

Spring 2019

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Patrick Kelley  
*College of DuPage*

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### Recommended Citation

Kelley, Patrick (2019) "Relationship of Dissolved Oxygen Availability and Chloride Contribution to Freshwater Macroinvertebrate Diversity Found in Various Ponds in Northern Illinois," *ESSAI: Vol. 17* , Article 24.

Available at: <https://dc.cod.edu/essai/vol17/iss1/24>

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Relationship of Dissolved Oxygen Availability and Chloride Contribution to  
Freshwater Macroinvertebrate Diversity Found in Various Ponds in Northern Illinois

by Patrick Kelley

(Biology 1152)

**A***bstract.* Dissolved oxygen levels and the presence of salt concentrations in freshwater systems serve as good indicators to the health of said systems and to relationships involving abundance and diversity of particular kinds of organisms such as benthic macroinvertebrates. Habitat differences, such as presence of aquatic vegetation can also influence invertebrate abundance. Research was conducted on 6 different freshwater sites on the College of DuPage campus. We collected benthic macroinvertebrates and water samples for analysis, specifically comparing organismal abundance and diversity to amounts of dissolved oxygen and salinity among sites, grouped according to habitat characteristics. There was a greater abundance of macroinvertebrates and lower chloride levels in ponds with emergent vegetation along their margins as compared to retention ponds without. We did not find clear differences in biodiversity between these groups of sites, possibly due to the broad groupings of taxa. Nonetheless, this study reflects the importance of benthic macroinvertebrates as indicators of human impact on freshwater aquatic habitats.

*Keywords: abundance; benthic macroinvertebrates; chloride; dissolved oxygen; diversity; freshwater ponds.*

## INTRODUCTION

Multiple elements must play a key role and be part of a freshwater ecosystem for it to flourish with its abundance of creatures lurking in, above, and around the system. It is vital that certain elements are sufficiently available for an organism to survive and reproduce in the environment. In aquatic habitats, dissolved oxygen (DO) is particularly important to organisms and is affected by rates of photosynthesis and cellular respiration (Coffin et al. 2018). For freshwater habitats, dissolved oxygen depends on key factors such as air temperature and even the salinity of water. Data around these factors enable scientists to grasp an understanding of how a network of abiotic processes influence a network of organisms. These processes have to occur for continued growth in developing macroinvertebrates. Changing levels of oxygen, salinity and temperatures happen frequently due to changes in atmospheric pressure, temperature fluctuations and air quality. For example, seasonal changes can impact DO levels, which tend to decrease as temperatures get warmer because oxygen can evaporate from water surfaces easily.

A relationship between temperature and quantifying rates of metabolic activity can be examined in aquatic organisms that factors in DO levels (Young and Rhodes 1974). As temperatures fluctuate, metabolic activity for ectotherms may increase or decrease to their environments, ultimately determining how much dissolved oxygen is available to the entire system (Verberk et al. 2011). Pollution may affect how much DO is readily available for aquatic organisms and serve as an indicator for the health of a system (citation needed). Another health indicator for freshwater ecosystems involves the diversity of macroinvertebrates present in the aquatic environment. Some macroinvertebrates are tolerant to high DO levels and some are tolerant to low levels of DO. Species richness, or diversity, and species abundance can then be observed based on how much DO is available.

Understanding the foundation of a habitat is imperative to studying other mechanisms that offer some relationship to diversity. Introduced salts can have a large impact on the behavioral aspects of macroinvertebrates and can also impact organismal diversity. Tolerance that aquatic macroinvertebrates have to salts varies. Too much or too little of these salts such as chloride may inhibit certain functions of macroinvertebrates (Berezina 2003). A part of understanding salinity stems from any freshwater pond or lake located near roads. It is not uncommon for salt on roads to end up in these waters, especially during the winter when salts become common practice to melt ice and even during heavy rainfall leading to salt runoffs. Ponds or retention basins sloped to quickly receive run-off may experience a rapid increase in salinity. If salinization continues to increase it could devastate macroinvertebrate diversity overall and drastically affect readily dissolved oxygen in the process (Dugan et al. 2017).

It would be ideal to look at most, if not all, of these factors in order to develop a proper analysis provided here in this specific study, however, through the techniques used to collect substrate at the bottom of ponds, marshes and swamps two hypotheses will be considered. The first hypothesis predicts that a higher diversity of macroinvertebrates will be associated with freshwater aquatic habitats exhibiting higher dissolved oxygen levels. The second hypothesis predicts that diversity of macroinvertebrates will be lower in freshwater habitats with high levels of salts as compared to nearby habitats with lower salt levels. This study took place around DuPage County located in Northern Illinois, home to many bodies of freshwater in the form of ponds, swamps and marshes. The College of DuPage in Glen Ellyn harbors several of these freshwater systems that were analyzed for macroinvertebrate abundance water chemistry tests for dissolved oxygen and salinity. This study will observe any noticeable relationship between dissolved oxygen levels and chloride levels to the impact on the diversity of macroinvertebrates present in the different aquatic environments.

## MATERIALS AND METHODS

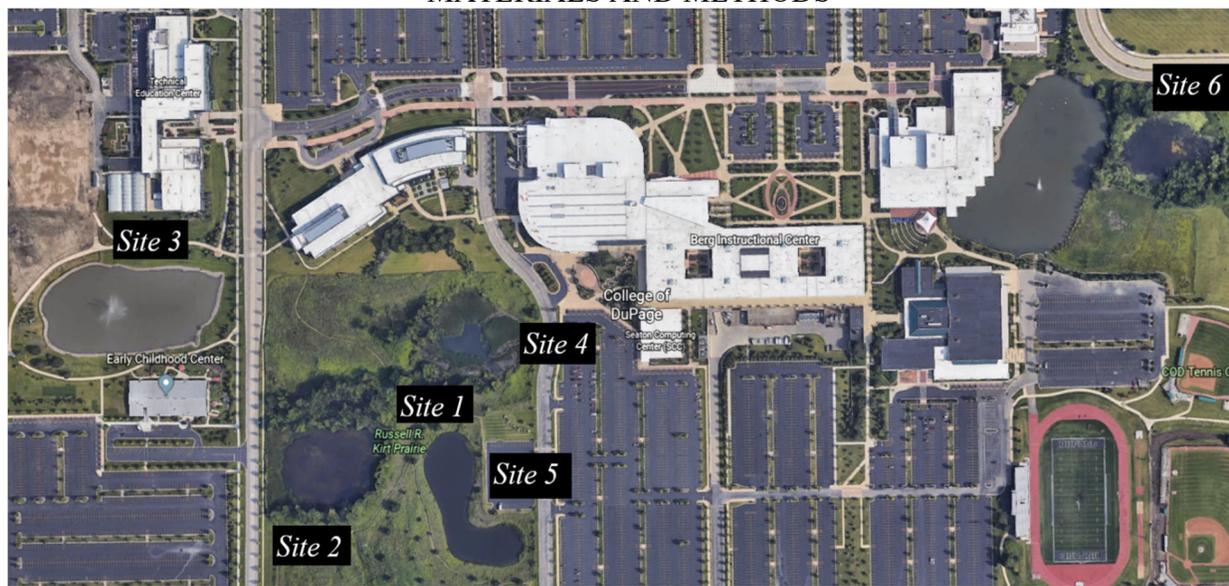


FIG 1. Aerial view of the DuPage Campus with locations of each of the 6 sites that were sampled (Note: Site 1 area falls underneath the tree line to the left of the “Site 1” label).

The study took place on April 1<sup>st</sup>, 2019, in the Russell R. Kirt Prairie, on the College of DuPage campus. Data was collected on 6 different sites: Site 1-Swamp (underneath tree line); Site 2-Pond By Swamp; Site 3-Reda; Site 4-Mini Swamp; Site 5-Retention Pond; Site 6-East Marsh. Sites 1

and 4 are ephemeral cottonwood swamps, Sites 2 and 6 are emergent vegetation ponds with cattails along the shore, and Sites 3 and 5 are retention ponds lacking emergent vegetation. The data was sampled at temperatures of 8.00 °C the first day following 11.00 °C the second day where site 5 was sampled and at water depths of 40.00-50.00 cm, with site 4 at 11.00 cm.

Substrate content was collected at the bottom of all 6 sites to observe macroinvertebrate diversity and abundance. Small groups of roughly 3 people collected substrate by using extended dip nets one meter out into the freshwater and followed a 3 jab technique into the substrate towards them. They exposed the net into the air after the third jab. The collected substrate was then dumped into a 19-L sampling bucket with rinse from that environment of another bucket to remove any still attached substrate to the net. Four 100-ml jars of water from each site were filled to capacity and tightly capped, to be used for chemical analysis. On April 3 in the lab, students sorted the substrate into trays and used identification keys (Opal 2016, UNH Center 2013) to group the macroinvertebrates into Family or Order, tolerance levels to pollutants and habitat differences. Organisms were observed under a dissecting microscope (8x-20x magnification) to aid in identification. In addition, chemical analyses on water samples for each site were performed in the lab on April 3. LaMotte test kits were used to measure dissolved oxygen levels and chloride levels by titration (LaMotte Co., Chestertown, MD). Data was collected, numerically, in parts per million (ppm). Average values from the mean of 2 water samples per site were reported.

A 2-sample Student's t-test was conducted to evaluate any significant differences in the abundances of macroinvertebrates present between groups of sites. Calculations of macroinvertebrate diversity for groups of sites were based on 2 indices: Simpson's (Simpson 1949) and Shannon's (Shannon and Weaver 1949). The Simpson Index gives more weight to the more abundant groups and the Shannon Index assumes all taxa are represented among random samples (DeJong 1975). The similarity of taxa between groups of sites was compared using Sorensen's Coefficient (Sørensen 1948), with values ranging from 0-1, and is influenced by the most abundant groups. Statistical inference was based on  $\alpha = 0.05$ .

## RESULTS

Of 6 sites visited, data was analyzed from 4 sites placed into 2 different groups: Group 1 consisted of Site 3 (1.25 ha) and Site 5 (0.65 ha) as two retention ponds without emergent vegetation; Group 2 consisted of Site 2 (0.85 ha) and Site 6 (0.60 ha) as two ponds with emergent vegetation and more gradually sloping shorelines than the Group 1 sites;. Group 1 had substrates with gravel and rocks, although more muck was present in Site 3 as compared to Site 5. Microscopic mites were common in Site 5 and uncommon in Site 3. Site 3's margins were more steeply sloped compared to Site 5. As for Group 2, an abundance of macroinvertebrates were found at Site 2 and at Site 6, the latter site sampled 1 day later than the rest. Approximately 2.5 times more macroinvertebrates were observed in Group 2 sites as compared to Group 1 sites (Fig. 2).

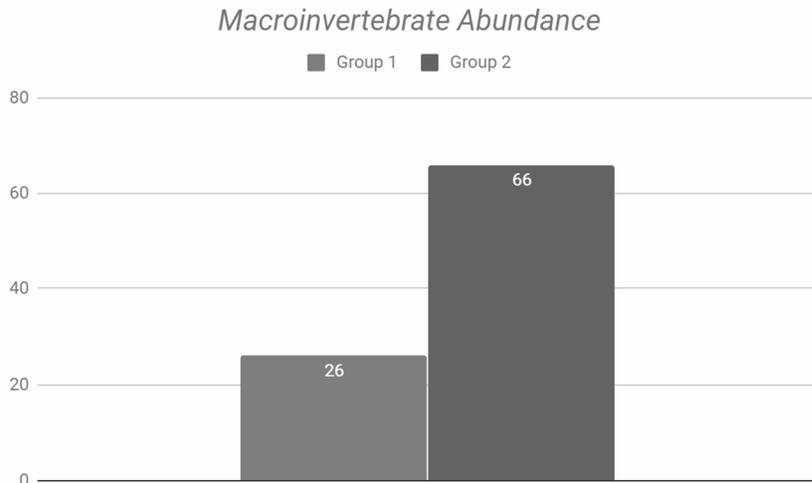


FIG 2. Abundance of macroinvertebrates found in Group 1 (Sites 3 & 5) and Group 2 (Sites 2 & 6).

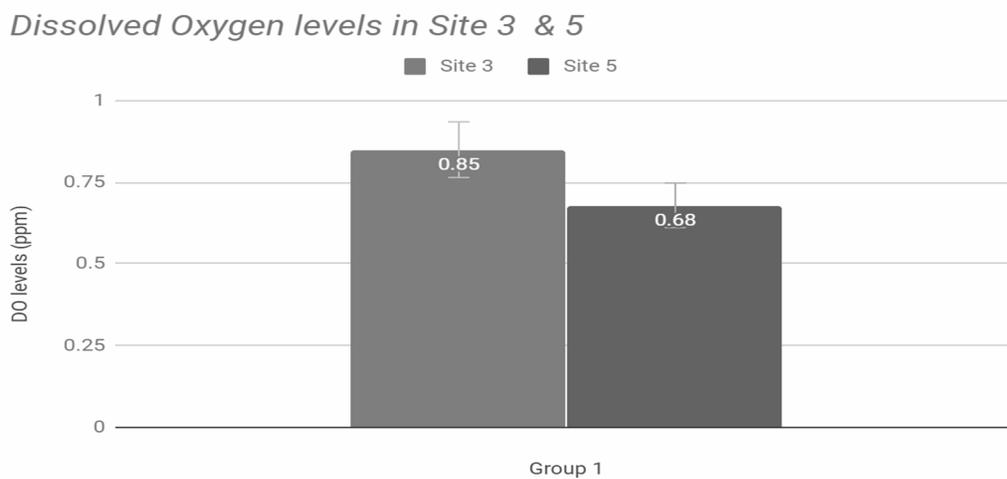
Macroinvertebrates were categorized into 11 unique taxa, mostly identified to order (Table 1). The two groups of sites shared 4 out of 8 taxa per group. The proportion of individuals across taxa  $(n/N)^2 = p_i^2$  was calculated to carry out sum values based on the Simpson Index, where  $D = \frac{1}{\sum p_i^2}$  and the Shannon Index  $H' = -\sum p_i \ln p_i$ . Diversity indices were similar for the sites in Group 1,  $D = 5.452$  and  $H' = 1.851$ , and Group 2,  $D = 5.542$  and  $H' = 1.860$  (Table 1). The Sorenson's Coefficient = 0.500. There was a marginally significant difference in the counts among taxa between the groups of sites (Fig. 2;  $t = 2.232$ ,  $df = 9$ ,  $P = 0.0525$ ). Small aquatic mites were not included in the analyses but Group 2 sites were observed to have an abundance.

TABLE 1. Abundances of benthic macroinvertebrates by taxa in the Group 1 sites (retention basins without emergent vegetation) and Group 2 sites (ponds with emergent vegetation). Blank counts indicate none observed. Taxonomic level identification (C) = Class, (O) = Order, (F) = Family

Taxon	Group 1 Sites count	Group 2 Sites count
Coleoptera (O)	1	5
Diptera: Chaoboridae (F)	2	4
Diptera (O): other	2	
Hemiptera (O)		1
Odonata (O)	5	12
Oligochaeta: Naididae (F)	6	8
Gastropoda (C)	1	20
Ephemeroptera (O)	7	
Trichoptera (O)	2	
Bivalvia: Spaeridae (F)		10
Tricladida: Planariidae (F)		6

Mean dissolved oxygen levels were similarly very low for all sites within and between the groups being compared (Fig. 3).

(a)



(b)

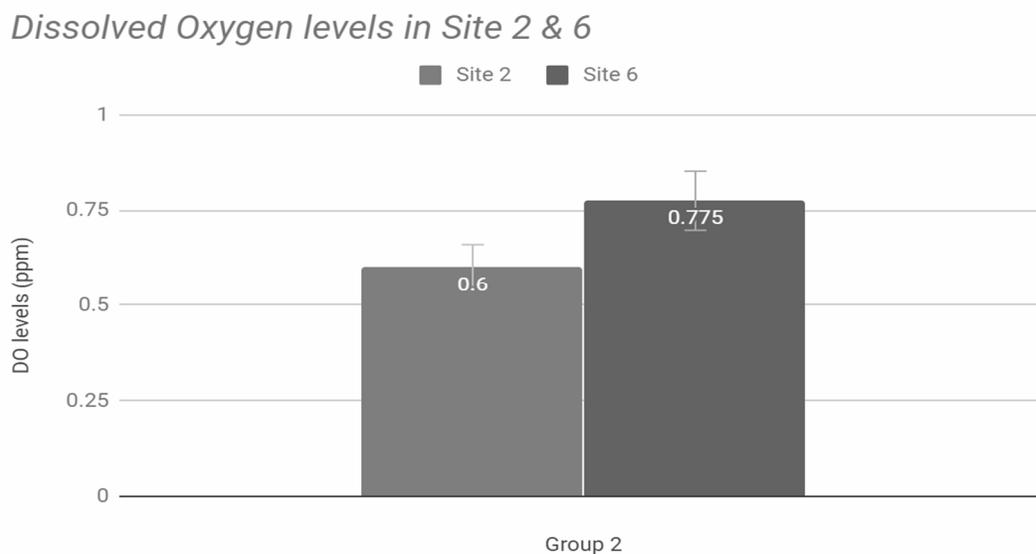


FIG 3. Dissolved oxygen levels in parts per million (ppm), with error bars, compared between paired sites and between Group 1 and Group 2. (a) Average DO level values taken for Group 1, Sites 3 (0.85 ppm) & 5 (0.68 ppm). (b) Average DO level values taken for Group 2, Sites 2 (0.60ppm) & 6 (0.78ppm).

The chloride levels among sites of the 2 different groups were notably different, with Group 1 (Site 3 = 7600 ppm, Site 5 = 7760 ppm) having chloride levels up to an order of magnitude greater than those of Group 2 (Site 2 = 3280 ppm, Site 6 = 720 ppm).

#### DISCUSSION

The first hypothesis suggested that higher levels of dissolved oxygen would increase the diversity for macroinvertebrates. This was not demonstrated through the samples collected in this study. Dissolved oxygen levels were not largely different from either groups. These samples were

collected on cold days (11 °C for air temperature and a range between 8 °C and 9 °C for the bodies of freshwater). This could have contributed to consistent levels of DO across the aquatic habitats. That is, the tendency for there to be less dissolved oxygen levels at higher temperatures may not have been observed based on it only being cold at the time of sampling. Despite colder temperatures outdoors, dissolved oxygen levels were unexpectedly low. This could be the result of the delay in measuring DO following water sample collection, and reduction of oxygen from cellular respiration of microorganisms in the water samples. It would seem that Group 2's presence of emergent vegetation might contribute to an increase of oxygen available in water based on the plant's release of that oxygen during photosynthesis. This difference might be pronounced during warmer seasons of plant growth. Ponds may go through cycles during the spring and summer known as pond turnovers that would deplete the amount of oxygen that can dissolve in these waters. The study took place on April first when temperatures were a lot colder.

There was not direct support for the second hypothesis that predicted biodiversity would be lower in sites with higher chloride levels. Chloride levels were much greater in the Group 1 sites, retention basins, as compared to the Group 2 sites, ponds with emergent vegetation and shallower slopes. However, biodiversity indices were similar between groups, although Group 2 sites with emergent vegetation had the greatest overall abundance of macroinvertebrates. In addition, the 2 sample t-test for means revealed that there was only marginally a significant difference between the abundances of both groups. This may be due to the large amount of variation among the counts of taxa in the Group 2 sites. The Shannon index values were a little bit greater for Group 2 than Group 1 which may factor into the fact that Group 2 had a little over a twofold difference in the abundance of organisms present. In addition, while both groups had mites present, Group 2 had a greater abundance. This could arguably be seen as a factor that could increase the diversity of Group 2. Although mites were not counted as macroinvertebrates, a higher presence of mites could suggest higher water quality which, in turn, can increase diversity amongst other organisms who can also adapt greater water qualities.

Considering the chloride levels between groups of sites, it is worth noting that chloride levels for Group 1 sites were relatively close, and high, compared to the Group 2 sites. This may make sense based on the fact that both of these retention ponds were located near busy roads, as shown on the map (Fig. 1). Busier roads, especially for northern Illinois, require the use of de-icers, or salts that can melt snow, which are used frequently in Illinois. Excessive use of salts may follow runoff during heavy precipitation into these steep retention ponds.

Future work has the potential to enhance this study even further. As a first, data can be sampled for weeks or even months. Given that seasonal changes tend to govern the available oxygen for freshwater organisms, collecting data over extended periods of time can account for these changes for one to infer the degree that seasonal changes affect the diversity of freshwater macroinvertebrates. In addition to seasonal changes, observing the composition of ponds that may be flooded or vulnerable to increasing salts during the winter can lead to potential relationships and can lead one to ask to what degree these factors have on DO levels and diversity. Decomposition rates, leaf litter, and tolerance to of certain organisms to pollution can be observed for data collected over a longer period of time to see how these factors affect DO levels and diversity among macroinvertebrates. Data was sampled for only two days which may not offer the full picture of what could happen that would support the two hypotheses presented in this study. Examining macroinvertebrates can also lead to further studies that relate human interactions directly or indirectly to freshwater systems. These interactions may include gases released by factories and cars, urban growth near freshwater bodies, and even the gradual changes with global warming. Macroinvertebrates are indicators for many of these freshwater systems and there may be other applications in studying them beyond just diversity, abundance and their tolerance to DO levels.

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