The Effects of Sunlight, Disturbance, and Food Availability on the Distribution of Chironomid Larvae

Marcus Lockett, Joseph Rhoades,
Drew Robison, Devan Rouse

University of Michigan Biological Station
EEB 381: General Ecology
Friday, August 14th, 2009
Robert Pillsbury

ABSTRACT

This study investigated the effects of sunlight and disturbance on the distribution of Chironomid larvae in river systems. Artificial streams were created using water pumped from the Maple River and plastic gutters. Tiles were placed in each stream as a surface for Chironomids to colonize. Half of the streams were shaded and disturbance was imposed on some of the tiles in each stream, while the others remained undisturbed. After six weeks, the number of Chironomids present on four of the seven tiles from each stream were counted.

There was no significant difference in the number of Chironomids on tiles exposed to direct sunlight and those in shaded streams. However, disturbance had a large effect on the number of Chironomids present; little or no Chironomids were found on disturbed tiles. In undisturbed areas, it appeared that Chironomids had a non-significant tendency colonize on shaded tiles. Therefore, disturbance seemed to effect Chironomid distribution while amount of sunlight had little to no effect.

INTRODUCTION

The distribution of most organisms is heavily dependent on environmental variables, such as temperature, pH, water and food availability, amount of sunlight, habitat structure, and disturbance patterns. Many of these factors are interdependent, for instance, sunlight affects temperature and, consequently, algae growth. Algae provide important resources to many aquatic organisms, such as food and habitat. Therefore, the abundance of algae largely affects the distribution of said organisms (Pennak 1978). Although light is necessary for algae to carry out photosynthesis, light can be harmful to other organisms. Excess amounts of UV radiation can damage cells and decrease the survival of organisms (De Cauwer et al. 2006).

Another factor affecting the distribution of organisms is disturbance. Disturbance to a habitat directly harms most individuals living there, but also encourages certain species that are more adapted to resist disturbance or recolonize after a disturbance than otherwise stronger competitors (Cain et al. 2008). In this study, both light and disturbance levels were examined to determine their effect on the distribution of Chironomid larvae. Also, the amount of algae on the tiles was examined to test whether algae and Chironomid abundance are correlated.

Chironomids are widely-distributed, non-biting midges that are obligated to lay their eggs and spend their larval stage in freshwater. Larvae are highly dependent on the availability of food, water quality and depth, and substrate. They preferentially colonize soft sediments, hard rocks, and woody debris (Pinder 1986). The larvae build protective tubes composed of silt, sand or algae, that are held together by saliva and act as a semi-permanent home from which they can feed. During the larval stage, they feed on algae, aquatic plants, and organic detritus (Pennak 1978).

The purpose of this study is to determine if there is a difference in the distribution of these larvae between shaded or unshaded, and disturbed or undisturbed habitats. If Chironomid larvae distribution is dependent on algal concentrations, a larger number of individuals should be present on the unshaded habitats where algae coverage is greater. If their distribution is instead dependent on amount of sunlight, then a larger number of individuals should exist in unshaded habitats even in the absence of algae. If disturbance hinders their distribution, as expected, then there will be fewer Chironomids on all disturbed tiles regardless of sunlight and algae presence. Disturbance of the habitat will partially determine the availability of food, but will also remove some Chironomids, perhaps leading to differences in the distribution versus undisturbed sites (Reice, 1985; Hax & Golladay 1998). If both shading and disturbance negatively influence Chironomid distribution, then unshaded, undisturbed habitats should be the ideal habitat in this experiment.

MATERIALS AND METHODS

This study was conducted at the Maple River Stream Lab at the University of Michigan Biological Station. Water was pumped from the river, filtered through panty hose, and delivered to a pair of outdoor head tanks. Each tank was used to supply a constant flow of water to eight artificial streams. Each stream was made using eight foot sections of plastic rain gutters. The streams were held on a table and elevated at the source end to ensure a flow downstream. Seven small ceramic tiles were placed in a line approximately one foot away from the stream head. These tiles were placed to act as artificial stones. Small bricks were placed toward the end of stream to increase depth at the tiles. To create shade, a double layer of black mesh screen was placed over the tiles in half of the streams in an alternating manner. The flow rate was adjusted on each stream to approximately the same flow in all streams. This setup was allowed to flow freely for three weeks to allow organisms from the Maple River to colonize the streams. The colonies on the tiles were targeted for this experiment.

After three weeks, a paint brush was used to lightly wipe the three tiles furthest downstream in each stream. These tiles were brushed three times to simulate a disturbance to the organisms growing on the tiles. Also, the sediments that had built up downstream of the tiles were brushed away each time the disturbance was induced. This disturbance was done twice a week for three weeks. The flow rate of each stream was checked and adjusted during these visits to ensure similar flow rates continued. After six weeks, two additional tiles were added to each stream: one at the upstream and downstream end of the line of tiles. Water depth at the tiles, temperature in the head tank, percent sunlight reaching the tiles (using a light meter), and flow rate were all measured during the trial.

A week later, the two one-week old tiles were collected from each stream along with one disturbed and undisturbed tile selected at random, for a total of 64 tiles. Chlorophyll-a was measured on these same streams. Samples were taken representing the four combinations of shading and disturbance. This data was used to determine the algae coverage for the light versus shaded streams. Using a dissecting microscope, the number of Chironomids of each species were counted and recorded according to the environment in which they colonized and were collected (Tables 1A and 1B). The data was divided into the following categories: light disturbed, light undisturbed, shaded disturbed, shaded undisturbed, light algae-free and shaded algae-free (Table 2).

A Kruskal-Wallis test was performed to find out if at least one of the four groups were significantly different from the others. Furthermore, Mann-Whitney U tests were performed to determine if there was a significant difference in Chironomid numbers between lit and shaded tiles or disturbed and undisturbed tiles. The tiles were categorized into two groups based on disturbance. Two more Mann-Whitney U tests were used to determine if there was a significant difference in Chironomid numbers between lit and shaded tiles in these groups.

RESULTS

A non-parametric one-way ANOVA (Kruskal-Wallis test) showed that there was a significant difference in at least one of the four groups: shaded/disturbed, shaded/undisturbed, light/disturbed, light/undisturbed (Table 3; $\alpha = 0.05$, $p = 0.008$). The Mann-Whitney U test showed no significant difference in lit or shaded tiles when considered as a whole (Table 4; $\alpha = 0.05$, $p = 0.399$). However, there was a significant difference in population size of Chironomids between all disturbed and undisturbed tiles (Table 5; $\alpha = 0.05$, $p = 0.002$). The Mann-Whitney U test conducted to compare the Chironomid distributions between the undisturbed light and undisturbed shaded tiles yielded insignificant results ($\alpha = 0.05$, $p = 0.109$). When the same test was used to compare the disturbed light and disturbed shaded tiles, slightly more insignificant results were obtained ($\alpha = 0.05$, $p = 0.643$).

DISCUSSION
By: Marcus Lockett

Our results strongly suggest that disturbance has a greater effect on Chironomid distributions than the amount of sunlight present. Even when disturbance was low, the results, while statistically insignificant, appeared to have a slight preference toward shaded tiles. This makes sense, as excess sunlight damages cells and decreases survival of organisms (DeCauwer et al. 2006). Furthermore, given that algae provide food and habitat for many aquatic organisms such as Chironomids, there should be a greater distribution of Chironomids where there is an abundance of algae (Pennak, 1978). Additionally, algae produce more chlorophyll under reduced-sunlight conditions (based on unpublished research by Barnas et al. 2009). Therefore, it makes since that Chironomids would migrate toward shaded areas. Because there was minor insignificance, further studies (and additional statistical power) is needed.

The slight statistical insignificance in regard to undisturbed Chironomid colonization differences under light and shade could possibly be due to our small sample sizes in each group. The data was too small to have a normal distribution. As such, non-parametric tests were used. Non-parametric tests are not as powerful as parametric tests, and as such, our chances of erroneously failing to reject our null hypothesis (i.e., no difference between undisturbed Chironomids under shade and under light) are higher when using these kinds of test. Therefore, if we had a larger sample size, we may have had a normal distribution. In turn, if our data had been normally distributed, we may have seen more statistical significance.

It was expected that disturbance would have such a significant effect on Chironomid distributions. A study done by Seth Reice was done in a similar way. In it, Chironomids dwelling on rocks were turned as much as twice in a 6 week span. According to Reice, there was a 21-95% reduction in taxa for a population under disturbance compared to control patches. Further, Reice concludes that disturbance is a major determinant of lotic community structure and species diversity (1985). The purpose of his study was to study recovery of macroinvertebrates after disturbances. From his study, he found that the macroinvertebrate community living on the rocks were resilient, returning to normal population numbers in 4 weeks. This has implications of the Intermediate Disturbance Hypothesis, which states that biodiversity is highest when disturbances are neither too rare nor too frequent. Therefore, it would be interesting to include diversity as a parameter in a future study similar to ours.

DISCUSSION
By: Joseph Rhoades

The Chironomid distribution found on the tiles shows that these small invertebrates are highly sensitive to disturbance, with highly significant results indicating that there were less Chironomids on the tiles which were disturbed twice a week. This makes sense considering the results from Hax and Golladay (1998), who found that it took several weeks for populations of invertebrates living on the substrate to recover from major disturbances, major non-regular flooding of the river in their case. It seems very likely that a disturbance occurring as often as two times a week is more than enough to prevent the Chironomids from becoming well-established on the provided substrate, a future test might disturb the tiles once a week for the same number of weeks, or every second week for a longer time period. If attempting to use Chironomids as water-quality indicators (Frouz, 1999), it is probably important to take samples from an area that has been relatively undisturbed for several weeks.

The data, though only almost significant, also suggests that the Chironomids display a slight preference for shaded areas over areas exposed to full sunlight. This may be related to temperature effects, as Pinder (1986) states that larva development in Chironomids is highly dependent on temperature, however he indicated only that low temperatures slowed development, not that high temperatures are detrimental. It is also possible that this may be related to the availability of food in the shaded streams, which had higher chlorophyll on the tiles retained for comparison of algae density. Since algae is one of the primary foods for larval Chironomids (Pinder 1986), it would make sense that they might preferentially colonize areas with higher food availability. One reason that the data was not significant may be the unusually cold and cloudy summer experienced during the test period. A future test to look more specifically at shade vs full sun might check the Chironomid distribution and replace the tiles on a more regular basis and compare the data to local weather, paying particular attention to weeks with mostly sunny days when comparing the shaded and unshaded streams.

DISCUSSION
By: Drew Robison

Based on the experimental results, Chironomid distribution was statistically related to disturbance, but not to light. Disturbance proved to be a dominant force in controlling Chironomid larvae numbers (Table 4). This observation agrees with the studies conducted by Hax and Golladay (1998) and Reice (1985), which showed it took weeks for Chironomids to recover from disturbances. Since the disturbance in this experiment was imposed biweekly, it would be difficult for Chironomids to recover to normal population levels. Conversely, light was not shown to significantly affect Chironomid numbers.

However, after the Chironomid population was split into groups based on disturbance, there was an almost significant correlation between Chironomid distribution and light levels on the undisturbed tiles. Although the p-value was over the alpha level, it was close to showing a significant relationship. The results suggested that Chironomids preferred shaded streams. Possible causes for this pattern could be the intensity of light, the temperature, or the availability of food. In an unpublished report by Barnas et al. (2009) on the same streams as used in this experiment showed that there was a significant difference in chlorophyll α densities between lit and shaded tiles. The shaded tiles had higher levels of cholorphyll α per unit area. Higher levels of chlorophyll α suggest that there is more food available for the Chironomid larvae, providing better habitat. Previous studies have shown that Chironomid distribution is directly correlated to chlorophyll-a abundance in lake systems (Jonasson 1972).

Sources of error in this experiment included unintentional disturbance and inconsistencies between streams. First, the imposed disturbance may have affected the other tiles. The brushing motion created a sloughing of the water in the stream which may have disturbed the tiles that were meant to be free of disturbance. Handling the tiles also imposed disturbance on them which could have changed the Chironomid density. Flow rates and water levels in the 16 streams were very similar, but not exact. These small differences may have caused variation in the results. Studies have shown that depth is a significant factor in Chironomid larvae distribution Real and Prat (1992). Differing flow rates may have made it more difficult for Chironomids to settle in certain streams. Taking more care to equate these factors in the streams would prevent them from affecting the results.

Further research in this area should include many more samples. Although 16 streams were used, very small numbers of Chironomids were found. Finding statistically significant results with small data sets is difficult. Greater numbers should confirm results found in this experiment, and might reveal trends not clearly shown here. Also, tests using only the variable of light could determine if there is a correlation between light and Chironomid density. Since disturbance was by far the dominating factor in Chironomid distribution, removing this variable could allow for testing of other variables. Investigating disturbance levels and their effect on Chironomid densities would also be interesting. The intermediate disturbance hypothesis states that species richness will be greatest at intermediate levels of disturbance. Imposing disturbance at different time intervals could reveal what level of disturbance significantly affects Chironomid numbers.

DISCUSSION
By: Devan Rouse

Disturbance appears to have a greater effect on Chironomid distributions than the amount of sunlight present. On tiles that experienced high disturbance, there was little to no Chironomid colonization regardless of the amount of sunlight present (Table 4). When disturbance was low, the results (while still statistically insignificant) were almost significant in that there appeared to be a non-significant preference for shaded tiles over those with a lot of sunlight (Table 5). A larger sample size or a longer time allotted to allow Chironomids to colonize tiles may yield statistically significant results for this observation.

The tiles that were placed in the stream for one week, were to control for food availability in lit and shaded regions. One week is not a sufficient amount of time for algae colonization to occur. If Chironomids were present on these new tiles, it can be assumed it is not because of food abundance. The only factor that contributed to Chironomid distribution on the new tiles was amount of sunlight. Because little to no Chironomids were found on these new tiles, despite amount of sunlight, the data were inconclusive in relation to our hypothesis. There may not have been enough time for the colonization of Chironomids to take place, or the lack of food may have encouraged colonization elsewhere.

There were many sources of error that could have affected the accuracy of the results. For example, too much disturbance may have been applied to tiles in this experiment and may have completely washed away all Chironomids each time so the data for disturbed tiles is inconclusive. Also, when disturbance was applied, the brushing motion created a sloughing of the water in the stream which may have disturbed the tiles that were meant to be free of disturbance. Handling the tiles imposed unintended disturbance on them as well which could have changed the Chironomid density. Flow rates of each stream were also slightly variable which could have some effect on Chironomid distribution. Chironomids are mobile invertebrates and may have crawled off the tiles before counting took place, so the population sizes could have been skewed.

The effect of disturbance on Chironomids has been previously studied in Spanish reservoirs. Reservoirs subjected to fluctuations of water level and mixing of sediments have a limited amount of Chironomid species present (Seminara and Bazzanti 1988). This supports our results that Chironomid distribution is affected by disturbance. Only populations that have evolved to resist disturbance can prosper in areas subjected to these interferences. Oxygen abudance has also been proven to greatly affect Chironomid distribution; the abundance of organisms is positively correlated with amount of oxygen in environment (Real and Prat 1992).

In a chlorophyll analysis of the algae growing on the tiles, it was found that the mass of chlorophyll-a on the shaded tiles was larger than on the lit tiles (based on unpublished research by Barnas et al. 2009; Graph 1). This could be because algae growing in shaded areas needed to compensate for the lack of sunlight to be productive. Chironomids were more abundant in shaded regions where chlorophyll-a mass was large. Previous studies have shown that Chironomid abundance is directly correlated to chlorophyll-a mass in lake systems (Jonasson 1972). However, Real and Prat (1992) found that the amount of chlorophyll-a was negatively correlated with abundance of Chironomids in Spanish reservoirs. Inconsistencies in these results could be due to many different environmental factors that influence each ecosystem.

Future studies may include testing whether Chironomids in river systems follow Connell's intermediate disturbance hypothesis, which states that species richness will be greatest at intermediate levels of disturbance. This could be done by imposing different levels of disturbance on different communities of Chironomids regularly over a long period of time and measuring the effect each has on species richness.

ACKNOWLEDGMENTS

We would like to thank the following people for assisting us in our research: Robert Pillsbury and Sharon Shattuck for providing assistance to us in every stage of our research, Troy Keller for providing us with our materials and allowing us to work at the stream lab, and the general ecology class at the University of Michigan Biological Station for aiding in the set-up of our experiment and analyzing chlorophyll mass.

	8	7	6	5	4	3	2	1	TOTAL	AVERAGE
New Top	0	0	0	0	0	0	2	0	2	0.444444
Undisturbed	2	0	1	3	2	0	6	1	15	3.333333
Disturbed	2	0	0	0	0	0	0	0	2	0.444444
New Bottom	0	0	1	0	0	1	0	0	2	0.444444
Depth	2.1	2.1	2.2	2.7	2.6	2.4	2.2	2.3		1.166667
last flow rate	163.333333	223.3333333	176.6667	310	270	210	86.66667	90		1.347222

Table 1A *- Stream set A with number of Chironomids present in each tile in each stream. Depth and flow rate of each stream. Average depth, average flow rate, and average number of Chironomids in each tile reported. Even-numbered tiles were shaded.*

	8	7	6	5	4	3	2	1	TOTAL	AVERAGE
New Top	0	0	0	0	0	0	1	1	2	0.444444
Undisturbed	0	1	4	1	1	1	1	0	9	2
Disturbed	0	1	0	0	0	0	0	1	2	0.444444
New Bottom	2	1	0	4	0	0	0	0	7	1.555556
Depth	2.5	2.5	1.9	2.5	2.4	2.7	2.3	3.1		1.111111
last flow rate	186.666667	183.3333333	186.6667	213.3333333	186.6667	210	176.6667	203.3333		1.277778

Table 1B *- Stream set B with number of Chironomids present in each tile in each stream. Depth and flow rate of each stream. Average depth, average flow rate, and average number of Chironomids in each tile reported. Even-numbered tiles were shaded.*

	Shaded	Unshaded
New Top	3	1
Undisturbed	17	7
Disturbed	2	2
New Bottom	3	6

Table 2*: Total number of Chironomids for each group of tiles under shaded and unshaded conditions.*

Test Statistics[a,b]

	Chironomid
Chi-Square	11.755
df	3
Asymp. Sig.	0.008

Table 3*: Kruskal-Wallis test results for investigating between four groups of Chironomid conditions: shaded/undisturbed, lit/disturbed, lit/undisturbed.*

Group	Count	Rank Sum
1	8	71
2	8	65
Mann-Whitney U		35
2-Tailed Significance		0.798
1-Trailed Significance		0.399

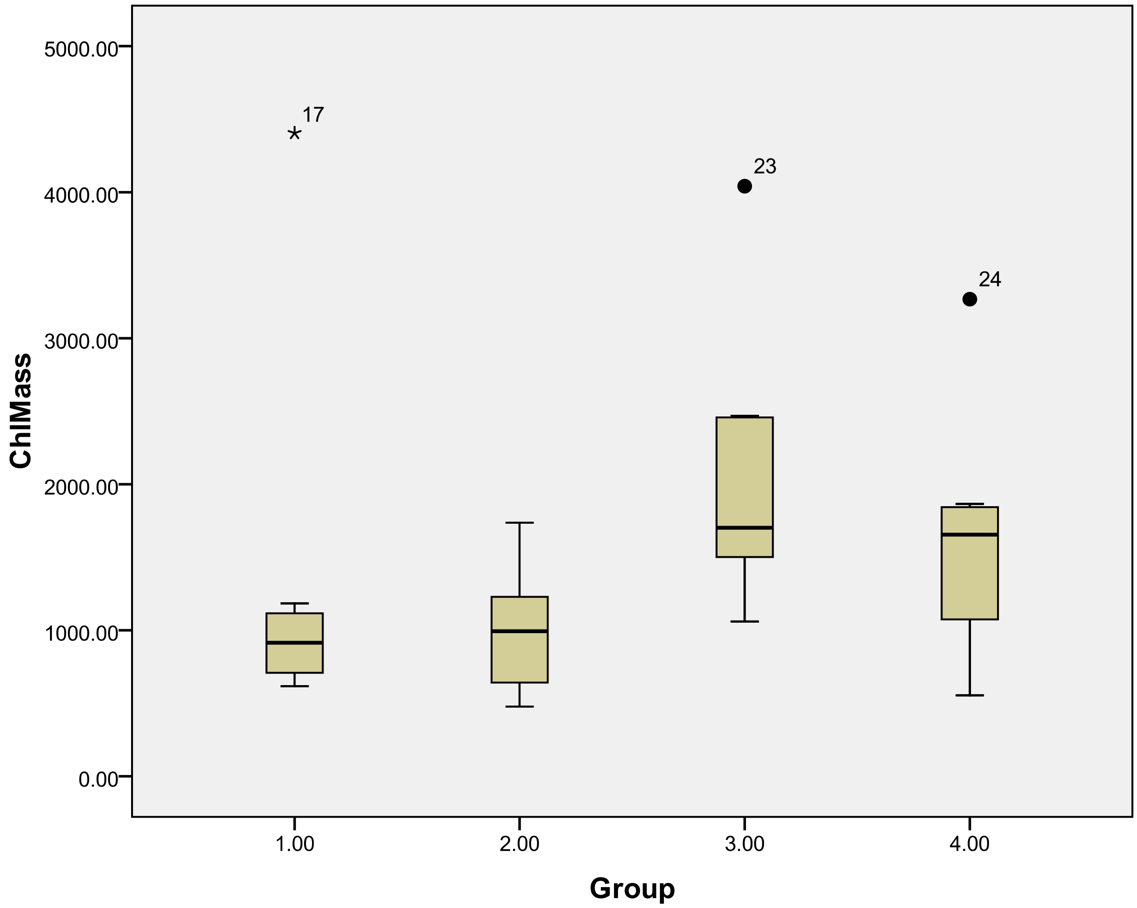
Table 4*: Mann-Whitney U test results for investigating differences in Chironomid colonization in all disturbed tiles with respect to lit and shaded conditions.*

Group	Count	Rank Sum
1	8	53.5
2	8	82.5
Mann-Whitney U		17.5
2-Tailed Significance		0.11
1-Trailed Significance		0.055

Table 5*: Mann-Whitney U test results for investigating differences in Chironomid colonization in all undisturbed tiles with respect to lit and shaded conditions.*

Test Statistics[b]	
	chironomid
Mann Whitney U	127.000
Wilcoxon W	263.000
Z	-0.050
Asymp. Sig.	0.484

Table 6*: Mann-Whitney U test results for investigating differences in Chironomid colonization in week-old tiles under lit and shaded conditions.*



Graph 1. ***Chlorophyll mass on tiles.*** *Group 1 = disturbed/light; group 2 = undisturbed/light, group 3 = disturbed/shade; group 4= undisturbed shade.*

LITERATURE CITED

Cain, Michael L., Bowman, William D., and Hacker, Sally D., 2008. *Ecology*. Sinauer Associates, Inc. Sunderland, MA.

De Cauwer, B., Reheul, D., De Laethauwer, S., Nijs, I., and Milbau A, 2006. "Effect of light and botanical species richness on insect diversity." *Agronomy for sustainable development* 26: 35-43.

Frouz, Jan, 1999. "Use of soil dwelling *Diptera* (*Insecta, Diptera*) as bioindicators: a review of ecological requirements and response to disturbance." *Agriculture, Ecosystems and Environment* 74: 167-86.

Hax, Carolyn L., and. Golladay, Stephan W., 1998. "Flow disturbance of macroinvertebrates inhabiting sediments and woody debris in a prairie stream." *The American Midland Naturalist* 139: 210-23.

Jonasson, P.M., 1972. "Ecology and production of the profundal benthos in relation to phytoplankton production in Lake Esrom." *Oikos Supplementum* 14: 1-148.

Pennak, R.W., 1978. *Fresh-water invertebrates of the United States.* John Wiley & Sons, Inc., New York, NY.

Pinder, L. C.V., 1986. "Biology of freshwater *Chironomidae*." *Chironomid Biology* 31: 1-23.

Real, Monttserrat and Prat, Narcis, 1992. "Factors influencing the distribution of Chironomids and oligochaetes in profundal areas of Spanish reservoirs." *Netherlands Journal of Aquatic Ecology* 26: 405-410.

Reice, Seth R., 1985. "Experimental disturbance and the maintenance of species diversity in a stream community." *Oecologia* 67: 90-97.

Seminara, M. and Bazzanti, M., 1988. “Trophic level assessment of profundal sediments of the artificial Lake Campotosto using midge larval community (Diptera: Chironomidae).” *Hydrobiological Bulletin* 22: 183-193.