Implications on Distribution and Abundance of Benthic Macroinvertebrates in the Maple River Based on Water Quality and Habitat Type

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Abstract

Macroinvertebrate communities are often diverse and variable in richness and abundance depending on water quality in aquatic ecosystems. Measures of water quality (e.g. alkalinity, pH, conductivity, salinity, macronutrient content) and habitat forms can vary in different locations and affect the abundance of macroinvertebrates there. Riffles (flowing water) and pools (standing water) form in streams and provide different habitats for macroinvertebrates. To test the effects water quality and habitat have on macroinvertebrate richness and abundance, three sites along the Maple River in Pellston, MI were sampled. In respect to water quality, we observed large differences between the East and West Branch locations, mainly involving higher levels of nitrate, total nitrogen, and conductivity from the West Branch located downstream from the Pellston Airport and multiple farming areas. Along with testing water quality at each site, macroinvertebrates distribution and diversity was analyzed to show a decrease in diversity among the West Branch as compared to the East Branch. Our findings suggest that pollution from anthropogenic sources impacts water quality of nearby streams and rivers, resulting in the decrease of macroinvertebrate diversity.

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Macroinvertebrate communities are often diverse and variable in richness and abundance depending on water quality in aquatic ecosystems. Measures of water quality (e.g. alkalinity, conductivity, salinity, macronutrient content) and habitat forms can vary in different locations and affect the abundance of macroinvertebrates there. Riffles (flowing water) and pools (standing water) form in streams and provide different habitats for macroinvertebrates. To test the effects that water quality and habitat have on macroinvertebrate richness and abundance, three sites along the Maple River in Pellston, MI were sampled. In respect to water quality, we observed large differences between the East and West Branch locations, mainly involving higher levels of nitrate, total nitrogen, and conductivity from the West Branch located downstream from the Pellston Airport and multiple farming areas. Along with testing water quality at each site, macroinvertebrates distribution and diversity were analyzed to show a decrease in diversity among the West Branch as compared to the East Branch. Our findings suggest that pollution from anthropogenic sources impacts water quality of nearby streams and rivers, resulting in the decrease of macroinvertebrate diversity.

Introduction

Rivers provide suitable habitats for diverse species of macroinvertebrates. The distribution and abundance of these macroinvertebrates can be assessed to determine the water quality of streams because certain macroinvertebrates can tolerate more extreme conditions (e.g. acidity and conductivity) than others (Lytwynec, 2003). The measures of water quality, including alkalinity, pH, macronutrient content, conductivity, and temperature, as well as habitat forms, can vary along the river and change the composition of macroinvertebrates found there. Conversely, the presence of certain macroinvertebrates can be an indicator of river system health. Specifically, the aquatic insect orders *Ephemeroptera* (mayflies), *Plecoptera* (stoneflies), and *Trichoptera* (caddisflies) can be studied, using the EPT Index, to determine water quality. The EPT Index is based on the premise that high-quality streams usually have the greatest species richness (EPT Index, 2015). Thus, we would expect a polluted river to have fewer species, especially because many benthic macroinvertebrates are not tolerant of pollutants (EPT Index, 2015). Water quality can be affected by natural sources such as groundwater springs, marshes, and precipitation in addition to anthropogenic point sources (drainage pipes) and nonpoint sources (fertilizer runoff from golf courses or farms).

The Maple River, located in Northern Michigan, is a primarily groundwater fed river by two branches (the East and West) that join together south of Pellston near a dam. The substrates of this region consist of sand and gravel that originated from glaciers (Heinen and Vande Kopple, 2003). The catchments for the East, West, and main branch of the river have coarse textured glacial till, which promotes groundwater movement in the underlying geology (Godby, 2014). Given the constant addition of groundwater from numerous springs along the river, pollutants that enter the water are likely to be diluted, which tends increase the level of water quality. The Maple River is a popular location for recreationalists to hike, kayak, or canoe. It is also known in the area as one of the best places to fish for trout, which prefer cold, groundwater fed rivers. In addition, the Maple River has been used for research purposes by the University of Michigan Biological Station (UMBS) for water quality since 1909. The UMBS now has a permanent stream lab that can manipulate variables (e.g. substrate, flow rate, water chemistry, shading) to study their effects on aquatic organisms in the Maple River (Heinen and Vande Kopple, 2003).

In this study, we explore whether macroinvertebrate distribution and abundance are affected by water quality depending on location along the Maple River. Our first hypothesis is that there will be higher species richness and abundance of macroinvertebrates at the farthest downstream location, being the Dam site rather than the East or West Branch. Rivers contain more biodiversity downstream due to the accumulation of nutrients and sediments. Temporal pattern of sediment addition to a river is positively correlated with flow volume (Becker, 172). Since the Dam site is located at the point where multiple water sources converge, the flow volume will be the highest, increasing the amount of sediment to the Dam site. When comparing only the East and West Branch, we hypothesize that the West Branch will have less biodiversity and abundance of macroinvertebrates because the West Branch site is located next to the town of Pellston and runs parallel to the Pellston Airport, making it more susceptible to anthropogenic pollutants. The East Branch runs primarily through the UMBS, which is a biosphere reserve that contains ecosystems worthy of conservation (Heinen and Vande Kopple, 2003), limiting the amount of pollution to this branch as compared to the West Branch. Many aquatic insects are intolerant of pollutants so the greater the pollution, the lower the species richness (EPT Index, 2015). We assume that the West Branch pollutants will become diluted by groundwater from the many springs along the river before reaching the Dam site, deeming this variable insignificant to macroinvertebrate distribution and abundance downstream. Therefore, more macroinvertebrates will be found in the East Branch site, which we observe to be a stream of better water quality as compared to the West Branch. Our second hypothesis is that more species will be located in pools as opposed to riffles. Pools have a lower velocity of water flow, allowing macroinvertebrates to inhabit these areas more easily as opposed to fast moving riffles. Lastly, we hypothesize that water quality downstream will have higher levels of conductivity, macronutrients, pH levels, and alkalinity. As the West and East Branch flow downstream and meet at the Dam site, water quality should decrease due to the accumulation of pollutants, sediment and nutrients.

Methods

Site Selection

We collected samples from three sites of the Maple River. Site 1 was located below the dam next to the Damsite Inn, Site 2 was located on the West Branch of the Maple River where it crosses US 31, and Site 3 was located on the East Branch of the river where it crosses Riggsville Road. We focused on these sites due to their accessibility and potential differences in water chemistry. Site 1 (Dam) was the furthest downstream, and potentially contains more biodiversity due to accumulation of sediments and nutrients (Angelier, 2003). Site 1 is also the point of intersection for the East and West branches of the Maple River. Site 2 and Site 3 were chosen for comparison because the branches run through different natural and anthropogenic landscape features (e.g. wetland and farmland) and may have different water chemistries and macroinvertebrate compositions. Water quality at the Site 2 and 3 is presumably affected by different factors. Site 2 may have fertilizer and other runoff from the airport and farmland, and Site 3 may have less runoff due to the fact that it runs through less human impacted areas. Additionally, Site 2 runs closer to the town of Pellston than the Site 3. Lastly, all sites have pool and riffle habitats.

Sampling Technique - Water Chemistry

We collected water samples from three sites along the Maple River. We took samples from Site 1 first, then Site 2, then Site 3 in order to avoid potentially contaminating downriver sites by disturbing the substrate upriver. We placed a transect across the river at each site and took water samples at five equally spaced intervals (9 ft. intervals at Site 1, 7 ft. at Site 2, and 6 ft. at Site 3) for a total of 15 water samples in total. The syringes used to fill the sample bottles were rinsed with deionized water three times and then fitted with a filter system. Each filter was removed from the syringe after each use and replaced using tweezers so the filters did not come in contact with any contaminates. We tested the pH level of each sample using pH strips. The water samples were then taken to the chemistry lab and analyzed for micronutrient composition and alkalinity (the amount of acid needed to bring the sample back to a pH of 4.2). We performed conductivity (the degree to which water conducts a specific electricity), temperature, and flow rate tests at the sites. Five flow rate readings were taken at each site at equally spaced distances across the transect and averaged together. Three conductivity and temperature readings were taken at each site, one at each shore and one in the middle of the stream. These readings were then averaged together.

Sampling Technique - Macroinvertebrate

Two riffle habitat macroinvertebrate composition samples and two pool habitat macroinvertebrate composition samples were taken at each site. We placed D-nets with the openings facing upstream, stood one foot back from the nets, and dug our feet into the sediment five to ten centimeters to kick up any macroinvertebrates on the bottom of the river or burrowed in the substrate. If the habitat was near a log or rock, we ran our feet over the rocks to knock any potential macroinvertebrates free and into our nets. We repeated this process three times at each riffle and pool for a total of thirty-six individual collection attempts. The contents of the nets were then dumped into a tray on shore and sorted through by adding approximately 15 mL of water and looking for movement. Forceps were used to pick out the macroinvertebrates from the substrate and place them in vials with an inch of 70% by concentration ethanol solution. The macroinvertebrates were taken back to the lab and identified to order and family using Merritt's key (1978). We counted the number of different macroinvertebrates from each sample to determine abundance and species richness differences at each site, and more specifically in riffle habitats versus pool habitats.

Statistical Methods

To test for independence and correlations between macroinvertebrates living in riffles and pools, we conducted a chi-squares analysis. We also performed a Biotic Index Calculation to determine whether the river was polluted or clean based solely on macroinvertebrate composition. The index categorizes different macroinvertebrate orders as Class I (Pollution Sensitive Taxa), Class II (Moderately Tolerant Taxa), or Class III (Pollution Tolerant Taxa). To calculate a river's biotic index, we multiplied the number of unique species found from Class I by two, and add that to the number of unique species found from Class II (Sharpe et al., 2015). If the resulting number is ten or greater, the river is considered clean with minimal or no pollution. The following is the equation for the Biotic Index Calculation:

$$BI = 2(n\ Class\ I) + (n\ Class\ II)$$

Where the variable "n" is the number of taxa (different organisms based on appearance).

Results

We found that Site 1 had the greatest aquatic macroinvertebrate abundance and second most richness, with 131 macroinvertebrates collected from eight different orders (Table 1). One hundred twenty macroinvertebrates from seven orders (Table 2) were collected at Site 2, and 64 macroinvertebrates from ten orders (Table 3) were collected at Site 3.

Table 1. Collection Totals from the Site 1 (Dam)

Dam Riffle 1	Dam Riffle 2	Dam Pool 1	Dam Pool 2
1 Odonata gomphidae 1 Oligochaeta 4 Hirudinea 1 Ephemeroptera leptophlebiidae 1 Ephemeroptera arthroplediae 3 Trichoptera limnephilidae 16 Diptera 2 Amphipoda	4 Oligochaeta 2 Hirudinea 2 Decapoda 1 Coleoptera 2 Odonata gomphidae 4 Odonata cordulegastridae 4 Trichoptera limnephilidae 3 Amphipoda 3 Isopoda 3 Ephemeroptera heptageniidae	3 Diptera 1 Odonata cordulegastridae 1 Hirudinea 2 Isopoda 1 Coleoptera 7 Amphipoda 2 Trichoptera limnephilidae 1 Trichoptera ecnomidae 6 Ephemeroptera ephemeridae	10 Amphipoda 7 Isopoda 33 Diptera

Table 1. shows the total collection of macroinvertebrates collected at Site 1, the Dam site. There were a total of 131 macroinvertebrates from 8 orders collected. The number of macroinvertebrates found in the riffles was 57 from 13 different species, and 74 in pools from 10 species.

Table 2. Collections Totals from the Site 2 (West Branch)

W.B. Riffle 1	W.B. Riffle 2	W.B. Pool 1	W.B. Pool 2
2 Ephemeroptera leptophlebiidae 1 Diptera 1 Trichoptera hydropsychidae	22 Trichoptera brachicentridae 21 Trichoptera hydropsychidae 4 Diptera 33 Ephemeroptera leptophlebiidae 3 Ephemeroptera arthropleidae 1 Ephemeroptera baescidae 2 Plecoptera peltoperlidae 1 Plecoptera capniidae	2 Trichoptera limnephilidae 1 Ephemeroptera ephemeridae 1 Ephermeroptera baescidae 6 Diptera 2 Isopoda	2 Trichoptera limnephilidae 1 Oligochaeta 1 Odonata petaluridae 3 Isopoda 7 Diptera

Table 2. shows the total collection of macroinvertebrates collected at Site 2, the West Branch site. There were a total of 120 macroinvertebrates from seven orders collected. The number of macroinvertebrates found in the riffles was 94 from nine different species, and 26 in pools from seven species.

Table 3. Collections Totals from Site 3 (East Branch)

E. B. Riffle 1	E. B. Riffle 2	E.B. Pool 1	E.B. Pool 2
3 Trichoptera limnephildae 1 Amphipoda 2 Coleoptera 1 Megaloptera carydalidae	2 Odonata cordulegastridae 5 Amphipoda 2 Plecoptera 3 Diptera 1 Decapoda	8 Trichoptera limnephilidae 2 Odonata calopterygidae 1 Plecoptera perlidae 15 Amphipoda 1 Odonata cordulegastridae 1 Odonata petaluridae 1 Ephemeroptera	4 Trichoptera limnephilidae 2 Odonata gomphidae 2 Odonata calopterygidae 2 Amphipoda 1 Ephemeroptera ephemeridae 1 Megaloptera sailidae

		arthropleidae	1 Hemiptera notohectidae 2 Diptera 1 Coleoptera

Table 3. shows the total collection of macroinvertebrates collected at Site 3, the East Branch site. There were a total of 64 macroinvertebrates from ten orders collected. The number of macroinvertebrates found in the riffles was 20 from eight different species, and 44 in pools from fourteen species.

We conducted a chi-square test to determine if the abundance of the macroinvertebrates in the riffles and pools was the same at each site. The chi-square test resulted in a value of 48.83 which, at the .05 alpha level, was statistically significant. We rejected the null hypothesis; there is a difference between macroinvertebrate habitation and habitat type. However, more macroinvertebrates were found in riffles (170) compared to pools (144), meaning a statistically significant large amount of macroinvertebrates inhabit riffles instead of pools.

Table 4. Aquatic Macroinvertebrate Biotic Index Pollution Tolerance Classification (Biotic Index Card)

Class I	Class II	Class III
Ephemeroptera Plecoptera Trichoptera (Case Building) Decapoda Pelecypoda	Trichoptera (Net Spinning) Coleoptera Isopoda Amphipoda Megaloptera Odonata	Diptera Gastropoda Tricladida Oligochaeta Hirudinea Coleoptera Hemiptera

Biotic Index calculations for Site 1, Site 2 , and Site 3 were all greater than ten (17, 17, and 20, respectively), so the river can be classified as "clean."

Table 5. Water Chemistry Averages for all Sites

Location	Alkalinity (mg/L as CaCO3)	Conductivity (uS)	Flow	Salinity (ppt)	Termperature (°C)	pH	NO3-N (ug/L)	NH4-N (ug/L)	PO4-P (ug/L)	TN (ug/L)	TP(ug/L)
Dam Site	31.6529	272.6	0.292608	0.1	14.45	7.802	125.863	24.04	3.3	518.237	6.53192
West Branch	96.2818	286.4	0.492252	0.1	13.4	7.718	201.371	26.12	2.2	587.923	8.23412
East Branch	130.063	263.6	0.308458	0.1	16	7.662	6.34872	14.58	1.58	329.105	5.28728

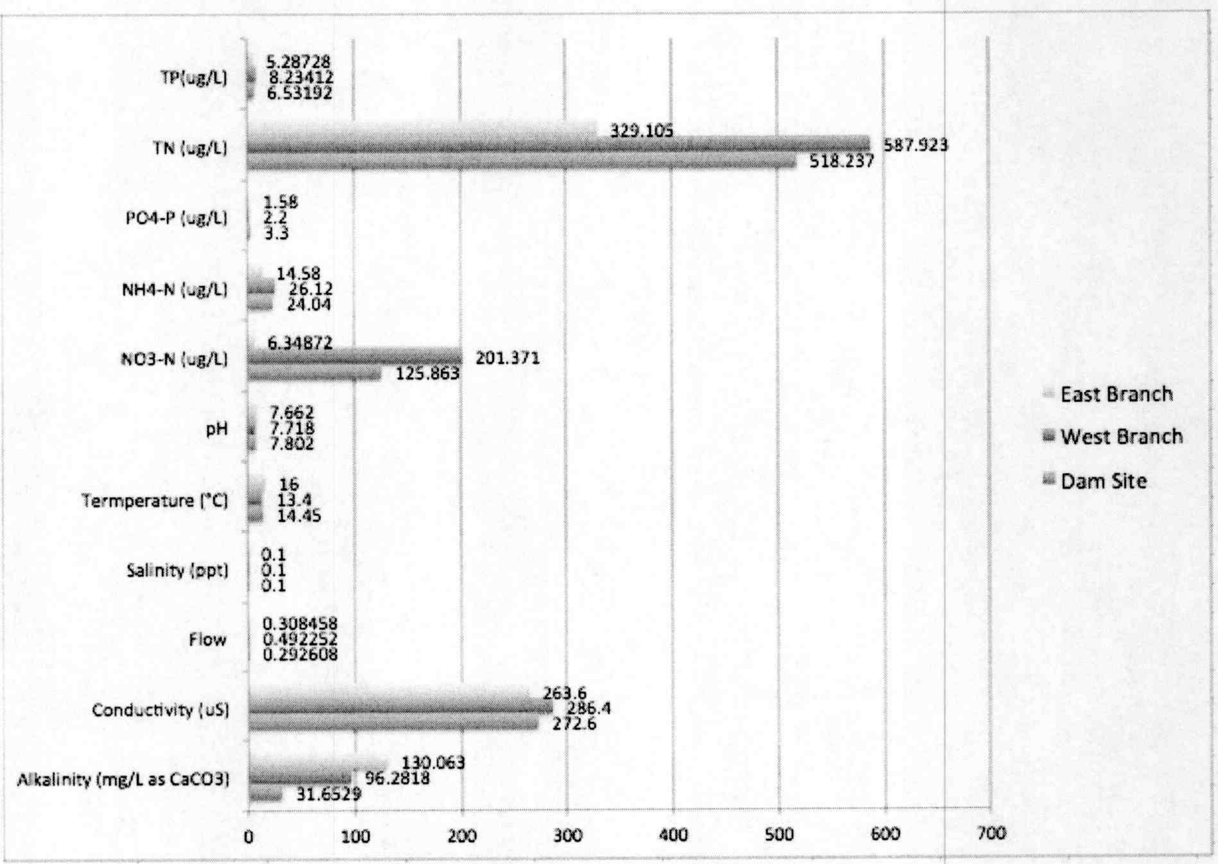

Figure 1. Water Chemistry Averages for all Sites

Table 5 and Figure 1 show the averages for all the water chemistry analyses from each site. Conductivity was highest at Site 2 with an average of 286.00 um. The average at Site 3 was 263.00 um and the average at Site 1 was 273.00 um. The average nitrate amount of Site 2 was

210.37114 ug/L and the Site 1 average was 125.86286 ug/L, while the Site 3 average was only 6.34872 ug/L. The average total nitrogen of Site 2 was 587.9226 ug/L and the Site 1 average was 518.2374, while the Site 3 averaged only 329.1048 ug/L. Alkalinity varied significantly, with Site 3 having the highest (130.063), followed by site 2 (96.1818), then Site 1 (36.6529).

Discussion

The data supported our hypothesis that there would be the largest amount of species diversity and abundance at the most down-river location (Site 1), due to the accumulation of runoff. We found that nearly twice as many macroinvertebrates were collected at Site 2 than Site 3, and the most found at Site 1. We expected the water quality at Site 2 to be worse than Site 3 based on the proximity of Site 2 to US 31 and the Pellston Airport. The runoff from cars, planes, and lawns could potentially impact the habitat and explain the lower macroinvertebrate diversity. We found 120 macroinvertebrates at Site 2 (Table 2), while only finding 64 at Site 3 (Table 3). However, Site 2 collection attempts yielded macroinvertebrates from seven different families while Site 3 had macroinvertebrates from nine different families (the most of any site).

Aquatic macroinvertebrates are good indicators of water quality. Different species have various tolerance levels, can live at least one year, and do not migrate or move often (West Virginia Department of Environmental Protection, 2015), which prevents them from relocating to a healthier part of the river system. If a river becomes contaminated where a population of pollution-intolerant macroinvertebrates live, the population will die out. *Ephemeroptera* (mayflies), *Plecoptera* (stoneflies), *Trichoptera* (caddisflies), and *Decapoda* (crayfish) are categorized as Pollution Sensitive Taxa (Sharpe et al., 2015), and their presence in a river indicates a high level of water quality. We found *Ephemeroptera, Trichoptera, and Decapoda* at

Site 1; *Ephemeroptera, Plecoptera, and Trichoptera* at Site 2; and all four orders at Site 3. The presence of these macroinvertebrates at all three sites suggests that the Maple River has a low level of pollution, which is supported by the results from the water quality tests performed at the sites and in the lab.

Some variation occurred in our water chemistry results in regards to alkalinity. Alkalinity is determined by the soil and bedrock through which the river passes and can indicate the supply of groundwater to rivers (Sircus, 2011). Site 1 had the lowest average alkalinity, while Site 3 had the highest average alkalinity (Table 5), indicating high groundwater intake by Site 3. The main sources of alkalinity in water are rocks containing carbonate, bicarbonate, and hydroxide compounds (Oram, 2014). We observed an abundance of rocks in the East Branch, which could be adding to the high alkalinity in the water.

Our water samples showed significant variation in conductivity, with the highest present in Site 2, followed by Site 1, and with the least in Site 3 (Table 5). Conductivity in water is affected by the presence of inorganic dissolved solids such as chloride, nitrate, sulfate, and phosphate anions or sodium, and aluminum cations (EPA, 2012). High levels of conductivity are associated with phreatotrophic (i.e. groundwater fed) environments, and both branches of the Maple River are fed by numerous groundwater springs. Site 2 had the highest nitrate levels, which could explain the increase in conductivity. In addition, conductivity of streams and rivers is affected mostly by the landscape of the water flow area (EPA, 2012). Streams that run through areas with clay soil tend to have higher conductivity as compared to streams running through bedrock because the clay has minerals that ionize in water (EPA, 2012). We observed clay-like substrate at Site 1, which could explain the increase in conductivity, and large rocks in Site 3, which could explain the decrease in conductivity. The actual effects contributing to conductivity are difficult to understand because, when measuring conductivity, the specific ions are not determined.

We observed differences in the nitrate (NO3) and total nitrogen of Site 3 as compared to Site 1 and Site 2 (Table 5). Site 3 of the Maple River was located upstream from any surrounding farmland or anthropogenic sources of pollutants, while Site 2 of the Maple River was located downstream from farmland as well as the Pellston Airport. Although nitrogen can be produced naturally in the environment, anthropogenic factors of fertilizer runoff and sewage waste can significantly impact the environment, which could explain the drastic difference between the nitrate and total nitrogen levels between the East and West Branches. Nitrate can enter the river system through leakage of manure, animal feed, and fertilizer directly from runoff, as well as entering from the atmosphere (USGS, 2014). Heavy rains can generate the runoff of excess nitrogen that can cause overstimulation of algae and aquatic plants. The excess growth of these organisms can clog water intakes, use up dissolved oxygen, and limit respiration efficiency of aquatic invertebrate, decreasing diversity (USGS, 2014). Site 2 had seven orders of macroinvertebrates while Site 3 had ten orders, indicating greater diversity in the East Branch. The limit of diversity of macroinvertebrates of the West Branch as compared to the East Branch could result from the increase of total nitrogen and nitrate levels.

Despite high levels of nitrate and ammonia, the Maple River and its branches are likely not polluted based on the presence of pollution intolerant macroinvertebrates. Nitrate encourages the growth of algae, a food source of many macroinvertebrates, including *Ephemeroptera*. We found 44 members of the order *Ephemeroptera* during our sampling at Site

2, but only twelve at Site 1 and six at Site 2. The high abundance of *Ephemeroptera* at Site 2 with the highest nitrate levels shows that *Ephemeroptera* may prefer environments with more nitrate because there may be more availability of food sources.

The chi-square analysis that tested for correlations between stream habitat, riffle or pool, and macroinvertebrate populations found that a statistically significant larger number of macroinvertebrates live in riffles than in pools. We predicted that there would be a larger number of macroinvertebrates in pools because that water velocity was lower, which could increase the organic material collecting there. This organic material could provide food and protection from predators. We did not find evidence to support our hypothesis, and found that most pools had sandy substrate with little or no organic material. The riffles we sampled from were often next to submerged logs or large rocks, which yielded the highest number of macroinvertebrates. Based on these results, the velocity of the water does not have as large an impact on the distribution of macroinvertebrates in rivers as we had assumed through our hypothesis. The number of different orders found in riffles compared to pools was higher for the former at Site 1 and Site 2.

Our study potentially had some limitations and flaws that could be sources of error in our accuracy. Collecting macroinvertebrate on a day after a rainstorm that increased water levels could affect the samples by altering the burrowing and locations of the macroinvertebrates. Additionally, possible variation in collection among individuals could occur. The number of kicks into the D-nets and the force behind the kicks may vary between collectors. The habitat where macroinvertebrates were collected (some from logs, some from stones, and others from various substrates) can impact the distribution and abundance of samples. Separating water quality sampling days with macroinvertebrate sampling days may impact results due to weather variations. It is possible that rainfall, as previously mentioned, can cause differing levels of nutrients in the water. If future replication is to be done, sampling should be taken on the same day.

For future research, more sites on each of the rivers could be tested. Originally, we had planned to test upstream and downstream locations on each of the branches to see if a correlation between locations and macroinvertebrate abundance existed. However, due to time and accessibility constraints, we altered the experiment. This study could be improved further by testing areas with varying levels of detritus and with different substrates to determine macroinvertebrate abundance and richness.

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