

# Effects of Salt Runoff on Aquatic Macroinvertebrate Colonies

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Aquatic macroinvertebrates bridge the gap in part between primary and secondary consumers, making them ecologically important for most aquatic systems. They are used as biotic indicators for water quality due to their sensitivity to pollutants and may be affected, specifically, by chloride ion concentrations in the water. Water quality can be evaluated by looking at species compositions and their respective tolerance values. To determine long-term effects of road salt on macroinvertebrate colonies, a total of 338 identified macroinvertebrates of 10 families (two generalized to class) at 9 sites were collected from before, after, and between three bridges on Turtle River in Bemidji, Minnesota. The most numerous families of invertebrates were Gammaridae, Ephemeridae, and Corixidae (116, 73, and 71, respectively), and the weighted average tolerance values ranged from 4.10 to 8.07. The conductivity was also measured at each collection by using 1000 mL of surface water, with an average of 294.0  $\mu\text{S}/\text{cm}$  for all sites, ranging from 285.5 to 305.1  $\mu\text{S}/\text{cm}$ . Conductivity had no significant relationship with tolerance values, diversity, or richness ( $P > 0.10$ ). There was, however, a significant positive, linear relationship between river kilometer and conductivity ( $P = 0.01$ ).

Faculty Sponsor: Dr. Andrew W. Hafs

## Introduction

Aquatic macroinvertebrates are present in many aquatic environments and are especially important in wetlands. Wetlands have been primarily researched for economic species, such as waterfowl, furbearers, and fish (Murkin and Wrubleski 1988). However, in recent years, more scientists have realized the importance to understand the food chain support, nutrient cycling, and overall productivity in wetlands (Murkin and Wrubleski 1988). Invertebrates play an important role in the food chain support, and are a link between plant litter, microorganisms, and invertebrate consumers (Murkin and Wrubleski 1988). Invertebrates both shred and scrape large leaf litter particles (such as amphipods and snails) for microorganisms to further decompose and consume the microorganisms directly (Nelson 1982, cited by Murkin and Wrubleski 1988). Most wetland birds feed on invertebrates at some point during their life cycle, but especially during breeding season, where protein and calcium requirements increase (Murkin and Wrubleski 1988).

Macroinvertebrates are commonly used as biologic indicators, specifically for water quality, as

there are many species which have intolerances to certain ion concentrations (Bant 2009). A potential threat to aquatic invertebrate density and diversity is unusually high salinity and chloride ion concentrations accumulated through road salt or road brines.

The United States uses over 10 million metric tons of road salt annually (Blasius and Merritt 2002). This does not include the millions of gallons of salt brine solutions such as Beet Heet, Apex-c, and RG8, which also increase chloride concentrations when polluted into water bodies (MN DOT 2019). It is important to study how the salinization of freshwater affects aquatic macroinvertebrate colonies, and to what extent. These salts dissolve and release chloride ions into the water, which can compromise the osmoregulatory processes in some aquatic insects (Benbow and Merritt 2004).

Inland freshwater typically has less than 120 mg/L salinity and chloride alike (Bourquin et al. 2013). The Maryland Department of Natural Resources is monitoring long-term non tidal concentrations of chloride and have found significant increasing trends at 80% of their stations

(Bourquin et al. 2013). This is likely due to the accumulation of road salt from consecutive years of heavy usage.

There have been many studies looking at the direct relationship between salinity and macroinvertebrate colonies using both field and laboratory methods. Blasius and Merritt (2002) have found that leaf litter decomposition rates were faster upstream from road bridges, however, the decrease downstream could be due to sedimentation rather than affected macroinvertebrate colonies. The same research found, in laboratory tests, that *Gammarus* (Amphipoda) were possibly affected by chloride concentrations greater than 5000 mg/L when exposed for more than 24 hours, but that most of the invertebrates were unaffected by concentrations up to 10,000 mg/L for 24 to 96 hours (Blasius and Merritt 2002). Other lab tests, however, showed 100% mortality of concentrations between 8,000 mg/L and 10,000 mg/L for 48 hours for *Hydroptila* sp. and *Cericotopus* sp. (Blasius and Merritt 2002).

Salt concentrations in rivers do not commonly reach these levels for lengths of time that cause complete mortality but are still susceptible to high enough concentrations of chloride to predict an effect on invertebrate populations.

Invertebrates are more inactive in the winter, which makes them more resistant to increased salinity during peak salting seasons, but not necessarily peak conductivity. Rupprecht et al. (2009) monitored conductivity in a stream over the winter and into the late spring, finding that the baseline conductivity over the winter was greater than 4  $\mu\text{S}/\text{cm}$ , and the baseline conductivity in late spring through fall was less than 1  $\mu\text{S}/\text{cm}$ , when temperatures were greater than 10°C. Overspray from winter driving caused small peaks, less than 6  $\mu\text{S}/\text{cm}$ , which returned to baseline in early spring, however, snowmelt in mid spring resulted in peak conductivity of 30  $\mu\text{S}/\text{cm}$ , during which invertebrates are more active and can be negatively affected by dissolved chloride (Rupprecht et al. 2009).

The objective of this study was to determine if salt runoff from bridged roadways over rivers impacts the annual downstream benthic macroinvertebrate colonies and water quality.

## Methods

### Study Area

This study analyzed macroinvertebrates collected from three bridges on Turtle River in Bemidji, MN. There were a total of nine collection spots, three per bridge, each with cattails *Typha latifolia* as the major vegetation, and with mucky, silty substrate. There were little to no rocks greater

than 1  $\text{cm}^3$ . All three bridges were constructed by concrete. Bridge one was on a paved road, and bridge two was on a gravel road, each intersecting with Turtle River on Turtle River Lake Road. The third bridge was on a gravel road, intersecting with Turtle River on Birchmont Beach Rd. NE. Bridge one had tresses supporting the bridge, and bridges two and three had culverts directing water flow.

### Collecting data

Samples were collected in 2020 on 10/15, 10/19, and 10/21 for bridges one, two, and three, respectively. Each bridge had three collection sites (a, b, c) associated with it, all on the left-hand side of the river when facing downstream. Collection site a was 61 m before the bridge, b was directly after the bridge, and c was 152 m after the bridge. After traveling to each collection site via boat, approximately 1000 mL of water was collected with a Nalgene bottle dipped into the surface water and stored for later analysis. Then, 0.0027  $\text{m}^2$  of substrate was scooped from the bottom of the river by inserting a 30 x 30 cm D-net 3 cm into the substrate with the opening at right angle with the bottom. The net was then moved in a straight line, parallel to the river bottom, for 30 cm, and quickly brought from the bottom to the surface at a 45-degree angle until the d-net was out of the water. Holding the opening of the net up, the water was drained until the mix of substrate and water could be completely emptied into a gallon zip loc bag. A stick was used to move around the substrate in the net to allow flow of water through the filter for most samples. This process was completed for each of the nine collection sites.

The water and dirt from the collection site were sorted through within four hours of being collected. One bag at a time, the contents were emptied into a large bin. The bag was rinsed out with cold water into the same bin. Small portions of substrate were poured into a 23 x 33 cm shallow tray, just covering the bottom. With a bright, warm light and tweezers, obvious invertebrates were picked first, with smaller ones collected by agitating the water and dirt, then looking for movement. The invertebrates were rinsed in a cup of cold water, then placed into a small, sealable bottle filled with approximately 500 mL of a 90% Ethanol solution. After thoroughly searching for invertebrates, the remaining water, substrate, and vegetation was poured into a disposal bin. Before proceeding to the new collection site bags, the first bin and dissecting tray were rinsed thoroughly.

### Identification and measurement

Species were identified using a 2x Ward's magnifying scope. Specimens were identified to the

family or class level by using Bouchard (2004) and Stroud (2020).

The water samples were tested for conductivity using a YSI. The probe was rinsed thoroughly with deionized water, then placed in each Nalgene sample for at least 20 s, or until the conductivity reading stabilized.

**Analysis**

Weighted averages were calculated for species tolerance levels at each collection site, with 1 being low tolerance and 10 being high tolerance (SWCS 2015).

Linear regression models were created to relate conductivity to the following variables: tolerance values, species diversity, species richness, and river kilometer downstream. A paired t-test was used to relate upstream and downstream conductivity differences.

**Results**

There were ten different identified families collected at all sites. The most numerous species were of the families Gammaridae, Ephemeridae, and Corvidae (Table 1). The tolerance value was 5.72 for the weighted average across all locations, ranging from 4-9.

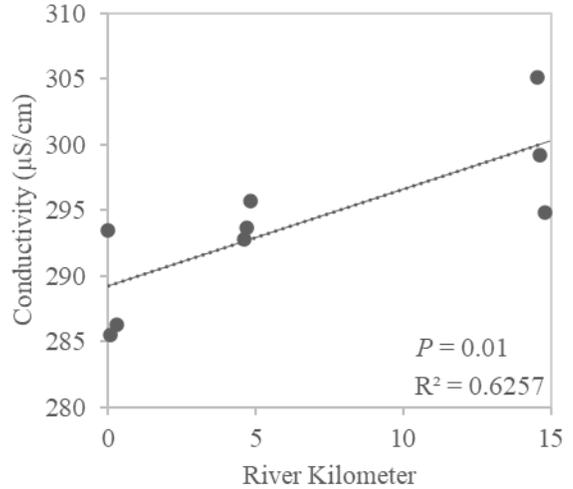
**Table 1:** Names of families/class of macroinvertebrates collected, number of macroinvertebrates found, and the tolerance value of each family or class. D-net invertebrate samples collected October and November 2020 at Turtle Lake River, Bemidji, MN.

Family/Class	n	Tolerance
Gammaridae	116	4
Ephemeridae	73	4
Corixidae	71	9
Gastropoda	26	7
Bivalvia	21	7
Coenagrionidae	11	9
Libellulidae	8	7
Polycentropodidae	6	6
Phryganeidae	5	4
Unknown	2	Null
Gyrinidae	1	4
10 Families/Classes	Total 338	

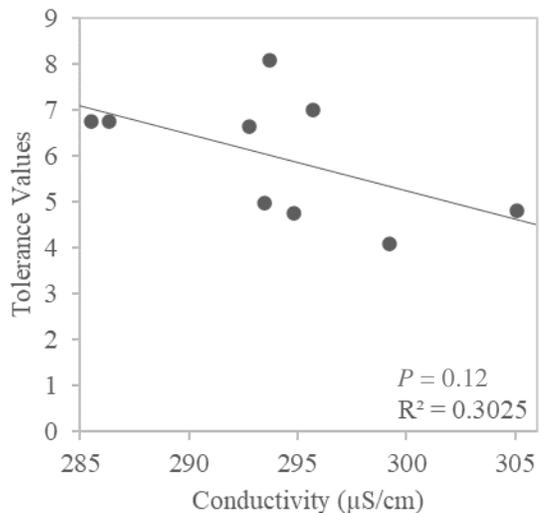
The conductivity ranged from 285.5  $\mu\text{S/cm}$  to 305.1  $\mu\text{S/cm}$ , and the average value for all sites was 294.0  $\mu\text{S/cm}$ . There was an increase in conductivity as river kilometer increased from upstream to

downstream ( $P = 0.01$ ), but not enough to influence macroinvertebrate colonies (Figure 1).

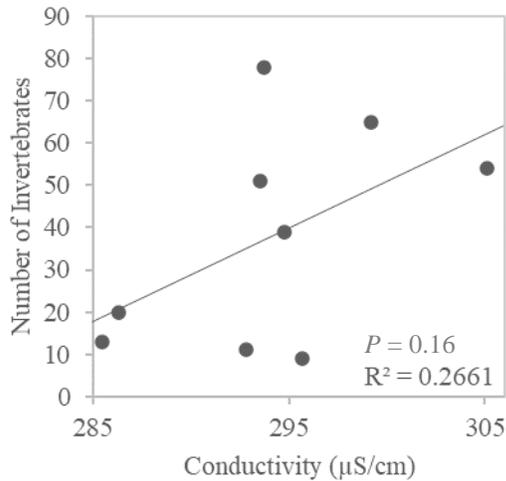
There was no significant relationship between conductivity and tolerance ( $P = 0.12$ ; Figure 2), conductivity and richness ( $P = 0.16$ ; Figure 3), or conductivity and diversity ( $P = 0.15$ ; Figure 4).



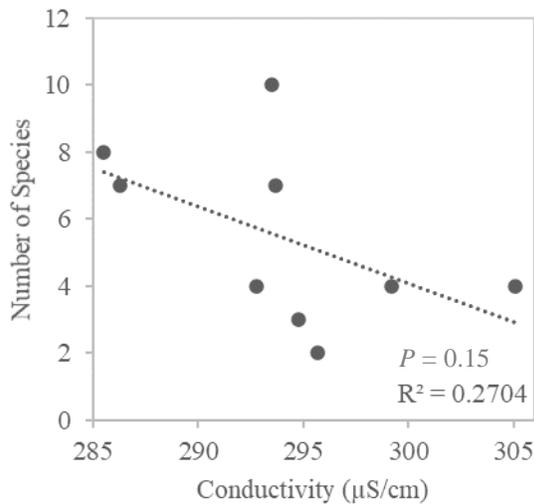
**Figure 1:** Conductivity ( $\mu\text{S/cm}$ ) river kilometer relationship. River kilometers began at collection site 1a (0 km) and ended at collection site 3c (14.8 km). Conductivity samples collected October and November 2020 at Turtle Lake River, Bemidji, MN.



**Figure 2:** Conductivity ( $\mu\text{S/cm}$ ) tolerance value relationship for macroinvertebrates. D-net invertebrate and water samples collected October and November 2020 at Turtle Lake River, Bemidji, MN.



**Figure 3:** Conductivity ( $\mu\text{S}/\text{cm}$ ) species richness relationship for macroinvertebrates. D-net invertebrate and water samples collected October and November 2020 at Turtle Lake River, Bemidji, MN.



**Figure 4:** Conductivity ( $\mu\text{S}/\text{cm}$ ) species diversity relationship for macroinvertebrates. D-net invertebrate and water samples collected October and November 2020 at Turtle Lake River, Bemidji, MN.

### Discussion

While investigating effects of road salt on stream macroinvertebrates in Michigan, USA, Blasius and Merritt (2002) found that upstream and downstream locations had no significant differences in diversity and composition that could be attributed to road salt. These findings corresponded with the results in this study, with no significant relationship found between conductivity and the three variables:

tolerance, diversity, and richness of macroinvertebrates. In their laboratory experiments, Gammarus and two caddisfly species had a response to NaCl concentrations of 7700 and 3526 mg NaCl/L, respectively (Blasius and Merritt 2002). At the greatest conductivity, species in this study were exposed to  $\mu\text{S}/\text{cm}$  values comparable to approximately 1/18th of the lower mg NaCl/L value. It is unlikely the conductivity in Turtle Lake River nears ranges that could significantly affect these two invertebrate groups.

While studying the effects of road salt from highway runoff on macroinvertebrate communities in three streams in Norway, Bant (2009) concluded that macroinvertebrates had no acute negative responses to road salt. This suggests that conductivity may not be the best measurement to determine if macroinvertebrate colonies are affected by anthropogenic pollutants. Even though conductivity was highly associated with river kilometer moving from upstream to downstream in Turtle River Lake, it was not useful in estimating macroinvertebrate colonies.

The variance seen in invertebrate colonies in this study could be attributed to factors other than road salt, such as the sediment loading patterns found from upstream to downstream. Sediment loading could have caused the accumulation of particles downstream, resulting in both greater conductivity readings with river kilometers and variance in invertebrate communities due to substrate changes.

In this study, the tolerance ranged from 4-9, with an average of 5.72. Turtle Lake River may not hold the best macroinvertebrate community for analyzing the effects of road salt runoff. Species such as some caddisflies, mayflies, and stoneflies have low tolerance and are good bioindicators. A river community with these species before and after a major source of pollutants could be better for analyzing effects on sensitive macroinvertebrates.

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