INVESTIGATIONS OF PHYSICAL AND CHEMICAL PROPERTIES AFFECTING CADDISFLY DENSITY

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ABSTRACT

Caddisflies of the order Trichoptera, construct cases in the pupae stage made of silk and materials found in streambeds. These cases can reside in a variety of freshwater habitats. Because variation exists within these environments, this paper explores optimal conditions for caddisfly larvae by comparing density of cases to several variables. These variables included: flow rate, depth, temperature, nutrient levels, and pH. In our results, nutrient and pH data were inconclusive, while shallower, colder, slower moving waters favored higher caddisfly density.

INTRODUCTION

The dynamic relationship between organisms and their environment is common throughout all biomes. Even more intricate is the link between organisms with complex life cycles and their habitats. An example of this is caddisflies of the order *Trichoptera*, an aquatic insect common to Northern Michigan. Caddisflies begin their cycle when eggs are laid in or near freshwater systems. Once hatched, larvae construct transportable cases with silk and materials from the water around them in order to feed and grow. After several months, full larval growth is reached and they anchor their cases to streambed material and seal them from predation (Borror, 1970). During this time they feed by filtering planktonic algae and other particles from passing water (Schlager, 2004).

The order *Trichoptera* can be subdivided by case morphology; those made of stone particles are referred to as stone houses, while those of plant material are called log cabins (Hilsenhoff, 1995). These cases are found in a variety of freshwater environments (Brown, 2004); however, the variation of freshwater habitats leads one to assume that conditions between these locations are likely to have an effect on resident populations. In addition, the chemistry of an ecosystem can drastically limit the types of algae and micro-organisms fit to grow there (Mackay, 1991).

We plan to investigate stream conditions in several locations on the Maple River in Pellston, Michigan. Previous studies have shown that preference in flow velocity can vary between species of filter feeders (Krusnik, 2005). We hypothesize that caddisfly larval density will be higher in slower moving waters because this may allow for more cases to be secured to substrates. Furthermore, the presence of their primary food source, planktonic algae, is known to be denser towards the surface of the water (Regents of University of California, 2004). Thus, we

expect shallower waters to provide higher larval densities because planktonic algae would be more accessible. In addition, we hypothesize that caddisfly larval density will be higher in warmer waters because higher temperatures commonly facilitate productivity (Mackay, 1991), thus fostering growth and development of caddisflies and their primary algal food source.

Additionally, nutrient and pH levels can provide insight into what organisms are able to thrive in an environment (Mackay, 1991). We hypothesize that caddisfly density will be higher in waters with moderate nutrient content because high concentrations may favor aquatic vegetation and take away available substrate, while low levels may not support algal life. We also hypothesize that caddisfly density will be higher in environments of more neutral water (pH 6.5-7.5) because too much variance on either side of neutrality may be to the detriment of algal life necessary to caddisflies.

METHODS

Our study was conducted at local stream sites in Pellston, Michigan (43°30' N; 84°80' W; T36N R4W, Sec. 14) (Figure 1). We chose three sites at varying points along the Maple River: one on the West Branch (site 1), one on the East Branch (site 2), and below the dam where both branches combine (site 3). To account for variability of streambed content, we used 25m transect tapes to lay transects across each site. Along each transect we evaluated the percent area covered by habitable rocks within a 15 cm radius at 1 meter intervals, rated on a scale of 0-5 (0=0%, 1=1-20%, 2=21-40%, 3=41-60%, 4=61-80%, 5=81-100%). The average size of rocks deemed habitable at each interval was estimated using common sporting items such as golf ball, tennis ball, baseball, softball, volleyball, bowling ball, and basketball. These items were

converted to measurements of surface area: 57.18 cm^2, 128.65 cm^2, 175.48 cm^2, 729.03 cm^2, 1464.38 cm^2, 1496.54 cm^2, and 1848.38 cm^2, respectively. These measurements were then used to calculate the average size of rock and percent coverage of habitable substrate for each stream.

For data collection, thirteen rocks from each site were chosen at random locations within the stream. For each rock, data were collected on: depth (cm), flow rate at surface level and at rock bottom (m/s), and temperature (°C). After collecting measurements with a meter stick, flow meter and thermometer, the rocks collected were each sealed in gallon-sized Ziploc bags for preservation. At each site, a 250 ml bottle was filled with water from throughout the stream and chemically analyzed for: pH, NO_3-N concentration (μg N/L), PO_4-P concentration (μg P/L), total P concentration (μg P/L), and total N concentration (mg N/L). In the lab we assessed each rock for caddisfly density. Houses were counted and categorized by material composition: stone house or log cabin, and total rock volume was measured by the technique of water displacement.

In calculating density (houses/cm^2), volume of each rock was converted into surface area by considering each rock a sphere. We then calculated the density of total caddisfly houses on each rock. These values were used to perform a one-way ANOVA test to determine if there was a significant difference in caddisfly density between sites. We continued to investigate our hypotheses based on those results. First, to test whether water speed affected density we analyzed top flow and bottom flow separately in comparison to density across sites. In order to run a Chi-Square test, we created standard divisions for flow rates 1-8 (Table 3), which were categorized in equal ranges to show distribution of density relative to flow speeds. Density levels were also grouped in standard categories 1-7 (Table 2). The Chi-Square test was run to compare significance values for top flow rates and bottom flow rates. These groupings were used to

construct graphs that reflected density distributions across different water speeds for top and bottom flow separately.

To test the affect of depth on caddisfly density across all sites, we used the same process as above. Varying depths were categorized in equal ranges to reflect distribution and ranked 1-5 (Table 4). Density levels were grouped in the same fashion as the previous test (1-7). These values were run using a Chi-Square test to measure significance. A bar graph was also constructed to reflect these distributions. After collecting data to evaluate temperature's relationship to caddisfly density, we realized that temperatures were taken at different times of the day for each site; this did not allow for comparison of temperature between sites. Temperature readings at site 3 were all taken within an hour of each other and are the best gauge of how temperature could affect density of caddisfly houses. Site 3 presented the most variation due to the influence of ground water; values ranging from 18°-20°C were plotted against the average densities found at each temperature.

RESULTS

The caddisfly densities and other variable measurements including pH, nutrient content, and streambed habitability can be found in Table 1 respective to each stream. Of the populations counted from each stream, relevant ANOVA test results showed density differences were significant between Sites 1 and 2 ($p= 0.001$) and between Sites 1 and 3 ($p= 0.000$), but not between Sites 2 & 3 (0.918). The subsequent tests run examined causes for such significance with a Chi-Square test looking at density with respect to flow rate at surface of the water. A significant p-value of 0.017 was found. Rocks with higher densities were found in water with

slower flow rate at surface level (Figure 2). The Chi-Square test looking at density with respect to flow rate at bottom of streambed similarly showed that rocks with higher densities were primarily found at lower flow rates (Figure 3). When examining the effect of depth on caddisfly density a Chi-Square test showed distributions of rocks in categorical densities based on depth (Figure 4). Additionally, Figure 5 presents average density at each temperature reading in site 3.

DISCUSSION

Significant results were only obtained from flow rate at the surface of the stream. However, the trend seen in Figure 3 parallels that of Figure 2 supporting our hypothesis that higher densities would be found in slower moving waters. With regard to depth, there was a definite trend showing rocks with the highest densities were at depths of 21-40 cm (Figure 4) thus supporting our hypothesis that higher densities would be at shallower depths. Additionally, our results show a trend that colder temperatures support more caddisfly density; thus our prediction of warmer temperatures supporting caddisfly growth is rejected. It should be noted that our results were gather from a very limited sample of variation, and may not be applicable to a wider range of temperature.

Our nutrient evalutations showed that Site 1 with the highest larval density had the lowest total phosphorous (Table 1). In contrast, the results of our other nutrient tests (nitrates, total nitrogen, and phosphate content) showed that site 1 continuously had the highest concentrations of each variable tested, and was consistently followed by Site 3 and lastly Site 2. This analysis did not support our hypothesis that intermediate nutrient levels would lead to higher caddisfly densities, and remains inconclusive. When examining pH in respect to caddisfly density, our sampling did not provide enough variance to uncover a true preference. It was shown that site 1

with the highest density of larvae also had the lowest pH reading of 8, but we suspect that these are not directly related. The other sites had only slight variation in pH comparatively (pH 8.04 for site 2 and 8.11 for site 3) (Table 1). This minute variance left us with inconclusive results.

A possible explanation for slower moving water containing higher caddisfly density could be more secured attachment to underwater substrates in areas of low flow. Cases built in turbulent waters may easily be detached from underwater substrates and likely resulting in larval death. In this scenario, the only larvae left would be those attached to substrates in less turbulent waters. Also, these species of caddisflies may prefer or be more effective filter feeders in this range of flow.

As shown above, slower moving water is preferred. We suspect the movement of water allows planktonic algae, which is known to be denser towards the surface (Regents of University of California, 2004), to reach depths of caddisfly larvae; therefore higher density at moderate depth is plausible. If in a situation of optimal depths the larvae would be surrounded by planktonic algae as well as sufficient water velocity to aid in filter feeding. Beyond this range, success of larvae may be challenged due to limited availability of resources. These factors both individually, and combined may indicate the caddisfly's placement as an act of optimal feeding.

Previous studies have shown that planktonic algae prefer and are more productive in cold water (Regents of University of California, 2004). Since planktonic algae serve as caddisfly larvae's primary food source, this likely explains why higher densities of caddisfly larvae were found in colder waters (Regents of University of California, 2004). In addition, it is possible that higher temperatures support growth of competing organisms therefore creating a shortage of resources required for caddisfly development.

Our nutrient analysis showed that Site 1 was always at the high end or low end of the nutrient range (Table 1). This could suggest that low levels of total phosphorous and high levels of nitrates, phosphates and total nitrogen align with higher caddisfly densities. Past studies have been unable to show any correlation between nutrients and larval densities (Cummins, 2003; Mackay, 1991), and our results need more investigation to solidify any true trend. Previous studies have also shown that some species of caddisfly larvae are able to survive in very acidic conditions with pH as low as 3 (Dropkin et al., 2009). This demonstrates that *Trichoptera* larvae are able to live in a large range of conditions and varied tolerance of pH can be attributed to different species of caddisflies. Our study did not distinguish between species; this, coupled with lack of pH variation, showed we could not conclude anything from our study of pH.

Although our study showed some significant results regarding the preferred habitat of caddisfly larvae, there remain areas in which our study could have been improved. First and foremost a larger sampling of rocks from more streams could have provided a better representation of the wide array of physical and chemical conditions. We also had trouble with some of the equipment used in this study. Furthermore, during transportation from the stream to the lab some caddisfly houses fell off their rocks, which did not allow us to evaluate densities on different faces of rocks.

Another way that site 1 noticeably differed from both sites 2 and 3 are in that both the average size of rock and percent of streambed covered in habitable rocks was substantially less (Table 1). Any correlation between streambed content and density is suspect, and may be further defined in additional studies. A relationship between substrate size and survival of larvae or the size of surrounding larval populations and fitness could further be investigated. It was noticed that a parallel trend of more log cabin houses was found in these areas with smaller average rock

size. We found no other studies to support this trend, but further examination of species may provide clearer insight.

Case morphology is one way to classify caddisflies; however, we were unable to identify each insect down to the species level. Additional studies may find more species-specific patterns among the tested variable. Because we analyzed each rock as a whole instead of individual larvae, the general locations of cases on each rock were not noted. Future studies could better quantify and analyze the effect location has on survival. Additionally, because our small testing range could have provided skewed results regarding caddisfly preference, a larger testing range is advised for future studies. A wider variation of variable such as pH, nutrients and temperature could better solidify any present relationship. Furthermore, a lab component isolating nutrients and pH could provide a more controlled testing environment.

We only considered abiotic factors in this study, but there are many biotic factors that may affect caddisfly larvae density, specifically the study of food abundance and density of adult caddisflies in each stream. Food amount could further explain why there seems to be an optimal temperature, flow rate and depth for the larvae. Similarly, a higher number of adults could lead to a higher number of larvae at that site. Studies of these factors could prove vital to the understanding of the distribution of these organisms.

We have shown that slower moving water, shallower depths, and lower temperatures facilitate caddisfly growth in Maple River. The results of this study show small trends found in a limited area, but can hopefully influence further investigations in optimal conditions for caddisfly larvae.

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FIGURE 1: MAP OF SAMPLE SITES

(Figure 1 displays the locations surveyed)

TABLE 1: Site-specific Variable Measurements

Variables	Site 1	Site 2	Site 3
Density (houses/cm^2)	0.06934	0.00932	0.00439
# Log Cabins	98	22	20
# Stone House	292	96	16
% range of habitable substrate	41-60	61-80	61-80
Average surface area of habitable substrate (cm^2)	215.58	845.48	820.57
NO_3-N content (µg N/L)	353.2	5.9	144.1
PO_4-P content (µg P/L)	2	1.6	1.9
Total N content (mg N/L)	0.545	0.407	0.52
Total P content (µg P/L)	10.1	13.6	27
pH	8	8.04	8.11

(Table 1: displays referenced variable measurements for each of the three sites)

TABLE 2

Density Categories Used in Fig 2-4	
Category	**Density (houses/cm^2)**
1	0.000-.025
2	0.026-.050
3	0.051-.075
4	0.076-.100
5	0.101-.125
6	0.126-.150
7	0.151-.175

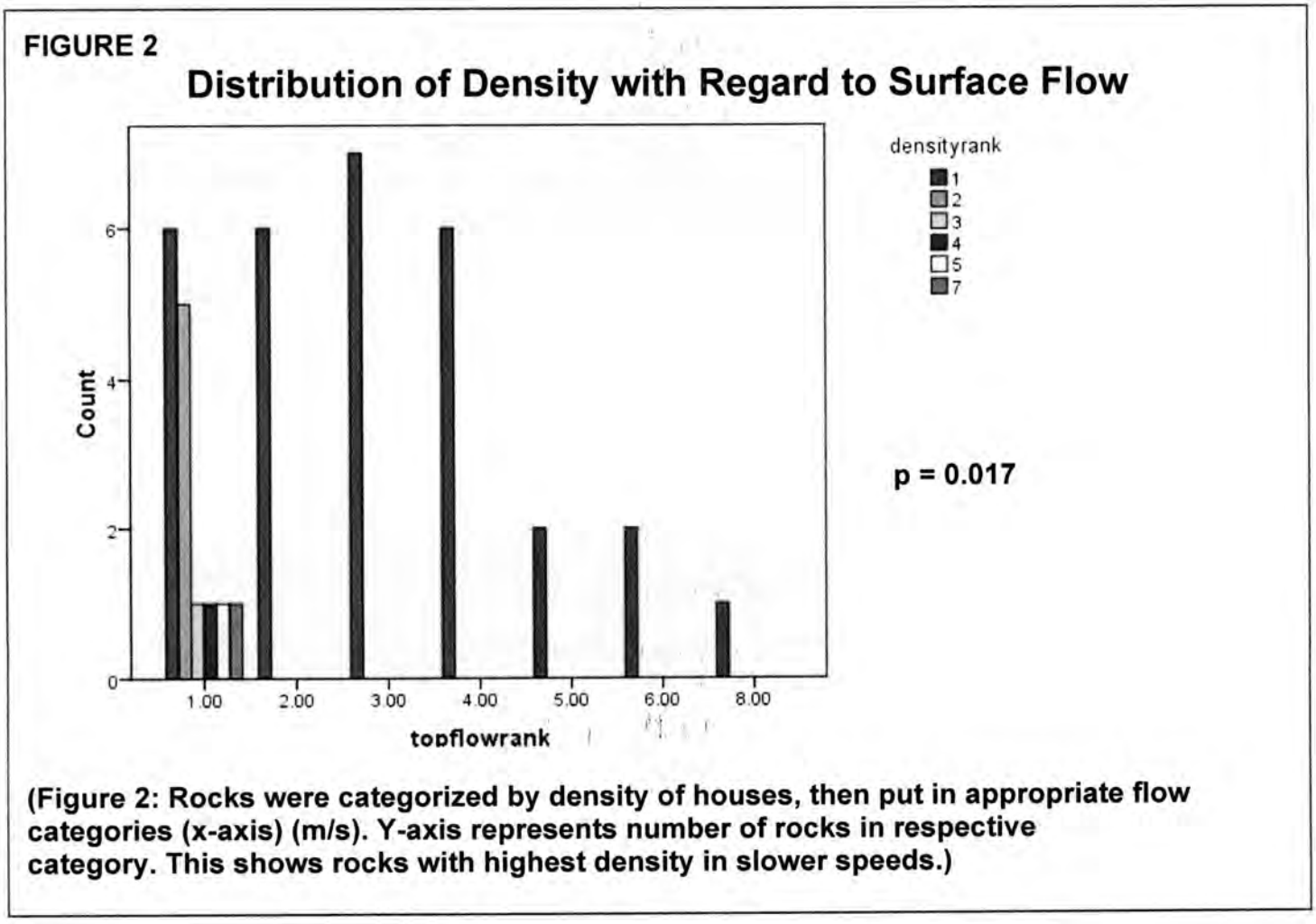
Categories of equal range to show distribution of relative densities.

TABLE 3

Flow Categories Used in Fig 2&3	
Category	**Speed (m/s)**
1	0-0.2
2	0.21-0.4
3	0.41-0.6
4	0.61-0.8
5	0.81-1.0
6	1.01-1.2
7	1.21-1.4
8	1.41-1.6

Categories of equal range to show distribution of relative flow speeds.

FIGURE 2



(Figure 2: Rocks were categorized by density of houses, then put in appropriate flow categories (x-axis) (m/s). Y-axis represents number of rocks in respective category. This shows rocks with highest density in slower speeds.)

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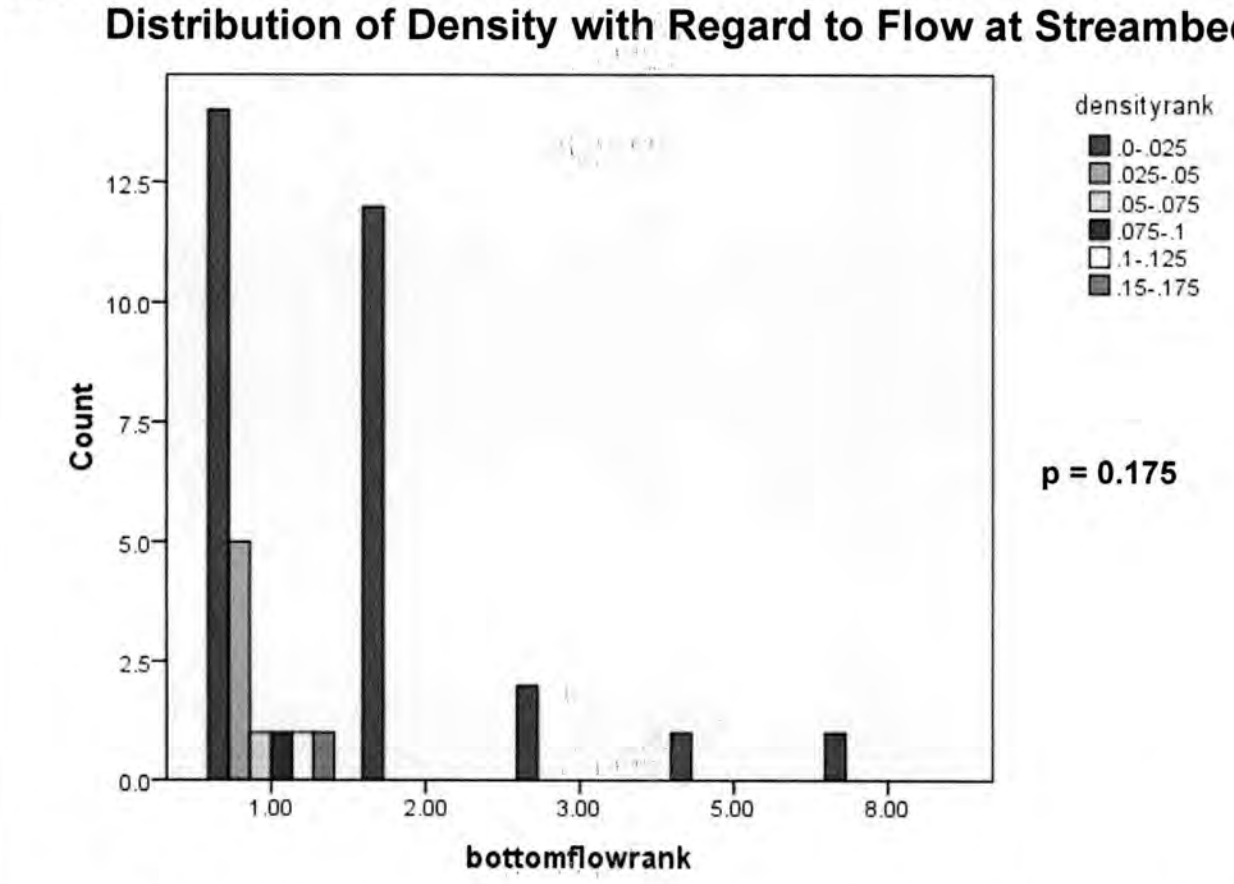
FIGURE 3



(Figure 3: Rocks were categorized by density of houses, then put in appropriate flow categories (x-axis) (m/s). Y-axis represents number of rocks in respective category). This shows that rocks with higher densities were found in slower water.

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Density Categories Used in Fig 2-4	
Category	**Density (houses/cm^2)**
1	0.000-.025
2	0.026-.050
3	0.051-.075
4	0.076-.100
5	0.101-.125
6	0.126-.150
7	0.151-.175

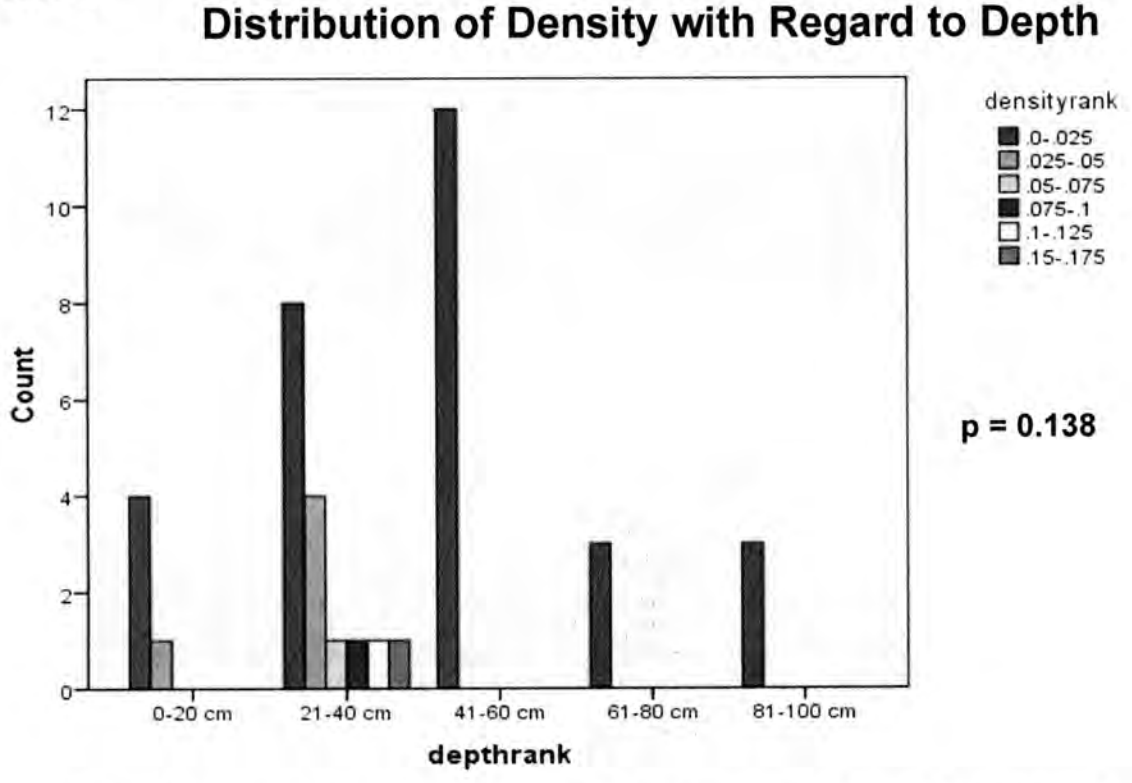
Categories of equal range to show distribution of relative densities.

TABLE 4

Depth Categories Used in Fig 4	
Category	**Depth (cm)**
1	0-20
2	21-40
3	41-60
4	61-80
5	81-100

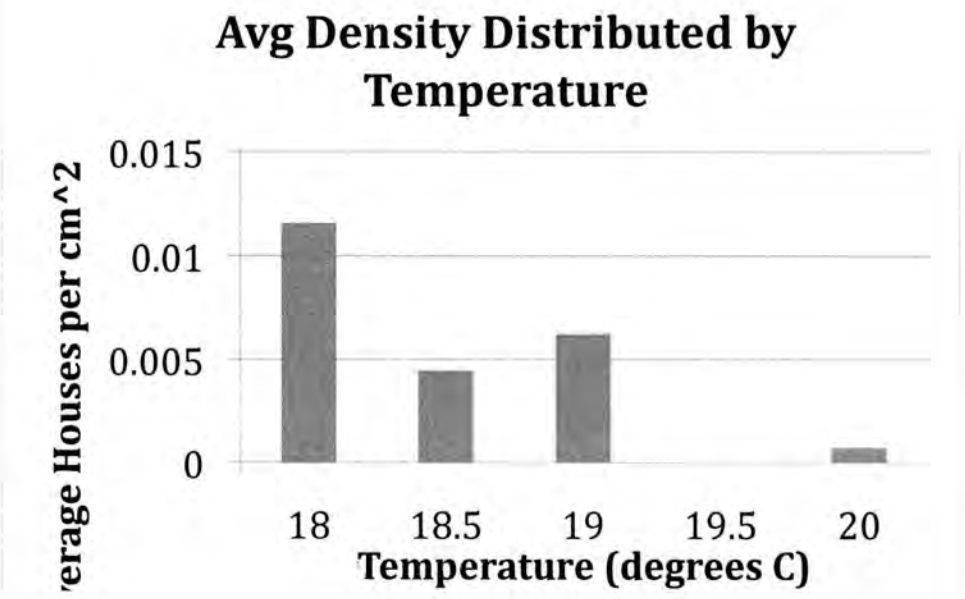
Categories of equal range to show distribution of relative depths.

FIGURE 4



(Figure 4: Rocks were categorized by density of houses, then put in appropriate depth categories (x-axis) (cm). Y-axis represents number of rocks in respective category). This shows that rocks with higher densities were primarily found in a depths between 21-40 cm.

FIGURE 5



This is a graph of average density of caddisfly larvae at different temperature throughout site 3. Though we cannot test for significance, there appears to be a negative trend between density and temperature.

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