Effects of Suspended Sediments on *Daphnia Magna*

Advanced Environmental Seminar
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Project Daphnia

Introduction:

The main purpose of the experiment was to find out if the suspended sediment problem in Lake Macatawa was hindering the invertebrates *Daphnia Magna*. The thought process of studying Daphnia was its potential to send a negative feedback through the lake's ecosystem. If Daphnia are the baseline food source and their populations are decreasing due to sediment loads, then there would be less food supply for bait fish which are used for game fishing. With the Daphnia being impacted by the sedimentation we believed it could have detrimental effects to future fishing in Holland. We modeled our experiment off of Neil A. Capper's work with the *Effects of Suspended Sediment on the Aquatic Organisms*. He studied the effects of different concentrations on aquatic organisms by using similar techniques that we implemented into our project.

Methods:

Water and Sediment Collection

Water and sediment samples were collected from Unity Bridge, Kollen Park, Pine Creek, and Paw Paw Park. Water samples were collected in 1 L plastic bottles attached to a sampling stick that was dipped into the body of water facing upstream. Sediment samples were collected using a sediment clam shell sampler as well as shoveling the top layer of the stream bed. Sediment was transported in clean five gallon buckets.

Once in the lab, Whatmen filter papers were weighed then used in a vacuum system. The water samples were poured through filter paper and vacuumed into a 1000 mL volumetric flask. The filter paper air-dried and was weighed to determine the total suspended solids. Each 1L water sample was filtered through three times. Sediment samples were dried in an oven at 100 ° Fahrenheit for 24 hours and then removed to cool. Samples were then weighed on an analytical balance (which read to the fourth decimal) and poured into a sieve stack. The sieve plate sizes used included: 2 mm, 1 mm, 500 µm, 250 µm, 125 µm, and 63 µm. Each sample was sieved for approximately two minutes. The characterized sediments were then further separated into plastic cups and individually weighed. After weighing the sediments, grain-size distribution charts (Figures 4-7) were created to visualize the percent finer than and analyze the sediments present at each location.

Suspended Sediment Settling Rate

The suspended sediment settling rate was determined for Pine Creek because it contained the highest levels of fine grained sediments. This pointed the experiment to learn more about the sediment, such as dropout rates since it was going to be chosen for future experimentation. To test the dropout rate, two vials were filled with 1.00 g of sediment and 4 mL of water. Both vials were shaken at time zero of the experiment followed by only one being shaken after a specific time. The settling rate of the Pine Creek sediment was observed over a 24 hour period and pictures were taken at one minute, five minute, ten minute, thirty minute, one hour, six hour, and twenty-four hour intervals. Once the test ran its course, it was concluded that the spin test that would be conducted with Daphnia later on in experimentation would need to be disturbed at least every hour for the majority of the sediments to remain suspended.

Daphnia with Sediment and No Stirring

About 500mL of spring water was added to three 1L beakers. 3.2 grams of kaolinite and montmorillonite were dumped in two beakers and allowed to settle before adding Daphnia. The third beaker was left with just water and deemed the control. After an hour with most of the sediment settled, ten Daphnia were added to each beaker by pipets which were new and clean for each trial. The beakers were then left alone for 15 hours. Once the trial period was over, the Daphnia were re-counted. In kaolinite and montmorillonite as well as the control, there was 100% survivability. It was then deemed acceptable for the next trials to start.

Daphnia with Suspended Sediment and Stirring

The Daphnia chamber was constructed by cutting about one inch by one inch holes on opposite sides of a 250 mL Nalgene plastic container. The holes were then covered with a Nitrix plastic 60 micron mesh and superglued in place. The containers were left to dry for 24 hours in order to cure before allowing contact with the Daphnia. The mesh was small enough to keep the Daphnia inside the chamber, but large enough to allow for the clay to circulate through during experiments. Cardboard sleeves were cut and placed around the neck of the bottles to keep the bottle suspended and eliminate contact with the stir bar at the bottom of the beaker during the experiment.

Results:

After tests 0.10 g - 6.00 g were run through the series twice, it was observed that the trials with more sediment (1.50 g - 6.00 g) were producing babies. This was specifically observed in the 3.00 g trials. It is thought this was caused by the new Daphnia kits that were used and. The first set of Daphnia kits could have been older and less robust, while the second kit could have been at the prime breeding age. This is complete speculation though as there was no true way to tell due to ordering them and not having raised them and watched the growth. Though the 15 hour test wasn’t long enough to allow fertilization and birthing, having the babies survive the conditions exemplifies the Daphnia’s resiliency. The tables and figures below illustrate that resiliency throughout the experiment. Figures 1-3 display the number of Daphnia alive after each 15 hour experiment with the indicated amount of sediment. Only the adults were calculated in because it was thought the babies would skew the data. All three figures show a dipsey do at 0.75 g and montmorillonite and Pine Creek continue with that dip until 3.00 g. Kaolinite and Pine Creek seem relatively unaffected once they hit the high levels of sedimentation while montmorillonite begins to show an increase in mortality. The grain-size distribution charts (Figures 4-7) show that most of the sites had a relatively even distribution. Paw Paw and Kollen Park had a wider range in distribution and Unity Bridge and Pine Creek contained more of the finer particles. Tables 1-3 display the number of living adult Daphnia after each trial. Table 4 displays the number of living adult Daphnia after being tested with contaminated sediments. Out of the normal sediments, only kaolinite was run through the trial series three times.

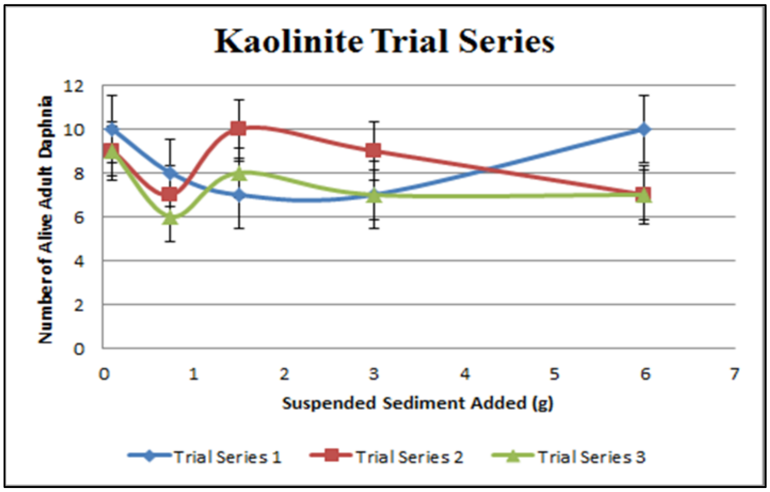


Figure 1: Number of living adult Daphnia corresponding to the total amount of kaolinite added.

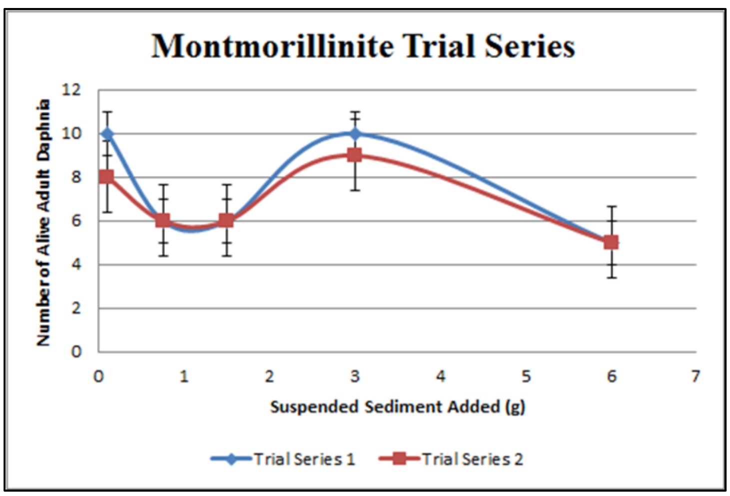


Figure 2: Number of living adult Daphnia corresponding to the total amount of montmorillonite added.

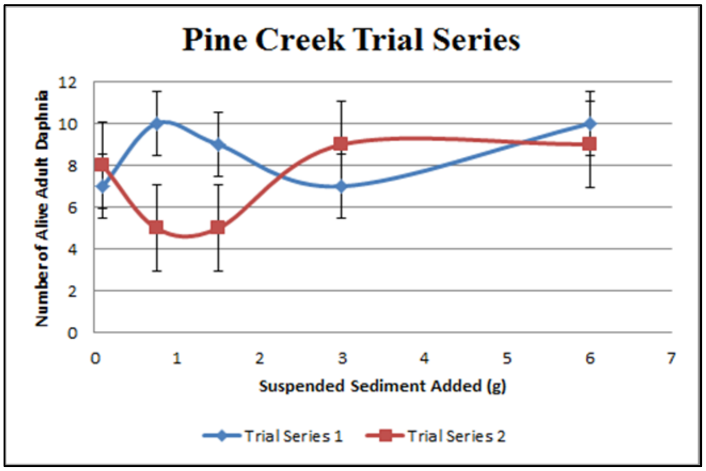


Figure 3: Number of living adult Daphnia corresponding to the total amount of Pine Creek sediment added.

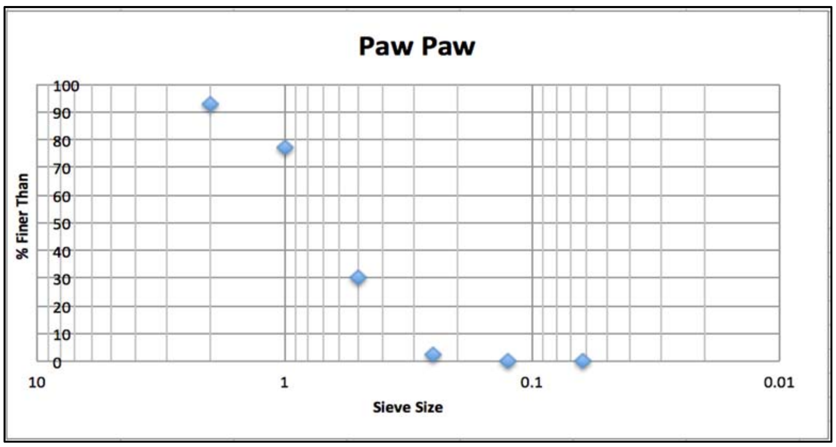


Figure 4: Grain-size distribution of Paw Paw Park sediments.

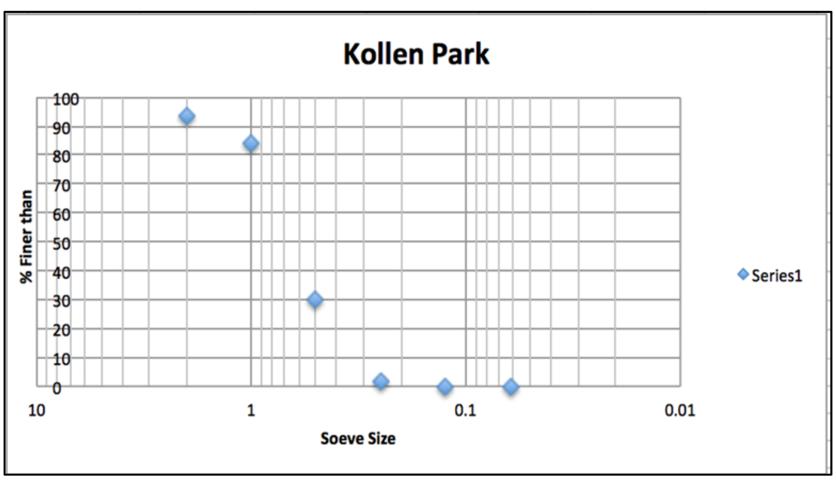
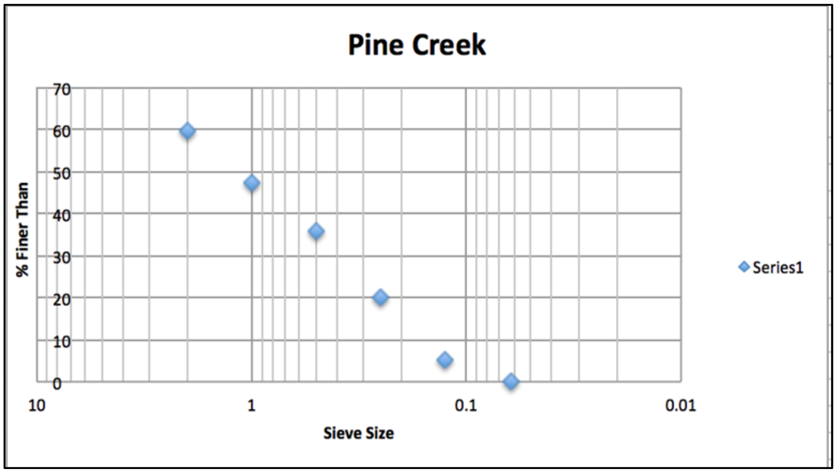


Figure 5: Grain-size distribution of Kollen Park sediments.



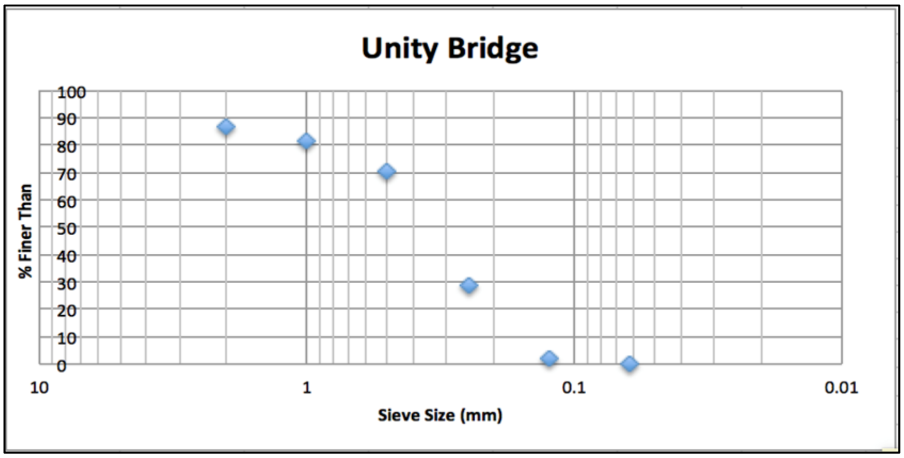


Figure 6: Grain-size distribution of Pine Creek sediments.

Figure 7: Grain-size distribution for Unity Bridge sediments.

0.10g/750mL	**Trial 1**	**Trial 2**	**Trial 3**
Montmorillonite	10	8	-
Kaolinite	10	9	9
Pine Creek	7	8	-

Table 1: Total living adult Daphnia of three trials with 0.10 g of sediment in 750 mL of spring water.

3.00 g/750mL	**Trial 1**	**Trial 2**	**Trial 3**
Montmorillonite	10	9	-

Kaolinite	7	9	7
Pine Creek	7	9	-

Table 2: Total living adult Daphnia of three trials with 3.00 g of sediment in 750 mL of spring water.

6.00 g/750mL	**Trial 1**	**Trial 2**	**Trial 3**
Montmorillonite	10	9	-
Kaolinite	10	7	7
Pine Creek	5	5	-

Table 3: Total living adult Daphnia of three trials with 3.00 g of sediment in 750 mL of spring water.

6.00 g/750mL	**Trial 1**	**Trial 2**	**Trial 3**
Purple Contaminant	9	10	-
Contaminated Pine Creek	8	6	6

Table 4: Total living adult Daphnia of three trials with 6.00 g of contaminated sediment in 750 mL of spring water.

Discussion:

Overall, the majority of grains in Kollen Park and Paw Paw Park were much larger than those collected from Pine Creek and Unity Bridge. Larger grain sizes had less of an effect than fine and clay-sized sediments on Daphnia mortality because clay-sized sediment was small enough to be ingested and clog the digestive tract. Daphnia survival rate in 0.10 g and 3.00 g of suspended montmorillonite sediment was much higher than in 6.00 g, where the survival rate was only 50%. 6.00 g of suspended montmorillonite sediment has a concentration that is than 0.14 g/L, which was the highest concentration of suspended sediment found in Lake Macatawa. Montmorillonite clay is also finer than most of the finest sediments found in Lake Macatawa. Thus, the increased mortality rate of Daphnia observed is likely due to the excessive amount of very fine sediment clogging the Daphnia digestive tracts. The unrealistic amount of fine suspended sediment used in the trials suggests the mortality of Daphnia would not be affected by the average suspended sediments found in Lake Macatawa or even high suspended sediment levels found after a large rain event. It was also found throughout the trials, below 3.00 g Daphnia had high mortality rate and at 3.00 g they seemed to recover, after that mortality increased again. The high mortality in Daphnia below 3.00 g was hypothesized to be because they did not have enough sediment to burrow in and became stressed in their environment. Because the Daphnia seemed to survive with 3.00 g, it is thought this is the preferred amount of sediment Daphnia need to live comfortably. Mortality rate then increased again because Daphnia became stressed and their digestive tracts clogged. Also, in each trial done with montmorillonite, kaolinite, and Pine Creek sediments, as the sediment amount increased the number of babies birthed increased, with the exceptions of 3.00 g of kaolinite and 6.00 g of Pine Creek sediments. It was observed that as sediment input increased, more sediment settled at the bottom of the Daphnia chamber. The increase in births may be due to the Daphnia preference to burrow and reproduce in more sediment.

Contaminated sediments were chosen due to the lack of strong trends in regular sediments. The contaminated sediments were used to prove our methodology could in fact kill Daphnia. The Daphnia trials done with 6.00 g of the purple contaminant showed Daphnia survival rate to be 95% and the Daphnia trials done with 6.00 g of the Pine Creek contaminant showed only a 66.6% survival rate. As a result, the purple aniline dye did not increase the mortality of Daphnia like originally thought and the heavy metals found in the Pine Creek contaminant had enough toxicity to significantly increase the mortality of Daphnia, but not so much as to rule out that death was just caused by the high concentration of suspended sediments.

Conclusion:

While the results were not exactly what were hoped for, it was observed that kaolinite and montmorillonite from about 0.10-1.50 g was somewhat lethal. Around 3.00 g it seemed as though it was a prime habitat for the Daphnia. They began to recover as well as have more babies. Once the concentration moved past 3.00 g, the mortality rates began to rise again. The most lethal dosage was at 6.00 g of sediment. The purple contaminated sediment and Pine Creek contaminated sediment proved neither were all that toxic. Pine Creek contaminated sediments did exhibit mortality, but no more so that the regular Pine Creek sediments at 6.00 g. This would point to that death was mainly caused by the concentration of the sediment and not the toxicity of it. During the experiments, the highest suspended sediment concentration after a rain event was around 0.14g/liter, indicating that there was no likely chance that the Daphnia would hit a concentration that would greatly affect them. With the sedimentation being so low it was concluded that the Daphnia would not create a negative feedback loop in the ecosystem. A likely issue that could cause problems for the game fish would be distorted sight due to murkiness of the waters, not necessarily the lack of food.