**Effects of anthropogenic influences on the Maple River using a biotic index and water chemistry analysis.**

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General Ecology

## ABSTRACT

Aquatic systems play an integral role in wide ranging ecological contexts by providing an outlet of nutrient and chemical buildup. Rivers are particularly vulnerable to deleterious anthropogenic effects given their popularity as transit and recreation waterways.  The health of a stream can accurately be assessed using an index of macroinvertebrate diversity as well as a battery of chemical concentration tests.  Our study found that the Maple River shows few negative effects of anthropogenic structures on macroinvertebrate health and on overall stream health.  The presence of a wetland may also have served to mitigate the possible human effects.

## INTRODUCTION

Human activities can impact aquatic systems in direct and indirect ways. In particular, anthropogenic disturbances such as agriculture and development can both directly and indirectly affect the stream ecosystem. (Maloney et al., 2011)  The geophysical extent of rivers and streams makes them a frequent recipient of human effects via fertilizer run-off and other non-point source pollution issues (Goodnight, 1973).  Commercial development and areas of land alteration are often linked to elevated concentrations of carbon, nitrogen, and phosphorus in a water bodies proximal to these unnatural disturbances (Dodds, 2006). Excessive concentrations of nitrogen in aquatic systems have been linked to extreme algal blooms leading to hypoxic conditions, which are damaging to other species and ecosystem health in general (Rabalais, 2002). Algal blooms cause many problems by releasing toxins and creating an absence of oxygen as dead algae decomposes (Carpenter, et al., 1998). Chemical composition changes of a water body can also have drastic effects on ecosystems. Certain aquatic invertebrates are particularly sensitive to

elevate chemical concentrations. Studying these macroinvertebrates to indicate water quality and the health of an ecosystem has become a common practice (Spieles, 2000). On the other hand, natural structures, as opposed to human ones, positively affect stream quality. Wetlands, for example, have been found to be efficient nitrate sinks. Less than one percent of nitrate entering wetlands eventually escape to downstream waters (Schindler, 1998). The sequestration properties of wetlands are critical to water bodies downstream of them.

The Maple River, located in northern Michigan, is known locally as a fishing spot and has a history of water quality research at UMBS. In 1981, UMBS student Matthew Kane conducted a water quality profile of the Maple River using a macroinvertebrate biotic index. Kane concluded substrate differences, current speed, and temperature differences affect macroinvertebrate diversity (Kane, 1981). Another study conducted in 1988 by UMBS student Monica Holley studied the effects of the Hidden River Golf Course on macroinvertebrate diversity. The golf course was not found to impact the stream in this study (Holley, 1998). Presently, this river functions as a site for fishing and recreation, and runs through a dam, a golf course, and a natural wetland.  In this study, we aim to quantify and compare the potential effects of these anthropogenic structures on water chemistry and macroinvertebrate diversity.

One method used in assessing water quality in the Maple River and the relative effects of the golf course and dam was to sample macroinvertebrate biota both before and after the anthropogenic structures.  A diversity index utilized was based upon the Hilsenhoff Biotic Index (HBI). A battery of water chemistry tests including pH, temperature, dissolved oxygen, nitrogen, and chloride concentration was also utilized as a criterion for relative stream health. By comparing these data from several sites on the Maple River, our aim is to determine the net impacts of anthropogenic structures on water quality and structure-specific impacts on water quality. We also are looking at how a natural structure, in this case a wetland, alters water quality downstream of anthropogenic structures.

## MATERIALS AND METHODS

*Study System*

The Maple River spans approximately 14.9 kilometers and is located in Northern Michigan in the city of Pellston. The East Branch of the Maple River begins in Douglas Lake

and flows to the dammed Lake Kathleen. The West Branch, however, is fed from groundwater and also empties into Lake Kathleen. Because of this, our study begins in Lake Kathleen to eliminate water source as a confounding variable. The area of study spans approximately 7.82 kilometers, beginning in Lake Kathleen and ending in Burt Lake. To calculate distance studied, the program ARCGIS 9.3 was used. Distance through the center of the river from the first to the last sample cite (ESRI, 2011). At the time of this study the percent dissolved oxygen was 93.74% (+/- 2.593) and the temperature was 17.28ºC (+/-1.061) across the Maple River.

Seven sites along the 7.82 kilometers were sampled in this study and chosen due to their proximity either prior to or following major anthropogenic impacts imposed on the Maple River. Sampling for the first site was done on the banks of Lake Kathleen approximately 180 meters from a dam. The lake had a sandy bottom and is slow moving. Also, the branches along the riverside appeared to be trimmed, a sign of additional human impact. The second site was approximately 25 meters past the dam. Water at this site was fast moving and the river bottom was rocky. Site three is approximately two kilometers downstream of the dam. This site was close to two small homes and appeared to be full of small fish and macroinvertebrates. Site four was just upstream of the Hidden River Golf Course and is a well-known trout fishing spot. The fifth site is approximately a kilometer downstream of the Hidden River Golf Course. Site six was very early in the wetland and was much more shallow than our other sites, and very sandy. The Michigan Department of Natural Resources has restricted vehicular use in this area in order to improve vegetation growth along the river and to reduce erosion. Our seventh and last site was Burt Lake. Sampling was done as close to the entrance of the Maple River into Burt Lake as possible. Many people were present here along with a multitude of boats, jet skis, and waterfront homes. There were many zebra mussels present at the site, as well.
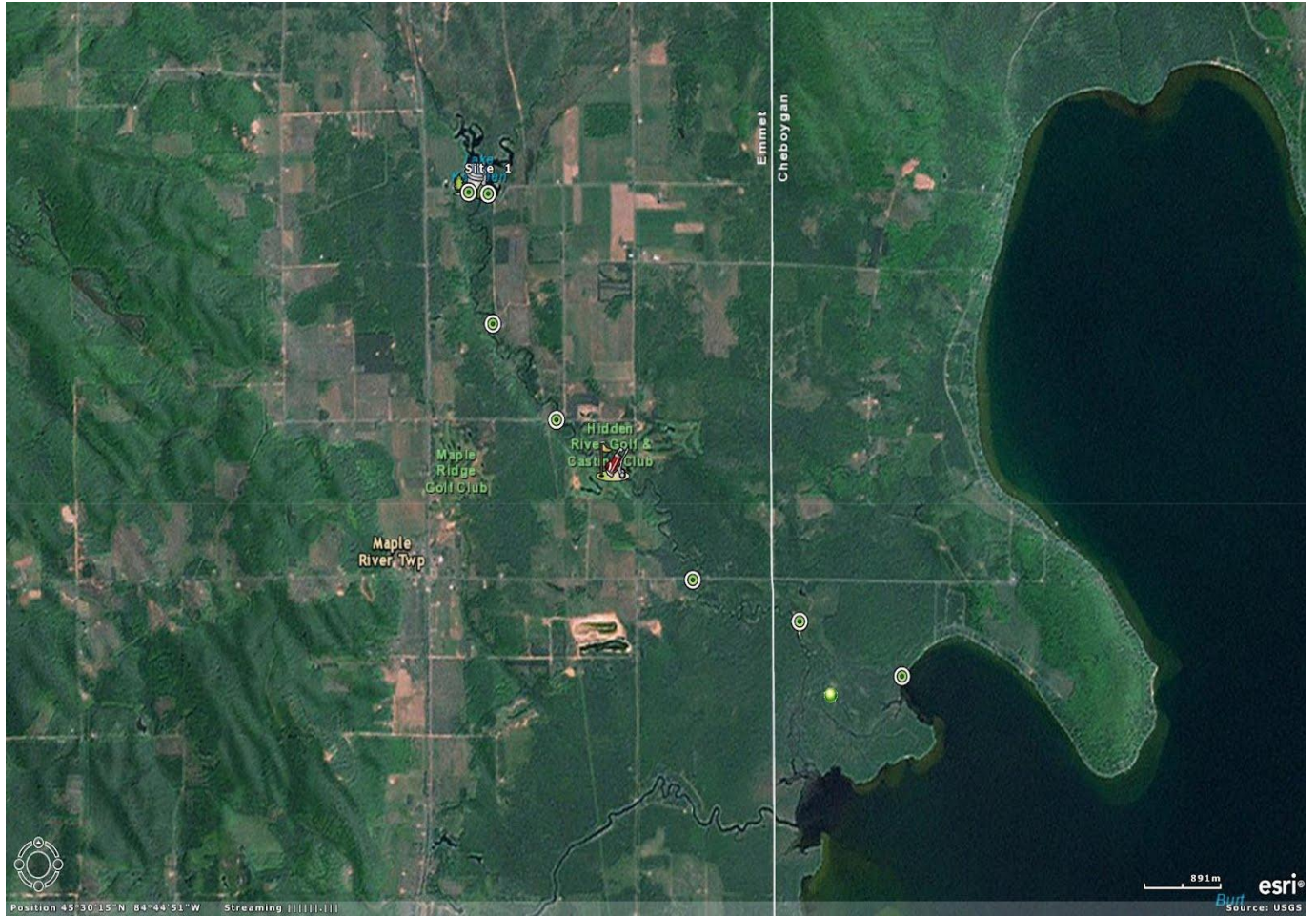
Picture 1. Maple River with marked testing sites.

*Sample Collection for Nutrient levels*

To determine water nutrient levels and pH, five water samples were collected at each site. The water samples were collected on the same day to reduce variance of the natural fluctuations seen in stream systems. The water samples were acquired using turkey basters and were stored in 250 mL sulfuric acid-washed bottles. An effort was made to keep the depth of these samples constant among each site. The samples were kept in a cooler until tested. The five samples were kept separate during testing.

*Sample Analysis for Nutrient levels*

Tyler Elias and Michael Grant tested water levels of pH, nitrate, and chloride in the University of Michigan Biological Station Lakeside Lab. Additional testing of the water was done on-site. Dissolved oxygen level (measured in mg/L) and temperature (measured in degrees Celsius) were measured using a dissolved oxygen probe.

To test pH level, a pH-testing probe was placed in a 15 mL portion of the original 250 mL water sample. In order to begin sampling, the pH probe must first be calibrated. In order to calibrate the probe, buffers of acidity 4, 7, and 10 were chosen. In between each calibration, the probe must be washed. After calibration, a standard curve was generated in which the water sample is compared to. Concentration is determined based on correlating values of the standard curve.

For chloride level testing, water samples must be filtered through a syringe to rid the sample of particulates that disrupt readings. From the original 250 mL water sample, 3 ml water samples were taken. These smaller samples were placed into cups washed with sulfuric acid. The samples were then put in to an auto flow analyzer which generated readings. Standard curves of chloride levels must be generated to serve as comparisons for the water samples. Water samples were then compared to the 5-point standard curve to generate chloride values.

To test the nitrate level, water samples were filtered and placed into the auto flow analyzer. Similarly to chloride testing, a 5-point standard curve was used to generate a standard curve for nitrate levels and concentrations.

*Sample Collection for Macroinvertebrate Diversity*

To determine macroinvertebrate diversity, three samples were taken at each site using Surber samplers. The Surber samplers were placed parallel and equidistant from one another across the river width. The samplers provide three .305 m x .305 m sample areas. To use the Surber sampler, the attached metal rectangle is placed on the river bottom and the net is extended in the direction of water flow. The sediment inside of the metal rectangle must be disturbed to catch organisms of interest. To do so, sediment was "kneaded" for ninety seconds at a depth of approximately 5 cm. This sample was then poured into a sifter and filtered with the river water.

Macroinvertebrates were removed from the sifter using tweezers and placed into a bottle containing 70% ethanol to prevent decomposition. These samples were then taken to the lab for identification. The three samples from each Surber sampler were kept separate.

*Sample Analysis for Macroinvertebrates Diversity*

The identification of macroinvertebrates was done using River Murray Waterwatch's Aquatic Invertebrate Key (RMCWMB, 2004). By using microscopes and magnifying glasses, organisms were identified and tallied for subsequent data analysis. To determine macroinvertebrate diversity and their relationship with water quality, the Hilsenhoff Biotic Index (HBI), a biotic index of arthropod fauna that evaluate stream pollution, was used (Hilsenhoff, 1987). Arthropod families found in streams of the Great Lakes region were each assigned with a "biotic index" that range from 0–10. This number indicates their tolerance for organic and nutrient pollution. Numbers closest to 0 are species most intolerant to pollution and species with numbers assigned closer to 10 are those most tolerant to pollution (Hilsenhoff, 1987). Since we have limited knowledge in the identification of macroinvertebrates down to family level, biotic numbers of each order were averaged. For example, we were able to identify Odonata in our samples, but unable to be more specific than this. An average for all Odonata families was calculated, and this value was used in place of a specific index value. This method was used for the Plecoptera, Odonota, Ephemeroptera, Trichopera, Amphipoda, and Coleoptera orders.  We were able to identify our "Midge", "Horsefly", and "Cranefly" macroinvertebrate samples down to the family level. These three macroinvertebrate families were quite distinctive, therefore easy to categorize by family. The HBI value for these three families are not averages which explains why they are whole numbers.

| Common Name (Order/Family Name) | HBI Values for Order/Family |
|---|---|
| "Stonefly"                (Order name: *Plecoptera*) | 1.14          (Average Order HBI Value) |
| "Odonata"                 (Order name: *Odonata*) | 5.26          (Average Order HBI Value) |
| "Mayfly"             (Order name: *Ephemeroptera*) | 3.36          (Average Order HBI Value) |
| "Caddisfly"               (Order name: *Trichoptera*) | 2.85          (Average Order HBI Value) |
| "Scud"                  (Order name: *Amphipoda*) | 7.33          (Average Order HBI Value) |
| "Scavenger Beetle"      (Order name: *Coleoptera*) | 4.81          (Average Order HBI Value) |
| "Horsefly"              (Family name: *Tabanidae*) | 5                    (Family HBI Value) |
| "Midge"              (Family name: *Chironomidae*) | 10                   (Family HBI Value) |
| "Cranefly"             (Family name: *Tipulidae*) | 4                    (Family HBI Value) |

**Table 1**. HBI Values used in data analysis. Values derived and adapted from Hilsenhoff's *Great Lake's Entemologist,* 1987.

*Statistical Analysis of Data*

Using IBM SPSS Statistics 19, multiple Independent-Sample t-tests and ANOVA tests were run to analyze the data for significance. Independent-Sample t-tests are used to compare the means of two different populations. Here they are used to compare the mean values of water chemistry (pH, nitrate, chloride) across barriers. Independent sample t-tests were also performed to compare mean Hilsenhoff Biotic Index (HBI) values across paired sites. No analysis was performed to compare the wetland's HBI to Burt Lake's because no macroinvertebrate were found within the Burt Lake samples. An ANOVA test was used to then compare the water chemistry, HBI values and EPT percentages for the "after" sites to see if they have different effects on the river (except for the HBI of Burt lake). A Scheffe Post Hoc test was included and indicated between which sites the significance occurred. In order to test if the each barrier had the same effect on water chemistry, HBI and EPT percentages we calculated a T-statistic. This equation gives a T-value, which was then input into Graphpad QuickCalc's website, along with the appropriate degrees of freedom, to find the p-value (Motulsky, 1999). The p-value given for each comparison will indicate if the difference between means is significant. Using an alpha

value of 0.05, a p-value of less than 0.05 is considered significant and p-values between 0.05 and 0.10 are considered marginally significant.

**RESULTS**

*Statistical Analysis of Data*

Table 2 displays the average pH, nitrate, chlorine levels, and HBI per site. The independent-sample t-tests performed to test the water chemistry across structures yielded significant differences in pH between the golf course barrier (t=-2.346, p=0.047) and the wetland (t=-10.042, p=0.000), while no significance was seen across the dam (t=1.188, p=0.269). Significance for nitrate levels across barriers was significant for all three pairs (p<0.05). Figure 2 shows that the nitrate levels in Burt Lake were much lower compared to the nitrate levels in the wetland. Chloride levels on the other hand, were only significant across the wetland (t=-39.497, p=0.000). Figure 3 illustrates that there is a marginally significant difference across the golf course (t=2.348, p=0.077). (see Independent Sample T-test Table #s) The figures below better illustrate the relationship of water chemistry across barriers (Figures 1, 2, and 3).

When comparing the relative effects of barriers on water chemistry, all relationships produced a significant difference (p<0.05) except for the effect on chlorine levels by the dam compared to the golf course (t=-1.9583, p=0.0859), which was marginally significant.

The ANOVA tests done to compare if each structure had a different effect on the river yielded a significant difference (p=0.000) between the three sites for pH and nitrate. Chlorine levels were only significant between barriers 1 and 3 (sites 2 and 7) and barriers 2 and 3 (sites 5 and 7) (p=0.000). There was no significant difference between 1 and 2 (sites 2 and 5) for their effect on chlorine levels (p=0.139).

The independent sample t-tests comparing HBI values across structures indicated no significance with all p-values greater than 0.200. The ANOVA comparing HBI after the structures yielded a p-value of 0.082 between barriers 1 and 2 (sites 2 and 7), meaning no significant difference existed. When testing to see if barriers had the same effect, all relationships were not significant (p>0.19).

The independent sample t-tests of percent EPT produced results similar to the tests of HBI. No significant difference was seen across barriers (p > 0.180), and no significant difference was seen when comparing the effect of each barrier on the river (p=0.946). The t-statistical test evaluating if the structures had the same effect on percent EPT were also all not significant (p>0.07).

**DISCUSSION**

In regards to our initial hypotheses, we were able to make solid conclusions based on our data.  We did not find significant differences in HBI index values between any of our sites. Therefore, we fail to reject our null hypothesis that anthropogenic structures have no effect on water quality based on the HBI index values. For our second hypothesis, we were able to reject our null hypothesis that anthropogenic structures have no effect on water quality based on nitrate, dissolved oxygen, chloride, and pH values. The only values we were able to discount from this hypothesis were in dissolved oxygen. Levels of dissolved oxygen did not show a significant difference among any sites tested.

For levels of nitrate, there was significantly less nitrate after the wetland as opposed to in the wetland.  Levels of nitrate were significantly higher after the golf course. Finally, levels of nitrate were significantly lower following the dam site. Differences in chloride levels were not found to be significant in the sites before and after the dam. Chloride levels were significantly lower following the golf course. The significance for chloride before and after the wetland was tested but discounted because we were only interested in nitrate levels at this site. Levels for pH were significantly higher both after the golf course and after the wetland. However, we must discount the wetland data because we only interested in nitrate levels at this site.

Results found in this study corroborate the expectation that Maple River is a healthy stream ecosystem, which was expected based on both past research and through conversing with

locals. We have drawn this final conclusion because our data ultimately was focused on the macroinvertebrate data. Since water chemistry fluctuates drastically day to day, we are unable to draw conclusions about stream quality based on this data alone.  Macroinvertebrate diversity is more stable, and so we are able to conclude that the river is quite healthy because we did not find significant differences among our sites in HBI index values. We are also able to come to the conclusion that wetlands are effective nitrate sinks, because the data indicated significantly lower nitrate levels after the wetland. Although our data did not support expected nitrate percentage absorbed, the absorbed amount was still significant and quite interesting.

Although we found our river system to be quite healthy, Burt Lake was quite a different story. Burt Lake displayed data of a degraded and human-impacted ecosystem. Macroinvertebrates were unable to be tested at this site, because no testable specimens were collected. The only macroinvertebrates collected during sampling were those that were not included in the HBI calculations, including a variety of shells.

After examining or research and results, we were astounded by how drastic the changes from the site in the wetland to Burt Lake. We have concluded that this is due to the high human traffic of the area.

Human activities have and will continue to negatively impact ecosystems in a variety of ways. It is our duty as ecologists to quantify and examine these effects in order to better understand them and to inform the public as to how best to ameliorate or prevent these byproducts of our modern society. Given the staggering breadth of the anthropogenically compromised environmental facets of our planet, from excessive carbon emissions to the deforestation of tropical rain forests, depletion of fisheries worldwide, and the literal tons of discarded plastics swirling in the Pacific Ocean, the best way to make a positive contribution to the causes of environmental conservation and protection and to raise public awareness of these causes is to focus on a particular ecosystem and issue, and carefully determine and report the natural and developing data of that area.

*Possible Sources of Error*

The largest source of error for this project is the last sample site, Burt Lake. This site was used to determine the effects of the wetland on nitrate levels. As stated in the introduction, it has

been previously concluded that wetlands allow a mere 1% of nitrogen to escape downstream (Schindler, 1998). In our study, however, 15.4% of nitrogen remained in the stream after the wetland. This difference could be attributed to the human traffic of Burt Lake.  Sampling was also limited due to the high density of water plants of the area. We are unable to draw any conclusions about differences in other chemicals or macroinvertebrate diversity from this site. Amount of human traffic could lead to few macroinvertebrates numbers and questionable and unknown chemical dumping in the area.

Early on in the study, a concern in the change in phosphorus levels was noted.  However, due to the properties of naturally occurring phosphorus, collection was not possible. Michael Grant was able to inform us that phosphorus often clings to particles such as rocks and debris in the river leading to inaccurate data. Because of this, phosphorus was not tested and nitrogen levels alone were used to test for possible effects of fertilizer run off.

Accompanying data from each test site such as stream width and depth as well as the flow rate could have helped to preclude the possibility of those variables confounding the results. Similarly, multiple readings for dissolved oxygen and temperature as well as additional macroinvertebrate collections would increase the precision of the data. Finally, an ideal design would take into account the stochastic effect of precipitation on chemical concentrations in the river by averaging chemical samples taken over the course of several weeks.  Testing multiple rivers with similar anthropogenic structures would also have enriched the data. Similarly, identifying the macroinvertebrates to the species level would have increased the accuracy of our study.

**ACKNOWLEDGEMENTS**

subject of General Ecology and aid in research. This project would not have been possible without outside assistance, and we would to extend our sincerest thanks to those listed above.

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**APPENDIX**

| Common Name (Order/Family Name) | HBI Values for Order/Family |
|---|---|
| "Stonefly" (Order name: *Plecoptera*) | 1.14 (Average Order HBI Value) |
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**Table 1**. HBI Values used in data analysis. Values derived and adapted from Hilsenhoff's *Great Lake's Entemologist,* 1987.

| Site | Nitrate (µg N/L) | pH | Chloride (mg Cl/L) | HBI |
|---|---|---|---|---|
| 1 | 153.3 (S.E) | 8.00 | 7.0 | 6.08 |
| 2 | 134.0 | 7.97 | 7.0 | 5.656 |
| 3 | 151.6 | 8.03 | 7.0 | 4.16 |
| 4 | 220.7 | 8.04 | 7.2 | 4.077 |
| 5 | 244.3 | 8.09 | 6.8 | 6.26 |
| 6 | 236.0 | 8.07 | 7.0 | 4.566 |
| 7 | 35.4 | 8.26 | 10.6 | N.A. |

**Table 2**. Water sample results by site. Nitrate, pH, HBI, and chloride are averages among five samples.
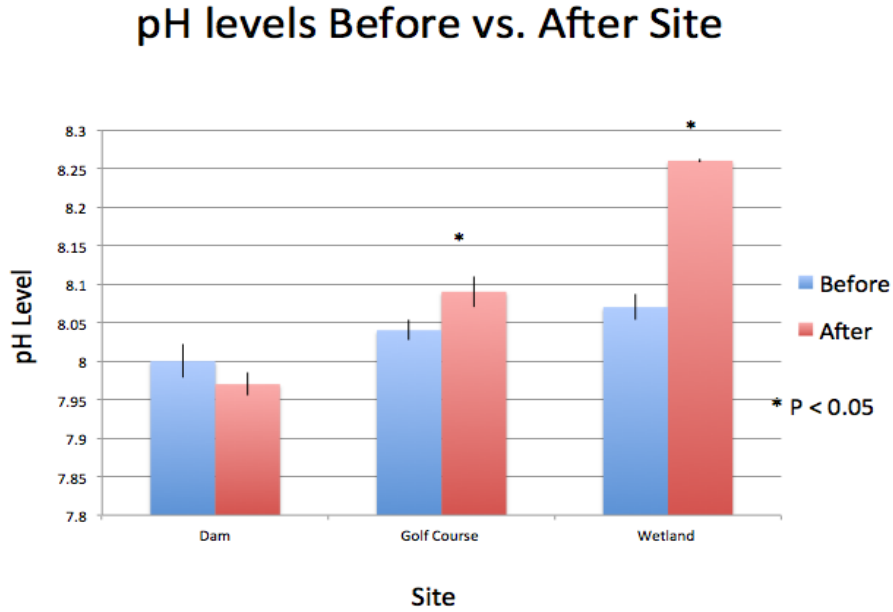
**Figure 1. Mean pH levels across bars. Bars represent standard error bars. In this case, n=5. Asterisk represents significance between groups. The alpha value is .05. P value found using an independent t-test.**
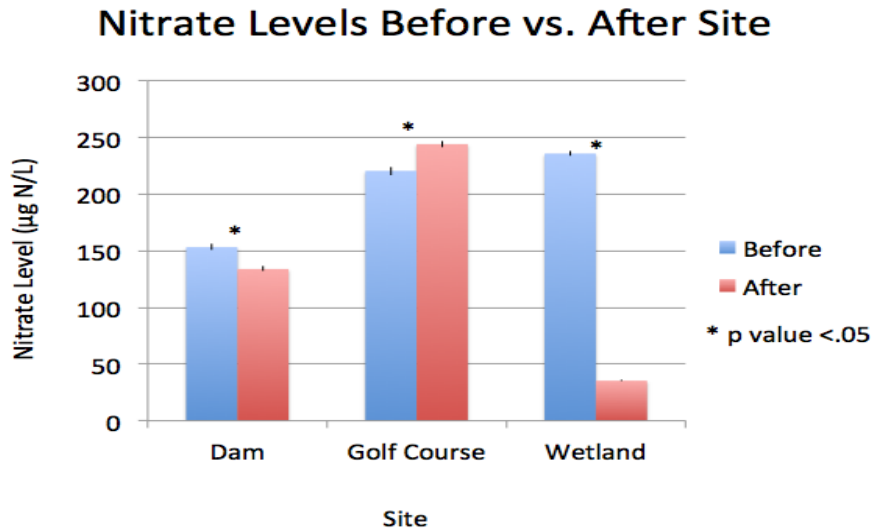


**Figure 2. Mean nitrate levels across bars. Bars represent standard error bars. In this case, n=5. Asterisk represents significance between groups. The alpha value is .05. P value found using an independent t-test.**

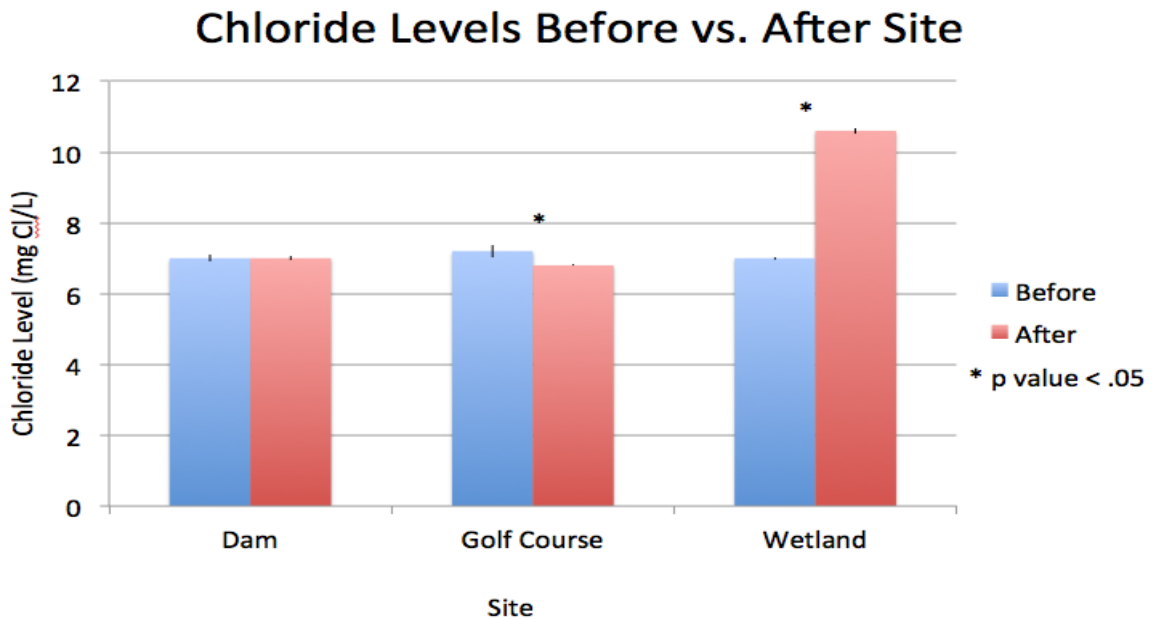## Chloride Levels Before vs. After Site



**Figure 3. Mean Chloride levels across bars. Bars represent standard error bars. In this case, n=5. Asterisk represents significance between groups. The alpha value is .05. P value found using an independent t-test.**
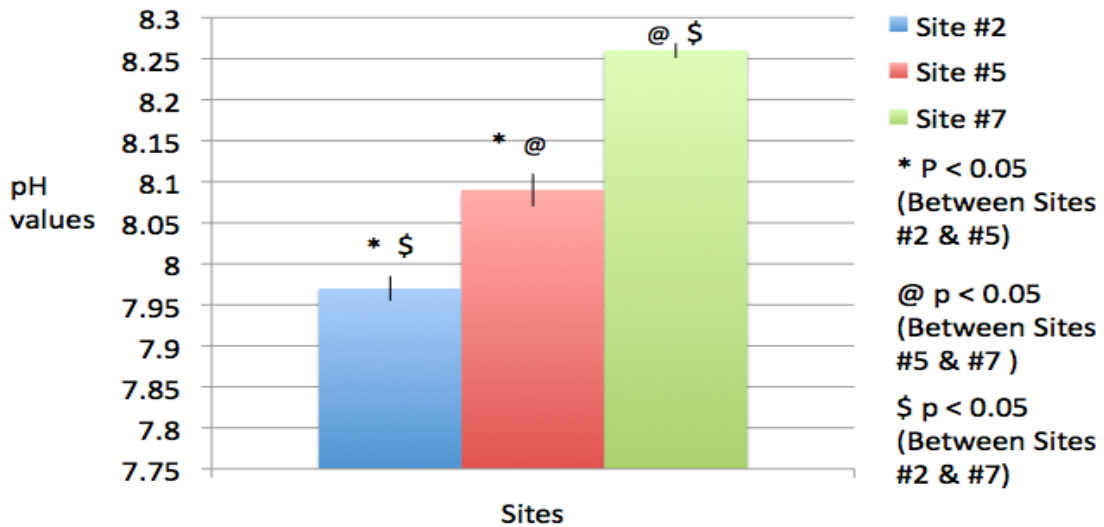
## pH Levels Between Sites After Structures



**Figure 4. Comparison of pH after structures. Bars represent standard error bars. Asterisk represents significance between groups. The alpha value is .05. P value found using an ANOVA**

**Figure 5. Comparison of Nitrate after structures. Bars represent standard error bars. Asterisk represents significance between groups. The alpha value is .05. P value found using an ANOVA**