Percent saturation of dissolved oxygen at both sites on April 2, 2005.

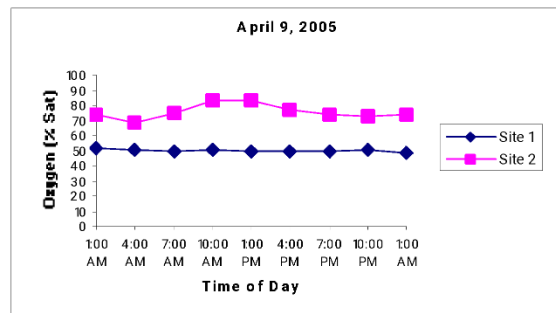


Figure 10. Percent saturation of dissolved oxygen at both sites on April 9, 2005.

To determine the metabolism of the spring, we also used our dissolved oxygen values to find the oxygen rate of change throughout the day (Figs. 11 and 12). The data for April 2 shows more variation, as

both Site 1 and Site 2 have unexpected fluctuations. Fig. 12 shows a more standard curve for the data on April 9.

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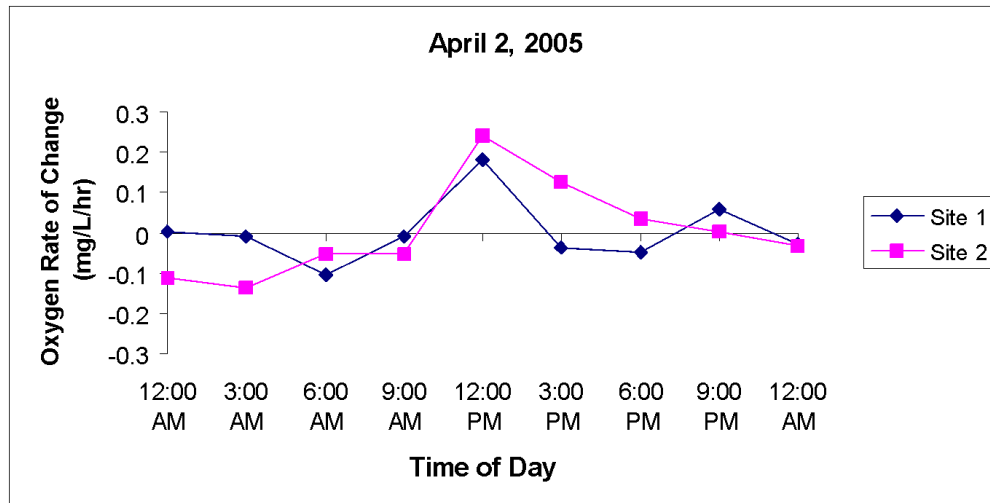


Figure 11. Oxygen rate of change at both sites on April 2, 2005. Sunrise occurred at 5:54 A.M. and sunset at 6:38 P.M. (U.S. Naval Observatory 2005).

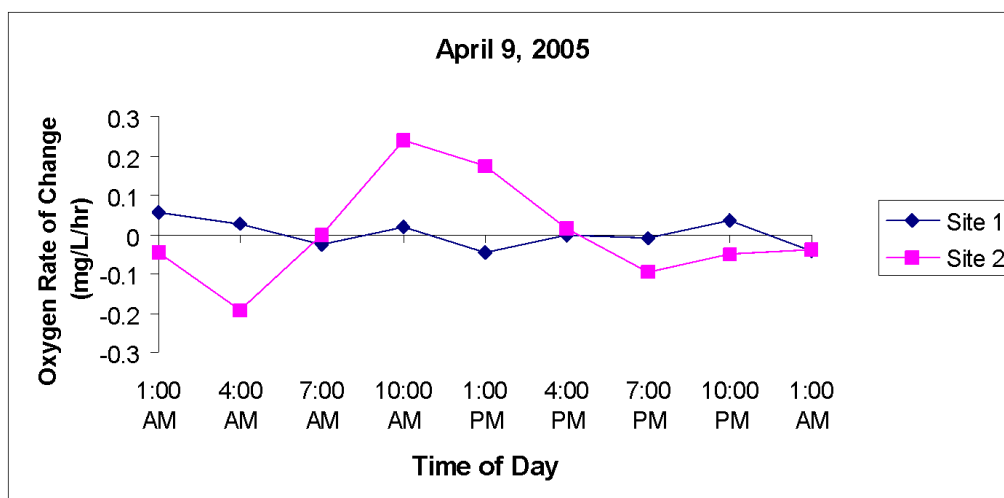


Figure 12. Oxygen rate of change at both sites on April 9, 2005. Sunrise occurred at 6:43 A.M. and sunset at 7:45 P.M. (U.S. Naval Observatory 2005).

Fig. 13 and 14 show a schematic representation of the metabolism of Warm Spring. From the data collected at Site 2 on each day, we were able to approximately estimate the gross respiration and productivity of the spring. On April 2, gross nighttime respiration equals 0.975 g oxygen/mL,

gross daytime respiration equals 1.35 g/mL, and gross primary productivity equals 2.55 g/mL. For April 9, gross nighttime respiration equals 1.2375 g/mL, gross daytime respiration equals 1.7625 g/mL and gross primary productivity equals 3.0375 g/mL.

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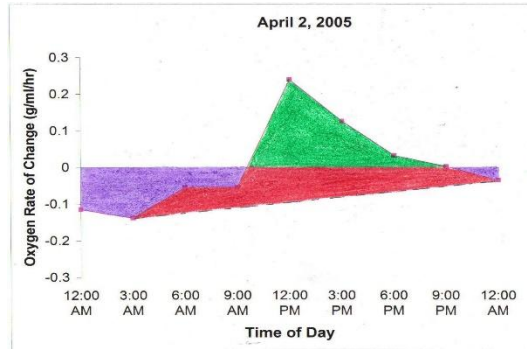


Figure 13. Ecosystem metabolism for Site 2 on April 2, 2005. Areas in purple represent an estimate of gross, nighttime ecosystem respiration. Areas in red are an estimate of gross daytime ecosystem respiration. Combined green and red area is an estimate of gross primary productivity.

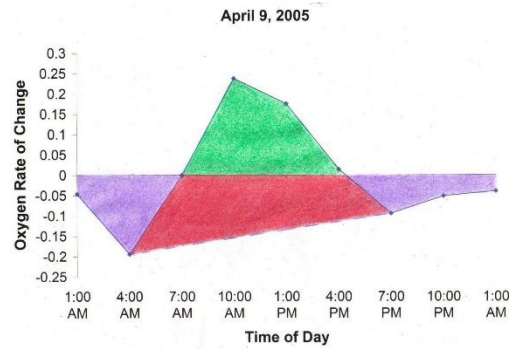


Figure 14. Ecosystem Metabolism for Site 2 on April 9, 2005. Areas in purple represent an estimate of gross, nighttime ecosystem respiration. Areas in red are an estimate of gross daytime ecosystem respiration. Combined green and red area is an estimate of gross primary productivity.

DISCUSSION

Several interesting trends were seen at the different sites on different days. Solar radiation was lower on April 2 when there was about 100% cloud cover than on April 9 when cloud cover was only about 5%. On April 2, the maximum solar radiation occurred at 3:00 P.M., which we believe was due to a break in the clouds and rain. However, this peak was less than that observed at 1:00 P.M. on April 9. This is most likely the normal time when the spring receives the most sunlight in April. On April 9, we also saw a difference between solar radiation at Site 1 and Site 2. The higher curve for Site 2 is explained by the fact that there was less tree cover at this site. If it is the sunlight that is driving this ecosystem, we should have been able to see differences between the two days in levels of air temperature, water temperature, dissolved oxygen and pH.

As shown in Figs. 3 and 4, diurnal water temperature varied differently between the two days. These changes, though significant to our study, are not as great as you would see in a stream because of the fact that a spring regulates its water temperature. April 9 showed a more cyclic trend in water temperature, most likely because of the warmer air temperature and increased sunlight during the middle of the day. Also, on both days Site 2 showed more temperature fluctuation, whereas Site 1 stayed more constant. This is due to the fact that Site 1 is closer to the source where water comes from underground where water temperature is constant. Site 2 is more subject to environmental conditions affecting its temperature, such as sunlight or air temperature. On April 9 there also seems to be a correlation between

the recorded air temperature and water temperature. As shown in Figs. 4 and 5, air and water temperature peak at the same time as solar radiation (1:00 P.M.). This shows that both air temperature and solar radiation play a role in determining water temperature.

One drawback of our data collection is our failure to obtain accurate pH readings on April 2. Because we cannot compare these data to those for April 9, we cannot make any generalizations regarding the effect of sunlight on pH. However, we can compare Site 1 and Site 2 on April 9. Fig. 6 shows that Site 2 has a pH consistently closer to neutral (pH = 7) than Site 1. Because pH levels are affected by carbon dioxide levels in the water, decreases in pH at night may be the result of plants taking in less carbon dioxide. During the day, the plants correct this imbalance and bring the pH back to neutral. We assume it is the plants in the ecosystem that cause this change because the water flowing to Site 2 passes over much more plant life and therefore Site 2 shows higher pH levels.

A major objective of this study was to see whether dissolved oxygen content was affected by the amount of photosynthesis taking place in the spring. Because we assume more photosynthesis occurs on a sunny day, we hypothesized that any changes in dissolved oxygen between the two days would be because of increased photosynthesis. In fact, we found that dissolved oxygen did increase during daylight hours when plants are more photosynthetically active (Figs. 7 and 8). Diurnal changes in oxygen are expected to be lower in relatively stable springs as compared to streams. A diurnal curve for a stream can often have dissolved

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oxygen fluctuating between 6 to 12 mg/L (McDiffett, Carr and Young, 1971), whereas in Warm Spring oxygen fluctuated only between 6.93 to 9.14 mg/L. The diurnal curve in Warm Spring was similar on sunny and cloudy days (see Figs. 7 and 8). Even correcting for water temperature by calculating percent saturation does not reveal any substantial difference between the two days (see Figs. 9 and 10). However, the rate of change in dissolved oxygen did vary slightly between the two days (see Figs. 11 and 12). April 9 shows a broader peak for change in oxygen, probably due to the increased sunlight absorption on this day. As for water temperature, Site 2 showed much more cyclic variability in dissolved oxygen than Site 1. This is also due to the fact that the water from Site 1 is coming directly from underground, not giving it time to fluctuate with in response to downstream environmental variables. Site 2 shows the expected curve for diurnal changes in dissolved oxygen in a stream ecosystem as documented in Wetzel and Likens (2000). This is most likely due to the fact that Site 2 has more biotic life than Site 1, leading to more changes. Figs. 11 and 12 show that dissolved oxygen is produced during the day and used up during the night. However, when the data is converted into a graph showing areas of primary productivity and ecosystem respiration (Fig. 13 and 14), one can see that April 9 shows more overall activity than April 2. A rough estimate of area in these graphs shows that April 9 had gross primary productivity totaling 3.0375 mg/L, whereas April 2 only had 2.55 mg/L. We infer that this difference is due to increased solar radiation on April 9.

It is important to note that our study does not include a calculation of the re-aeration flux of oxygen, which is the amount of oxygen lost to the air based solely on air-water diffusion (Wetzel and Likens, 2000). This value is used to correct a double curve method where the researchers would use the change between two sites to calculate change in dissolved oxygen. The data from a double curve method is then combined into one graph. Although we collected data from two sites, we only used a single-curve method for both sites. Instead of integrating the data into one curve, each site was graphed separately (Figs. 11 and 12). We recommend using the single-curve method for similar studies in other springs.

Further research regarding the metabolism of Warm Spring should be conducted during different times of year. Our study was done in the early spring, before trees have leaves and most plants start growing. It would be interesting to see the effect of tree cover, increased vegetation, and warmer air temperature upon the spring's metabolism. We suggest performing this study during different seasons to see how seasonal

changes in various environmental variables may affect the spring. Further research should be sure to include accurate triplicate measurements for each variable and site. Another field site that may yield interesting comparisons would be a spring with less vegetation due to acid conditions, such as Dubbel Spring in Huntingdon, PA. If our interpretations are accurate, we would expect Dubbel Spring to show little or no diurnal changes. Another interesting way to look at this issue is to compare a spring with a stream.

ACKNOWLEDGMENTS

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ARE ALGAE INDICATORS OF NITROGEN LEVELS IN SPRINGS?

Lauren Forster, Heather Hassel and Zach Laubach

ABSTRACT

Algae are commonly used as an indicator of nitrogen overload in aquatic systems. Variation in nitrogen levels is a result of a water system's relative location to input sources and/or buffer areas. By examining these differences among various springs, the causes for such nitrogen variations can be better understood. In this study, there was significant variation in nitrate levels between the springs of Huntingdon County and Centre County. However, the results of this study do not indicate that a correlation exists between these nitrate levels and the percent algal cover or algal species composition of the respective springs. Although this study was inconclusive, it has provided useful information for future researchers that indicates several other environmental factors in addition to nitrate concentrations that may play a role in controlling algal species composition and population growth.

Key words: Algae, Centre County, Huntingdon County, nitrates, springs

INTRODUCTION

Algae are commonly used as bioindicators for several types of pollution, including nitrate overload (Graham and Wilcox, 2000). There is extensive data that indicates that nitrogen availability is one of the two major factors limiting algal growth in freshwater systems (Biggs, 1989; Krebs, 2001; Cascallar *et al.*, 2003). As a result, the amount (biomass) of algae in a system can be indicative of the concentration of nitrates present in the water column (Round, 1965). The species composition of an algal community may reflect water quality, as well (Biggs, 1989; Burford and Pearson, 1998; Wunsam *et al.*, 2002).

Nitrate overload is one of the most common pollution problems in freshwater systems because of the use of nitrates in agricultural fertilizers (Miller and Miller, 1992). Nitrates are highly soluble in water, and thus are readily leached from soils and deposited into freshwater systems (Power *et al.*, 2000). Most species of algae require nitrogen in the form of ammonium (NH_4^+) to manufacture essential amino acids (Graham and Wilson, 2000). Ammonium is often in limited supply in aquatic systems (Webster *et al.*, 2003).

However, many species of algae are also capable of processing nitrates (NO_3^-) introduced through anthropogenic processes, converting them to ammonium via the nitrate reductase enzyme (Graham and Wilcox, 2000).

The process of eutrophication resulting from nutrient overload is known to change the species composition of algal communities in freshwater habitats, including springs. Typically, healthy springs contain a variety of diatoms and green algae; however in situations of pollution blue-green algal species dominate (Krebs, 2001). Phytoplanktonic and macrophytic algal species are extremely rare in waters overloaded with nitrates, because the cyanobacteria (blue-green algae) that dominate these systems are more efficient users of nutrients and quickly grow in blooms that block sunlight from reaching other algal taxa (Ferber *et al.*, 2004). For water-quality assessment, researchers have compiled lists that indicate "clean water" versus "polluted water" algal species (Palmer, 1962).

Using data collected from five Central Pennsylvanian springs, our group tested the hypothesis that percent algal cover and the presence of "polluted water species" will correlate with nitrate concentration.

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FIELD SITES

We conducted this study at five different springs in central Pennsylvania. Three of the springs were located adjacent to Route 45 in Centre County, near the town of Spring Mills. The other two springs were located just off of Route 26 in Huntingdon County, north of the borough of Huntingdon.

METHODS AND MATERIALS

At each of the five spring sites, data collection was completed at both an upper location (which corresponded with the spring source) and a lower region (which corresponded to an area approximately 20 meters down flow of the source). Consistent tests, methodology, and sampling techniques were used at all sample sites. We estimated pH with a Markson pH meter. We estimated water temperature ($^{\circ}$ C) and conductivity (μ S) with a YSI conductivity meter. Originally we measured levels of dissolved oxygen in the springs, but because of equipment failure these data set were cut short before completion.

We also estimated free nitrate and nitrite levels in each spring. To do this we collected two water samples from both the upper and lower sites at each spring. When collecting these samples the collection bottles were completely submersed during filling and sealed off while still submersed. This specific procedure was to prevent accidentally sampling part of the surface film as opposed to the actual water column. Next we mixed individually packaged NitraVer 5 as a reagent in each sample. Once the NitraVer 5 was added, each solution was mixed by gently shaking for one minute and then set aside for five minutes. After the conclusion of the 5-minute time

interval, solutions were placed in a spectrophotometer. The spectrophotometer was zeroed using distilled water containing NitraVer 5. Then each of the five spring samples was tested, and a value of free nitrate concentration was given as mg/L.

By visual inspection we estimated percent vegetative cover in each spring. We also sampled all the various types of vegetation that were growing in the spring itself, except for bank vegetation. Species of algae and moss were identified in the laboratory.

Using the five measurements of free nitrates taken at each spring, statistical tests were conducted using Minitab 14. The nitrate measurements were tested for equal variances and normality to test assumptions for a one-way analysis of variance (ANOVA). In the case of a violation of the ANOVA assumptions, the data were transformed using square root and log transformations before the non-parametric Kruskal-Wallis test was employed (Carver, 2004). We used Kruskal-Wallis and Mood Median tests to determine which of the springs had significantly different nitrate levels.

RESULTS

Pairs of conductivity, pH, and temperature measurements were made at each spring (Table 1). Although insufficient sample sizes prevented statistically analysis, these variables showed some trends. The three springs in Centre County had noticeably higher conductivities than the two springs in Huntingdon County. Also, the pH in Cold Spring was markedly lower than the pH values gathered at the other springs, whereas the water temperature of Warm Spring was markedly higher than that of the other springs.

Table 1. Water chemistry data for each of the springs sampled.

Spring	County	pH	Cond. (μ S)	Temp ($^{\circ}$ C)	Nitrates (mg/L)
Rising/Spring Mills	Centre	6.52	503.0	8.9	2.9
		6.63	505.0	8.9	2.5
					2.9
					3.0
Behind Trailer	Centre	6.84	626.0	9.7	3.8
		6.80	627.0	9.7	3.5
					2.9
					3.9
Farm	Centre	6.88	535.0	10.7	1.7
		6.61	526.0	10.6	1.8
					1.8
					1.8
Cold	Huntingdon	4.46	37.0	10.3	0.2
		4.40	36.8	10.9	0.2
					0.1
					0.2
Warm	Huntingdon	6.34	180.0	16.2	0.0
		6.04	164.0	16.6	0.0
					0.1
					0.1

The normality test indicated that the nitrate data were not normal, and the variances were found to be unequal. The nitrate data therefore violated the assumptions of the one-way ANOVA. After using two different transformations, the data still violated the assumptions, so ANOVA could not be used to analyze the data. The non-parametric Kruskal-Wallis test was used instead and indicated that there are statistically significant differences in the spring nitrate values ($H = 17.67$, $df = 4$, $P = 0.001$). The Mood Median test indicated that Trailer Spring (median = 3.65) and Rising Spring (median = 2.90) were not significantly different from each other, but they had significantly higher nitrate levels compared to all the other springs. Farm Spring (median = 1.80) had the

next highest nitrate levels of the five springs. The nitrate levels for Farm Spring were significantly lower than Trailer and Rising Springs; however Farm Spring had significantly higher levels compared to Warm and Cold springs. The nitrate levels for Warm (median = 0.05) and Cold (median = 0.20) springs were not significantly different from each other, but they had the lowest nitrate levels of all the springs.

Overall, five genera of algae and two families of moss were found among the five springs (Table 2). There was no noticeable trend between the nitrate levels and the aquatic vegetation in each of the springs. The vegetative coverage was greatest in Warm Spring and least in Cold Spring (Table 2).

Table 2. Algae and moss data for each of the springs sampled.

Spring	Aquatic Vegetation Cover (% est.)	Algae Genus	Moss Family
Rising/Spring Mills	1.0	Rhizoclonium	
Behind Trailer	40.0	Microspora	Fissidentaceae
			Fontinalaceae
Farm	15.0	Microspora	Fontinalaceae
Cold	0.1	Diatoma	
		Mougeotia	
Warm	98.0	Microspora	Fissidentaceae
		Batrachospermum	Fontinalaceae

DISCUSSION

The hypothesis that percent algal cover and species richness of polluted water algae would correlate with nitrate concentration was not supported. Although the three springs sampled in Centre County did have significantly higher nitrate concentrations than the two springs sampled in Huntingdon County, there was no corresponding trend in percent vegetative cover between the respective areas. The lack of a significant correlation between nitrate load and algal growth was likely the result of other environmental factors specific to the individual springs.

Our hypothesis was based on the false assumption that nitrates were the only factor affecting algal growth. Past research has indicated that this is not always true. Webster et al. (2003) presented data indicating that algal biomass is positively correlated with temperature and sunlight availability. Our experimental design did not include estimations of direct sunlight, so we are unable to analyze this possible factor. However, temperature measurements were taken at all sites. The spring with the highest temperature, Warm Spring, also had the greatest percentage of vegetative cover. This may indicate that temperature serves as a limiting factor for algal growth at these springs. Similarly, past studies indicate that water chemistry, specifically conductivity and pH, can affect the amount of algal cover in a water source (Graham and Wilcox, 2001). Our data do not show any definitive trends in conductivity and pH in relation to algal cover. However, it is possible and likely that several of these factors may be working in unison to control algal growth in spring systems.

Our hypothesis that the species richness of polluted-water algae would correlate with nitrate concentrations was not supported, because no

polluted-water species were sampled at any of the five springs. Typically, blue-green algae dominate polluted systems (Krebs, 2001). In this study, blue-green algae were not found at any of the five springs sampled. The species identified in this study were members of the green algae and diatom taxa, which are considered clean-water species (Krebs, 2001). This result is logical considering that all of the nitrate levels tested in this study were consistently lower than in waterways that are considered highly eutrophic or polluted (Osmond, 1995).

Despite the low level of pollution in these springs, it is possible to explain the variation in nitrate levels seen between Huntingdon and Centre Counties. The springs sampled in Centre County had significantly higher levels of nitrates, as a result of their proximity to past agricultural and industrial (corn processing plant) facilities. Some of the springs were adjacent to farm fields and roads, which can result in direct runoff and infiltration of various forms of nutrients (from fertilizers), including nitrates (Miller and Miller, 1992). On the other hand, the springs in Huntingdon County, which have significantly lower nitrate concentrations, are located in woodlands, a habitat that can act as a buffer for the removal of nutrients and pollutants.

Several steps could be taken to improve future studies concerning this topic. Flow rate measurements could help future researchers understand both the movement of nitrogen through the system and what types of vegetation are likely to be found. Also because algae and other aquatic vegetation are photosynthetic organisms, a measure of available sunlight would be a useful indicator of their potential for growth. Similarly, it would be useful to test phosphorous concentrations in the springs because it is the other significant limiting nutrient for plant growth in aquatic systems. To achieve more statistical significance, a greater quantity of samples for

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estimating pH, conductivity, and temperature need to be taken. Also, for completeness this study should also be continued for an entire year, which would allow for seasonal trends to be observed in the springs. This sampling strategy may indicate the effects of temperature fluctuation, light availability, and nutrient infiltration corresponding to fertilizer application and other seasonal inputs.

While, our hypothesis was not supported by the data obtained at the five springs sampled, this study has provided insight for future investigators. The lack of correlations between nitrates and algal growth indicate that several other environmental factors may also affect the growth and species composition of algal communities. These other factors include pH, conductivity, temperature, sunlight, and flow rate.

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ANALYSIS OF MACROINVERTEBRATE COMMUNITIES IN STREAMS OF VARYING WATER QUALITY USING BIOTIC INDICES

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ABSTRACT

Biotic indices are often effective indicators of pollution levels. One can evaluate water quality of streams by sampling macroinvertebrate communities, and then characterizing them with indices such as species richness, species diversity, and the EPT Index. We hypothesized that a stream suffering from acid mine drainage should have lower biotic index scores than a higher quality stream.

Key words: EPT biotic index, Hilsenhoff biotic index, macroinvertebrate, pH, water quality

INTRODUCTION

The purpose of this study was to compare the macroinvertebrate communities of two very similar watersheds with differing water chemistry and quality. Both Shoup's Run and Great Trough Creek are located in the Entriken quadrangle in South Central Pennsylvania. Both creeks are also tributaries to Raystown Lake and have comparable drainage areas. In addition, when analyzed using Geographic Information Systems (GIS) software, both watersheds exhibit similar land use and land-cover features. This includes vegetation types and amounts, human interference, paved surface area, and other physical characteristics. The major difference is that Shoup's Run encompasses a section of the Broad Top region.

This region is known historically for coal mining and includes the Joller Mine, which is now closed. In the mid 1990's the Joller site was found to exhibit acid mine drainage (AMD). In 1998 a water treatment system was installed to prevent and monitor the AMD occurring there. AMD occurs when water enters abandoned coal mines and reacts with iron pyrite in the coal to form very acidic water that can dissolve heavy metals in solution. This system is composed of seven different collecting ponds. Each pond has noticeably different biotic and abiotic features related to clarity, clay, reeds, cattails, rock,

and algae. Water collects in the top pond and slowly makes its way downhill through all seven ponds. Ideally, the water will be clean when it leaves the last pond and flows down the mountain side into Roaring Run and other local streams. This system was established after the Department of Environmental Protection discovered a pair of highly acidic and metal-rich deep mine discharges flowing into Roaring Run (DEP, 2002). Since then various problems have occurred with the system. In 1999 and 2000, the DEP and multiple private contractors went to Joller to pinpoint and fix the problems occurring there (EBTS, 2000). After multiple tries, the system was fixed, which according to the DEP, is currently working very effectively (DEP, 2002).

Since some macroinvertebrates show significant tolerance to particular pollution levels and because they can be easily analyzed both qualitatively and quantitatively, they were the subject of this study. Healthy streams can have several hundred different kinds of benthic macroinvertebrates with total numbers ranging in the thousands per m². Different species of macroinvertebrates have been characterized by the US Environmental Protection Agency (EPA) as sensitive, moderately tolerant, or pollution tolerant species. This characterization was the basis for the development of a variety of biotic

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indices, including the Hilsenhoff index and EPT index used to analyze one aspect of watershed health.

The Hilsenhoff index, or Familial Biotic Index, is a measure of the macroinvertebrate assemblage tolerance toward organic (nutrient) enrichment (PSU, 2003). In many ways this index is both an indicator taxa metric and functional group metric, since those taxa which become more dominant in moderately enriched streams are those which are taking advantage of shifts in the available food base in the stream. Each taxon is assigned a tolerance value ranging from 0-10 (0 being least tolerant) which is designated (x_i). These values are then multiplied by the number of individual organisms collected for that particular species (n_i), and the sum is divided by the number of total individual organisms collected for that particular site (PSU, 2003). The Hilsenhoff Equation is as follows:

$$\text{Familial Biotic Index (FBI)} = \left[\sum (x_i \cdot n_i) \right] / n$$

The EPT Index focuses on three orders of aquatic insects that are easily sorted and identified [i.e., Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies)], and is commonly used as an indicator of water quality. The EPT Index is based on the premise that high-quality streams usually have the greatest species richness of these orders. The greater the pollution, the lower the species richness expected, as only a few species are pollutant tolerant. It sums the total number of individuals found from each order. The resulting product is then analyzed using the EPT scale (See Figs. 6 and 7) (NRCS, 2001).

We hypothesized that Shoup's Run, which receives acid mine drainage, should have lower biotic index scores than the higher quality Trough Creek.

FIELD SITES

Shoup's Run (Huntingdon and Bedford Counties, Pennsylvania) and Trough Creek (Huntingdon County, Pennsylvania) were chosen

because of their similar land use and land cover, as well as relatively comparable flows and stream widths. Other physical features of both streams varied at specific locations. For this reason comparable forested sites (sites 1-3) and open sites (sites 4 and 5) were sampled in both stream systems. Sampling sites 1-3 along Shoup's Run were taken in Saxton, Pennsylvania, whereas sites 4 and 5 were taken on the outskirts of the town of Saxton. All sampling sites along Trough Creek were taken in Trough Creek State Park.

METHODS

At Shoup's Run, sample sites 1-3 were taken approximately 15-25 ft. apart and were sampled upstream from sample sites 4 and 5, which were taken approximately 15 ft. apart. Sites 1-3 and 4 and 5 were an approximate distance of 5 miles apart. Sites 1-3 contained 50% boulders and 50% cobbles, while sites 4 and 5 contained 15% boulders, 20% gravel, 5% sand, and 60% cobble. Shoup's Run drops significantly in elevation, and the water is quite turbulent, likely contributing to the healthy level of dissolved oxygen in the water. All five of the Shoup's Run monitoring sites were located in a wooded area.

At Trough Creek, sample sites 1-3 were taken roughly 15 ft apart and were far upstream from sample sites 4 and 5. Sites 1-3 contained 60% boulders, 20% cobbles, 5% gravel and 5% sand. All five sites were located in wooded areas.

Sampling at each site consisted of quantitative macroinvertebrate counts, physical measurements, and water chemistry readings. Sampling was done using a large 3-ft wide kick net. Macroinvertebrates were collected using a two person system in which one individual would hold the net while the other would shuffle their feet upstream of the net for approximately 30-60 seconds. Macroinvertebrates were collected in the net and the net was then transferred to the bank for field identification, as seen in Figure 1.



Figure 1. Sampling at Trough Creek.

Findings were recorded and tallied in the field. Physical measurements included determining the rate of stream flow using a flow meter, and width and average depth were measured using a two-person method utilizing a measuring tape, as seen in Figure 2.



Figure 2. Obtaining physical measurements at Shoup's Run.

The dissolved oxygen, pH, stream flow, temperature and conductivity of each site were measured using appropriate equipment in order to get an accurate water chemistry baseline. Other stream characteristics were also noted, such as stream bed makeup, width and depth, and descriptions of the surrounding environment.

Hilsenhoff and EPT biotic indices were used to determine the relative quality of each stream. These indices were used in the standard accepted method to provide a total value that was assigned to each site based on the weighting of each macroinvertebrate species.

RESULTS

At Shoup's Run, sample sites 1-3 had conductivity values of approximately 300 μS and pH values ranging from 3-4, which are associated with the acid mine drainage within the area. Downstream sample sites 4 and 5 had lower conductivity and higher pH. Macroinvertebrate diversity is relatively low in Shoup's Run. Caddisflies represented the most abundant taxon (Table 2).

Table 1. Physical and chemical characteristics of 5 sites along Shoup's Run.

Sample Site	Temp. (°C)	Depth (ft.)	Width (m)	Conductivity (μS)	pH	DO mg/L	Flow rate (mph)
1	10.1	1.5	6.7	298.5	3.26	11.04	2.4
2	9.6	1.8	6.7	299.8	3.9	11.02	3.56
3	9.9	1.65	6.3	299.7	3.89	11.04	2.87
4	10.2	1.2	13.8	194	4.94	10.89	3.5
5	10.2	0.7	13.7	192	4.7	10.80	1.74

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Table 2. Number of macroinvertebrates collected at 5 sites along Shoup's Run.

	Site 1	Site 2	Site 3	Site 4	Site 5
Caddisfly	26	13	28	5	4
Annelid	4	1	2	2	1
Stonefly	1	1	1	0	0
Water Beetle	2	0	1	0	1
Alderfly	0	0	1	0	0
Total	33	15	33	7	6

The pH in Great Trough Creek (6-7.5) was higher than that seen in Shoup's Run. Great Trough Creek also had a greater diversity of

macroinvertebrates than that of Shoup's Run. Caddisflies and mayflies were the most abundant taxa in Trough Creek.

Table 3. Physical and chemical characteristics of 5 sites along Trough Creek.

Sample Site	Temp. (°C)	Depth (ft.)	Width (m)	Conductivity (uS)	pH	DO mg/L	Flow rate (mph)
1	7.9	1.5	18.8	70.6	6.7	11.05	1.18
2	8.1	1.5	18.8	71.2	6.79	11.45	3.24
3	8.3	1.3	18.8	70.3	6.69	11.46	3.45
4	9.1	1.3	15	69.1	6.7	13.3	1.1
5	9.3	1.4	13	70.3	7.24	10.9	1.15

Table 4. Number of macroinvertebrates collected at 5 sites along Trough Creek.

	Site 1	Site 2	Site 3	Site 4	Site 5
Caddisfly	3	15	2	14	4
Annelid	2	0	7	3	0
Stonefly	3	8	3	4	3
Water Beetle	2	0	4	4	0
Dragonfly Larvae	0	0	4	1	1
Mayfly	6	25	0	10	12
Hellgrammite	1	0	0	1	0
True Fly	1	0	0	0	0
Water Penny	1	0	5	0	0
Maggot Larvae	0	1	0	0	0
Damselfly	0	1	0	0	0
Crayfish	0	0	2	0	0
Amphipod	0	0	1	0	0
Riffle Beetle	0	0	3	0	0
Total	19	50	31	37	20

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Table 5. Biotic Indices for Shoup's Run and Trough Creek.

Average Taxic Richness (S):	
Shoup's Run	3.4
Trough Creek	6.6
Equitability (E):	
Shoup's Run	0.440329
Trough Creek	0.742337
Diversity (D):	
Shoup's Run	1.49712
Trough Creek	4.899424

Taxic richness, equitability, and diversity (Simpson's) were calculated for each stream. Taxic richness was calculated for each site, and averaged for each stream, whereas equitability and diversity were calculated using all the data from each stream. Tables 6 and 7 show the data used to calculate the EPT indices

for the two streams. Trough Creek's EPT index was 19, indicating high species diversity and likely low pollution. Shoup's Run EPT index was 10, substantially lower than Trough Creek's score and potentially indicative of polluted waters (see Table 8).

Table 6. EPT (Ephemeroptera, Plecoptera, and Trichoptera) Index for Trough Creek

	Great Trough Creek					Total Combined Spp.
	1	2	3	4	5	
Ephemeroptera	3	4	0	2	3	4
Plecoptera	3	4	2	2	1	4
Trichoptera	2	9	2	8	3	11
	8	17	4	12	7	Trough Creek EPT Score: 19

Table 7. EPT (Ephemeroptera, Plecoptera, and Trichoptera) Index for Shoup's Run

	Shoup's Run					Total Combined Spp.
	1	2	3	4	5	
Ephemeroptera	0	0	0	0	0	0
Plecoptera	1	1	1	0	0	1
Trichoptera	7	5	7	2	3	9
	8	6	8	2	3	Trough Creek EPT Score: 10

Table 8. EPT Biotic Index Score Value Table

Rating	Excellent	Good	Good-Fair	Fair	Poor
EPT	>27	21-27	14-20	7-13	0-6

(Taken from NRCS, 2001)

Tables 9 and 10 illustrate the data used to calculate the FBI index for each stream. According to this index, Trough Creek's FBI index indicates the water quality is "Very Good" as it falls between 3.76

and 4.25, whereas the FBI for Shoup's Run is 4.40, which qualifies it as "Good" water quality with "some organic pollution probable" (see Table 11).

Table 9. Hilsenhoff Index (Familial Biotic Index) for Trough Creek

Trough Creek	
Sum of Total Species Found	142
Sum Hilsenhoff Number * Individual Species	537
Sum Hilsenhoff Number / Sum Total Species	3.78

Table 10. *Hilsenhoff Index (Familial Biotic Index) for Shoup's Run*

Shoup's Run	
Sum of Total Species Found	96
Sum Hilsenhoff Number * Individual Species	422
Sum Hilsenhoff Number / Sum Total Species	4.40

Table 11. *Hilsenhoff Biotic Index Score Value Table*

Family Biotic Index	Water Quality	Degree of Organic Pollution
0.00-3.75	Excellent	Organic pollution unlikely
3.76-4.25	Very good	Possible slight organic pollution
4.26-5.00	Good	Some organic pollution probable
5.01-5.75	Fair	Fairly substantial pollution likely
5.76-6.50	Fairly poor	Substantial pollution likely
6.51-7.25	Poor	Very substantial pollution likely
7.26-10.00	Very poor	Severe organic pollution likely

(Taken from PSU, 2003)

DISCUSSION

Our data on water chemistry, and macroinvertebrate biotic indices indicated that Shoup's Run has a lower water quality than that of Trough Creek. Conductivity in Shoup's Run is approximately 5.5 times greater than that in Trough Creek. This difference may be at least partly due to higher metal concentrations in Shoup's Run, which is associated with acid mine drainage. Dissolved oxygen concentrations are also higher in Trough Creek than in Shoup's Run, perhaps because of greater plant life.

Taxic richness, equitability, and diversity of macroinvertebrates are all greater in Trough Creek than in Shoup's Run. The EPT index and Hilsenhoff Index are both higher in Trough Creek than in Shoup's Run, as well. These biotic indices corroborate the water chemistry data showing that water quality is better in Trough Creek. Acid mine drainage is apparently contributing to the lower water quality of Shoup's Run.

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EFFECTS OF SNAKE ODOR ON THE BEHAVIOR OF WILD AND CAPTIVE MICE

Laura A. Daugherty, Nathan Pugh, Tim Shrout, Amy Sutherland and Mike Weaver

ABSTRACT

Wild mice such as the white-footed mouse (*Peromyscus leucopus*) are preyed upon by the black rat snake (*Elaphe obsoleta*) in Pennsylvania. However, captive house mice (*Mus musculus*) have been removed from their natural habitat for many generations. We hypothesized that captive mice have lost some of their instincts to avoid natural predators, such as the black rat snake, as opposed to wild mice which should be expected to avoid areas permeated with a predator odor. In this experiment, a divided aquarium which contained odorless and odor stimulus compartments was used to test the location preference for each mouse species. Captive mice showed a marginally significant preference to avoid the compartment with the snake odor stimulus ($P = 0.069$). No other significant preferences were found.

Key words: chemo-sensing, *Elaphe obsoleta*, *Mus musculus*, *Peromyscus leucopus*, predator-prey interactions

INTRODUCTION

The white-footed mouse (*Peromyscus leucopus*) and the house mouse (*Mus musculus*) are both widely distributed across North America and are common throughout the entire state of Pennsylvania (Merritt, 1987). Large habitat distributions and high population densities make these rodent species optimal specimens to study. Mice form “the base of the vertebrate food chain and thus serve as food for many other animals” (Merritt, 1987). The white-footed mouse and the house mouse occupy similar niches in the wild and are vulnerable to the same predators. In general, they are preyed upon by skunks, weasels, raccoons, foxes, owls, hawks, domestic cats, and various species of snakes. In particular, black rat snakes are common throughout Pennsylvania and the northeastern United States (Hulse et. al., 288). Black rat snakes (*Elaphe obsoleta*) are known to prey upon both white-footed mice and house mice.

Previous studies have shown that spiny mice showed significant changes in behavior patterns and activity when exposed to snake odors (Carere et. al., 1999). This study was designed to compare the behavioral responses of wild mice (*Peromyscus leucopus*) and captive mice (*Mus musculus*) when

exposed to a similar predatory stimulus. Two different species of mice were used in this study because a corresponding captive mouse population was not available for the white-footed mouse. It was hypothesized that wild mice should exhibit behavioral preferences in which they would avoid areas that had been permeated with the scent of a predator that shared the same home range. It was thought that wild mice would tend to avoid areas with black rat snake odor due to natural instincts. Through many generations bred in captivity, evolution would not have selected this as a necessary trait for survival because it was unnecessary for them. Therefore, mice bred in captivity may not have the flight instincts needed to identify the black rat snake as a predator. So, it was also hypothesized that mice bred in captivity would not show an avoidance of the black rat snake odor. If this hypothesis is true, then there should be a difference between the expressed behaviors of wild and captive mice in their location preference.

METHODS AND MATERIALS

The experimental animals included 5 wild (*Peromyscus leucopus*) and 5 captive mice (*Mus musculus*). The wild mice were captured using small

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mammal traps that were set in a small wooded lot behind the Juniata College Brumbaugh Academic Center, as well as at the Juniata College Raystown Field Station. The captive mice were purchased from a local Petsmart.

The mice were tested in an enclosure which consisted of a large aquarium with dimensions of 46 x 12 x 13.5 inches. The aquarium was divided into 3 compartments using plywood walls with cut doors allowing the mice to go from one side to the other. The middle compartment had an area of ~144 in² and the side compartments had an area of ~204 in². Both compartments had a layer of clean newspaper as a substrate and shredded paper towels at the far ends of the chambers to provide some cover (see Fig. 1). One side compartment contained a snake-odor stimulus and the compartment on the opposite side contained no snake odor. The odor was introduced into the compartment using scat covered newspapers and shed skins from black rat snake cages.

The mice were placed sequentially into the central compartment of the enclosure and released after one minute. This was done in order to reduce their stress and to allow them to acclimate themselves to their new surroundings. The location of each mouse was recorded every 15 seconds for a period of 3 minutes. The duration of time spent in each compartment was also recorded for every trial. If a mouse was completely in the middle compartment, it was not recorded as being on either side; however, if a mouse was halfway between compartments, it was recorded as if it had entered the compartment it was facing. Each mouse was labeled for the purpose of the comparison, and the 4 sets of trials were completed in rotation to prevent the mice from becoming habituated to the enclosure. It is important to note that our samples are not completely independent because choices were recorded repeatedly for each of the mice.



Figure 1. Picture of the testing enclosure

RESULTS

Chi-square tests were performed to determine any significant differences between the compartment preferences of the wild and the captive mice for the data involving the mice locations at 15 second intervals. In the control trials, no significant preference was found between the left and right compartments for both species (*Peromyscus*: $\chi^2 = 0.211$, $P = 0.646$; *Mus*: $\chi^2 = 1.056$, $P = 0.304$). In the treatment trials, no significant preference was found between the snake odor and odorless compartments for the wild species ($\chi^2 = 1.162$, $P = 0.281$); however, a marginally significant preference for the odorless compartment was found for the captive species ($\chi^2 = 3.302$, $P = 0.069$). Tables 1 and 2 show the location frequencies of both mouse species for the control and

treatment trials. The χ^2 test for independence was also applied to determine if wild mice showed a significant difference from the captive mice for either control or experimental trials. ($\chi^2 = 0.381$, $P = 0.573$; $\chi^2 = 0.373$, $P = 0.541$). This test showed no significant difference between the compartment preferences of the wild and the captive mice.

Paired t-tests were also applied to determine any significant differences in the time durations that each mouse spent in the compartments. The data showed no significant preferences for either compartment in the control trials of wild and captive mice ($t = 0.01$, $P = 0.993$; $t = 0.12$, $P = 0.908$). The data also show no significant preference for either compartment in the treatment trials of wild and captive mice ($t = 1.17$, $P = 0.256$; $t = 1.25$, $P = 0.228$). Table 3 shows a summary of these results.

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Table 1. Total positional frequencies for control trials of both species.

	Odorless Compartment (L)	Odorless Compartment (R)	Total
Wild Mice	80	72	152
Captive Mice	86	68	154
Total	166	140	306

Table 2. Total positional frequencies for treatment trials of both species.

	Snake Odor Compartment	Odorless Compartment	Total
Wild Mice	61	79	140
Captive Mice	67	100	167
Total	128	179	307

Table 3. Summary of mean and total durations in which mice occupied a compartment.

	Compartment	Mean $\pm$ Stand. Dev. (sec)	Total Durations (sec)
<i>Control</i>			
Wild Mice	Odorless (L)	54.60 $\pm$ 50.08	1092
	Odorless (R)	54.45 $\pm$ 47.50	1089
Captive Mice	Odorless (L)	48.75 $\pm$ 40.07	975
	Odorless (R)	46.75 $\pm$ 49.85	935
<i>Treatment</i>			
Wild Mice	Snake Odor	46.50 $\pm$ 36.58	930
	Odorless	61.80 $\pm$ 44.36	1236
Captive Mice	Snake Odor	58.60 $\pm$ 42.15	1172
	Odorless	80.45 $\pm$ 41.39	1609

DISCUSSION

This study was designed to test the possible behavioral differences between wild and captive mice when they are in the presence of a predator's odor. The results from the control trials confirmed the hypothesis that with the lack of odor in either compartment, the mice would not show a preference. However, captive mice showed a marginally significant preference for the odorless compartment during the treatment trials, whereas the wild mice did not. These results contradicted our hypothesis that the wild mice should have shown a preference, whereas the captive mice should have not. More data involving more animals are needed to verify our results.

Interestingly the wild mice exhibited grooming behavior after exiting the compartment with the snake odor, possibly to rid themselves of the stench of the snake. The captive mice did not groom themselves at all. Future studies should investigate this behavior further.

The lack of support for our hypothesis may be the result of several factors, besides low sample sizes and lack of independence of our data points. One factor is that two different mouse species were used in trials: an Old World (*Mus*) and a New World species (*Peromyscus*). The species may have different levels of chemo-sensory abilities, allowing them to detect the odors. Another reason could be that the design of the enclosure was flawed. The compartments were not airtight and odor may have permeated throughout the entire enclosure, which may have prevented the mice from accurately determining the location of the snake scent. The enclosure was also not cleaned between the trials. This may have affected our results because mice leave odor trails, which they may later follow. This became evident in that it seemed that once a mouse chose one direction, the others seemed to show the same preference, possibly by recognizing and following the odors left by the previous mice. An additional possibility is the level of stress. Wild mice are very adapted to the presence of complete cover, while the captive mice are acclimated to being relatively exposed. This could have affected our trials

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in that the wild mice continually traveled from one chamber to the other, possibly seeking shelter. Their drive for cover may be strong enough that they ignored the presence of the odor. On the other hand, the captive mice were comfortable without the presence of cover and so could avoid the scented chamber. Finally the enclosure should have been covered so that the mice would not be affected by differential lighting and other environmental features surrounding the enclosure.

In conclusion, more research on this study is needed to further understand the behavior of wild and captive mice species. In future studies, researchers should compare the same species of mice. They should also examine the effects of non-predator odors as a control on the effects of predator odors. It would also be interesting to provide a realistic visual of a snake, either combined with snake odor or not. This would allow one to ascertain whether the mice were relying on their sense of smell or sight to identify predators.

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