

# DISTRIBUTION OF SALAMANDERS IN WARM SPRING

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## ABSTRACT

Warm springs are a form of thermal springs that house a variety of salamanders, including Northern dusky salamanders (*Desmognathus fuscus*), spring salamanders (*Gyrinophilus porphyriticus*), and two-lined salamanders (*Eurycea bislineata*). These salamanders live in warm springs to help them thermoregulate and keep their skin moist. They also rely on the warm spring microhabitat to survive. We visited a warm spring in Huntingdon, Pennsylvania to observe the number and species of salamanders we could find in six 15-meter interval plots down the stream. We observed how the substrate changed as we traveled down the stream. We began at the head of the stream with a rock-filled, gravelly, and sandy substrate with mid-sized boulders, lots of vegetation, including duckweed, and a well-covered canopy. As we moved down the stream, the vegetation and coverage dissipated, and it became sandier with few rocks and duckweed. In addition, the stream narrowed as we moved further. Additionally, we took water quality and temperature at each site. After observing different types of salamanders through each plot of stream, we found a total of 7 Northern dusky salamanders, 18 Two-lined salamanders, and 6 Spring salamanders, for a total of 31 salamanders. Our results imply that these microhabitat changes downstream result in different species abundance and richness for salamanders.

*Keywords:* *Eurycea bislineata*, *Desmognathus fuscus*, *Gyrinophilus porphyriticus*, *Warm Spring*, *species area relationship*

## INTRODUCTION

Northern dusky salamanders (*Desmognathus fuscus*), spring salamanders (*Gyrinophilus porphyriticus*), and two-lined salamanders (*Eurycea bislineata*) are lungless salamanders of the Plethodontidae family. Because of their lack of lungs, they rely on respiration through their moist skin. The need for moist skin limits the salamander's habitat selection and ability to thermoregulate. These ectothermic salamanders are known to be thermoconformers and choose temperature-specific microhabitats to thermoregulate (Lunghi 2016).

Furthermore, warm springs fit into the larger category of thermal springs. It is defined as having water temperatures higher than the average air temperature but lower than a human's body temperature (Pentecost 2005). Additionally, a well-known ecological theory is the species-area relationship theory. This theory states that as habitat

area increases, species richness increases as well. The habitat heterogeneity hypothesis also states that the more heterogeneous a habitat is the more species it will have (Kallimanis et al. 2008).

For this study, Warm Spring (Huntingdon, PA) was chosen as our study site because of its compelling stream characteristics and abundance of salamanders. The goal of the study was to see if the microhabitat changes downstream result in different species abundance and richness for salamanders. This study is significant because it provides insights into the ecology and biodiversity of warm springs in central Pennsylvania. Through visual observation of the study site, it is noticeable that the stream appears narrower and feels colder downstream. For these reasons, it was hypothesized that as the distance from the source increases, salamander abundance and diversity should decrease due to lower temperatures and narrower stream conditions further downstream.

## METHODS AND MATERIALS

Warm Spring was divided into 15-meter interval plots starting at the head of the stream, moving downstream to differentiate the habitat. Each section was measured for width, flow rate, pH, temperature, oxygen, conductivity, and substrate. To conduct water chemistry analysis, a YSI meter was used. After calculating the data for each plot, a 20-minute timer was set for the purpose of looking for salamanders. Our methods included scanning the stream bed as well as flipping boulders and rocks to see if any salamanders were hiding under them.

## FIELD SITE

During April 2025, *D. fuscus*, *G. porphyriticus*, and *E. bislineata* were observed from one freshwater stream in Huntingdon County, Pennsylvania. Warm Spring (40.54753°N, 77.97424°W) (see Figure 1) has a few species of aquatic vertebrates and a notable population of crayfish (*Decapoda*). These crustaceans may prey on the salamanders in the stream. We also found numerous other macroinvertebrate species swimming around as well as under the rocks/boulders covering the spring floor.

well-covered canopy. **Plot 2 (15-30 meters)** had a width of 5.50 meters, this trend continues downstream as each plot gets narrower. This plot had a similar makeup of gravelly and sandy substrate, mid-sized boulders, and pebbles with a well-covered canopy. This plot also had a downed tree across the stream, though it didn't touch the water. **Plot 3 (30-45 meters)** was 5 meters wide. This portion of the stream also had a gravelly and sandy substrate. Though with a large portion of duckweed covering the stream's bed, it was difficult to see the substrate makeup completely. Also, this portion had fewer large boulders than the last two. **Plot 4 (45-60 meters)** was up to 4.8 meters wide. This plot had no boulders and very few small rocks for cover. The floor of the stream also had become finer and sandy instead of gravelly as it was before. The duckweed was also much less abundant. **Plot 5 (60-75)** narrowed down in width to 3 meters wide which is a significant decrease from the last (4.8 meters). This plot's floor makeup was almost entirely made up of sand with small amounts of gravel. Besides the small amount of gravel mixed in the floor of the stream, there were no rocks or small boulders in this plot. As for the vegetation, there was little to no duckweed and very small amounts of moss found on a fallen log in the plot. **Plot 6 (75-90)** was 2 meters at its widest point. This final plot was extremely shallow and narrow with substrate made up of only sand. There were no boulders or vegetation in this plot.



Figure 1. This map shows the location of the Warm Spring at 4879 Cold Springs Rd in Huntingdon, Pennsylvania. Standing Stone Creek and the golf course serve as a reference point for location.

Moving down the stream starting at the spring source, the plots were as follows. **Plot 1 (1-15 meters)** was the widest, with a width of 6.1 meters. The plot consisted of a mostly gravelly and sandy substrate, mid-sized boulders, pebbles, and a

## RESULTS

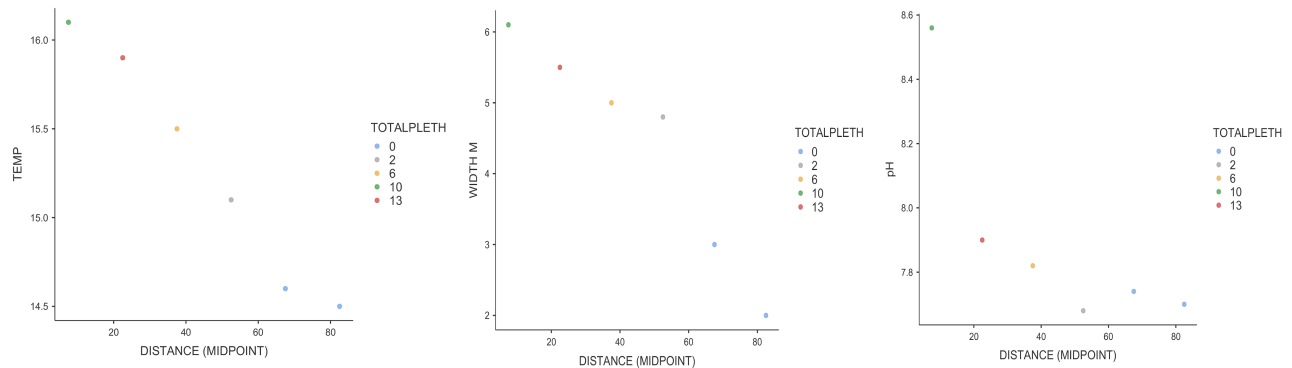
Using Jamovi, Pearson's correlation analyses were conducted to examine the relationships between salamander abundance and environmental variables across six 15-meter stream plots located downstream from the spring source (Table 1).

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Midpoints were calculated to represent distance more accurately. Descriptive statistics were initially used to explore the data and identify trends. As shown in

Figure 2, scatter plots suggest that temperature, stream width, and pH decline with increasing distance. The correlation analysis confirmed several of these trends.

*Figure 2. Descriptive statistics on which values could have the most correlation to salamander richness. TOTALPLETH represents salamander abundance, DISTANCE (MIDPOINT) represents the midpoints calculated to accurately account for the plot's distance from the spring headwaters in meters, pH represents potential of hydrogen, and TEMP represents temperature.*



There was a strong negative correlation between distance downstream and total salamander abundance ( $r = -0.911$ ,  $p = .011$ ) and between distance and species richness of salamanders ( $r = -0.969$ ,  $p = .001$ ). This indicates that both abundance and richness decrease as distance downstream increases. Abundance was positively correlated with species richness ( $r = 0.971$ ,  $p = .001$ ), and strongly associated with temperature ( $r = 0.944$ ,  $p = .005$ ), and stream width ( $r = 0.833$ ,  $p = .039$ ). Richness showed an even stronger correlation with temperature ( $r = 0.993$ ,  $p < .001$ ) and width ( $r = 0.924$ ,  $p = .009$ ). Environmental variables such as pH, specific conductance, and velocity did not significantly correlate with salamander abundance or richness. However, velocity did positively correlate with specific conductance, suggesting a hydrologic relationship.

Further Pearson's correlation analyses were conducted to determine specific relationships between the abundance of individual salamander species and environmental variables (Table 2). The results yielded some significant biological patterns.

Correlation Matrix

|                        |                | TOTALPLETH | RICHNESS  | DISTANCE<br>(MIDPOINT) | pH    | SPC     | TEMP     | WIDTH<br>M | VELOCITY |
|------------------------|----------------|------------|-----------|------------------------|-------|---------|----------|------------|----------|
| TOTALPLETH             | Pearson's<br>r | —          |           |                        |       |         |          |            |          |
|                        | df             | —          |           |                        |       |         |          |            |          |
|                        | p-value        | —          |           |                        |       |         |          |            |          |
| RICHNESS               | Pearson's<br>r | 0.971 **   | —         |                        |       |         |          |            |          |
|                        | df             | 4          | —         |                        |       |         |          |            |          |
|                        | p-value        | 0.001      | —         |                        |       |         |          |            |          |
| DISTANCE<br>(MIDPOINT) | Pearson's<br>r | -0.911 *   | -0.969 ** | —                      |       |         |          |            |          |
|                        | df             | 4          | 4         | —                      |       |         |          |            |          |
|                        | p-value        | 0.011      | 0.001     | —                      |       |         |          |            |          |
| pH                     | Pearson's<br>r | 0.624      | 0.696     | -0.789                 | —     |         |          |            |          |
|                        | df             | 4          | 4         | 4                      | —     |         |          |            |          |
|                        | p-value        | 0.185      | 0.124     | 0.062                  | —     |         |          |            |          |
| SPC                    | Pearson's<br>r | 0.684      | 0.699     | -0.596                 | 0.010 | —       |          |            |          |
|                        | df             | 4          | 4         | 4                      | 4     | —       |          |            |          |
|                        | p-value        | 0.134      | 0.122     | 0.212                  | 0.985 | —       |          |            |          |
| TEMP                   | Pearson's<br>r | 0.944 **   | 0.993 *** | -0.989 ***             | 0.747 | 0.654   | —        |            |          |
|                        | df             | 4          | 4         | 4                      | 4     | 4       | —        |            |          |
|                        | p-value        | 0.005      | < .001    | < .001                 | 0.088 | 0.159   | —        |            |          |
| WIDTH M                | Pearson's<br>r | 0.833 *    | 0.924 **  | -0.959 **              | 0.645 | 0.688   | 0.950 ** | —          |          |
|                        | df             | 4          | 4         | 4                      | 4     | 4       | 4        | —          |          |
|                        | p-value        | 0.039      | 0.009     | 0.002                  | 0.166 | 0.131   | 0.004    | —          |          |
| VELOCITY               | Pearson's<br>r | 0.522      | 0.602     | -0.562                 | 0.015 | 0.835 * | 0.599    | 0.754      | —        |
|                        | df             | 4          | 4         | 4                      | 4     | 4       | 4        | 4          | —        |
|                        | p-value        | 0.288      | 0.206     | 0.246                  | 0.977 | 0.039   | 0.209    | 0.083      | —        |

Note. \*  $p < .05$ , \*\*  $p < .01$ , \*\*\*  $p < .001$

Table 1. Correlation matrix of salamander species richness versus various environmental variables. Significant values are highlighted for reference. TOTALPLETH represents abundance, RICHNESS represents species richness, DISTANCE (MIDPOINT) represents the midpoints calculated to accurately account for the plot's distance from the spring headwaters in meters, pH represents potential hydrogen, SPC represents specific conductance, TEMP represents temperature, WIDTH M represents width in meters, and VELOCITY represents current velocity.

The abundance of spring salamanders (*G. porphyriticus*) was significantly negatively correlated with increase in distance from the spring headwaters ( $r = -0.878$ ,  $p = 0.021$ ). This indicates a strong decrease in abundance downstream. This species also showed a strong positive correlation with temperature

( $r = 0.907$ ,  $p = 0.013$ ), suggesting a possible preference for warmer upstream conditions. Northern dusky salamander (*D. fuscus*) abundance was strongly positively correlated with pH ( $r = 0.989$ ,  $p < 0.001$ ). This is a possible indication that this species prefers less acidic conditions. There was a slightly

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negative correlation between this species and distance ( $r = -0.735$ ,  $p = 0.096$ ), although this result was not statistically significant. The abundance of two-lined salamanders (*E. bislineata*) showed a significant positive correlation to Specific conductance ( $r = -0.959$ ,  $p = 0.002$ ), indicating this species may prefer areas of higher ionic concentration. However, there were no other significant correlations between

two-lined salamanders and other environmental variables.

Table 2. Correlation Matrix of salamander species abundance versus various environmental variables. Significant values are highlighted for reference. *D. FUSCUS* represents Northern dusky salamanders, *E. BISLINEATA* represents two-lined salamanders, *G. PORPHYRITICUS* represents spring salamanders,

| Correlation Matrix  |             | D_FUSCUS  | E_BISLINEATA | G_PORPHYRITICUS | DISTANCE (MIDPOINT) | pH    | VELOCITY | WIDTH M  | TEMP  | SPC |
|---------------------|-------------|-----------|--------------|-----------------|---------------------|-------|----------|----------|-------|-----|
| D_FUSCUS            | Pearson's r | —         |              |                 |                     |       |          |          |       |     |
|                     | df          | —         |              |                 |                     |       |          |          |       |     |
|                     | p-value     | —         |              |                 |                     |       |          |          |       |     |
| E_BISLINEATA        | Pearson's r | 0.022     | —            |                 |                     |       |          |          |       |     |
|                     | df          | 4         | —            |                 |                     |       |          |          |       |     |
|                     | p-value     | 0.967     | —            |                 |                     |       |          |          |       |     |
| G_PORPHYRITICUS     | Pearson's r | 0.532     | 0.683        | —               |                     |       |          |          |       |     |
|                     | df          | 4         | 4            | —               |                     |       |          |          |       |     |
|                     | p-value     | 0.277     | 0.135        | —               |                     |       |          |          |       |     |
| DISTANCE (MIDPOINT) | Pearson's r | -0.735    | -0.600       | -0.878 *        | —                   |       |          |          |       |     |
|                     | df          | 4         | 4            | 4               | —                   |       |          |          |       |     |
|                     | p-value     | 0.096     | 0.208        | 0.021           | —                   |       |          |          |       |     |
| pH                  | Pearson's r | 0.989 *** | 0.090        | 0.635           | -0.789              | —     |          |          |       |     |
|                     | df          | 4         | 4            | 4               | 4                   | —     |          |          |       |     |
|                     | p-value     | < .001    | 0.866        | 0.175           | 0.062               | —     |          |          |       |     |
| VELOCITY            | Pearson's r | -0.013    | 0.641        | 0.438           | -0.562              | 0.015 | —        |          |       |     |
|                     | df          | 4         | 4            | 4               | 4                   | 4     | —        |          |       |     |
|                     | p-value     | 0.981     | 0.170        | 0.385           | 0.246               | 0.977 | —        |          |       |     |
| WIDTH M             | Pearson's r | 0.599     | 0.599        | 0.790           | -0.959 **           | 0.645 | 0.754    | —        |       |     |
|                     | df          | 4         | 4            | 4               | 4                   | 4     | 4        | —        |       |     |
|                     | p-value     | 0.209     | 0.209        | 0.061           | 0.002               | 0.166 | 0.083    | —        |       |     |
| TEMP                | Pearson's r | 0.691     | 0.668        | 0.907 *         | -0.989 ***          | 0.747 | 0.599    | 0.950 ** | —     |     |
|                     | df          | 4         | 4            | 4               | 4                   | 4     | 4        | 4        | —     |     |
|                     | p-value     | 0.128     | 0.147        | 0.013           | < .001              | 0.088 | 0.209    | 0.004    | —     |     |
| SPC                 | Pearson's r | -0.087    | 0.843 *      | 0.716           | -0.596              | 0.010 | 0.835 *  | 0.688    | 0.654 | —   |
|                     | df          | 4         | 4            | 4               | 4                   | 4     | 4        | 4        | 4     | —   |
|                     | p-value     | 0.870     | 0.035        | 0.110           | 0.212               | 0.985 | 0.039    | 0.131    | 0.159 | —   |

Note. \*  $p < .05$ , \*\*  $p < .01$ , \*\*\*  $p < .001$

*DISTANCE (MIDPOINT)* represents the midpoints calculated to accurately account for the plots distance from the spring headwaters in meters, *pH*

represents potential hydrogen, *VELOCITY* represents current velocity, *WIDTH M* represents width in

meters, *TEMP* represents temperature, and *SPC* represents specific conductance.

## DISCUSSION

Pearson's correlation analysis in Jamovi revealed that both salamander abundance and species richness significantly decreased as distance from source of Warm Spring increased. Temperature and stream width also declined downstream and were positively correlated with both salamander abundance and richness, indicating that upstream areas provide more favorable conditions. These results aligned with our hypothesis that as distance from the headwater increased, temperature and favorable stream conditions would decrease, resulting in reduced salamander diversity and abundance. Similarly, these findings coincide with both the species-area relationship theory and the habitat heterogeneity hypothesis. Lungless salamanders' tendency to seek out temperature-specific microhabitats, as explained by Lunghi (2016) is also supported by these results.

Individually, *G. porphyriticus* were more abundant in warmer upstream areas, while *D. fuscus* showed a strong preference for areas with higher pH (less acidic). *E. bislineata* abundance was positively associated with specific conductance, suggesting a link to water chemistry. However, pH, conductance, and velocity showed no overall significant relationship with total salamander abundance or richness. These findings suggest that both physical (temperature, width) and chemical (pH, conductance) factors may influence the distribution of individual salamander species differently, but overall habitat structure plays a key role in supporting greater diversity and abundance upstream.

Because we initially only hypothesized that water temperature, substrate type, and stream width should affect salamander abundance, the correlation with other factors (pH and conductance) was unexpected. Similarly, we expected to find more than just three species of salamanders in this stream, especially as this survey was conducted during favorable salamander conditions in the springtime. However, because of the large coverage of duckweed and other vegetation present in the stream, it was often difficult to find salamanders without disrupting their natural habitat. Similarly, because we looked at

each 15-meter plot for only 20 minutes, there could have been other species that we missed, and thus a more thorough investigation could return different results.

For the purposes of this study, we chose to look only within the stream itself, and not at the surrounding streambank or forest area. Subsequent studies could investigate these habitats as well to check whether terrestrial species diversity and abundance is also dependent on temperature and distance from the spring headwater. Further research could also look at how these factors affect the characteristics of individuals, such as salamander length, weight, sex ratio, and life history traits. Furthermore, surveys of additional warm springs in different locations across central Pennsylvania could help clarify the findings presented in this paper and provide more data to further test this hypothesis.

## ACKNOWLEDGEMENTS

We thank Dr. Douglas S. Glazier and Travis Russel for supplying materials and support for this project. We also thank Juniata College for providing this opportunity.

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