

Analyzing the Impact of Zebra Mussels (*Dreissena polymorpha*) on Water Nutrient Levels

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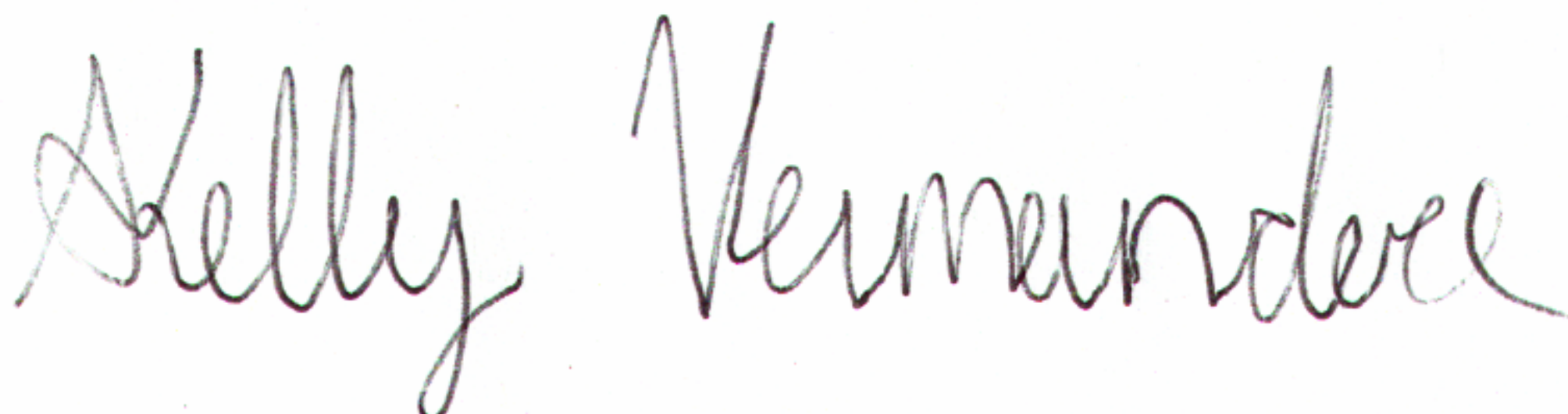
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Abstract

We studied changing water nutrient levels in tanks filled with different densities of zebra mussels to explore their influence on water quality. This study took place on University of Michigan Biological Station Property in the northern Lower Peninsula of Michigan. We placed zebra mussels from Douglas Lake into five tanks at the Alfred H. Stockard Lakeside Laboratory. We then took water samples every hour for 8 hours so that we could analyze changes in nutrient levels over time. Through chemical analysis, we were able to examine the concentrations of 14 nutrients; however, only 3 nutrients (nitrate, ammonium, and calcium) had changed significantly. The results showed that calcium and nitrate decreased with increasing density, and ammonium increased both over time and with increasing zebra mussel density. These shifts in nutrient levels indicated that zebra mussel infestation can have a huge impact by harming native species, the transfer of nutrients and energy through aquatic food chains, and the health of aquatic ecosystems. Essentially, zebra mussels can use up calcium that is necessary for the survival of some native species, changes in nitrogen concentrations can cause cascading trophic effects at regional and global scales, and an increase in ammonium can cause eutrophication, leading to dead zones and a decline in aquatic organisms.

Introduction

Zebra mussels (*Dreissena polymorpha*) were introduced into the Great Lakes from Russia in 1986 via ships' ballast water (MacIsaac, H, J., 1996). In their new environments, they "cemented" themselves on any available hard substrate (e.g. rocks, wood, man-made walls, native mussels, and other organisms) in clusters and were able to withstand heavy water flow

(Davis, A., Hanewish, S.M., et al. 1990). They had a high fecundity rate of 30,000 eggs per female in their first years of sexual maturity, and up to 40,000 by the third and fourth years. Zebra mussels and their offspring were highly dispersed through water and wind currents, ships, and waterfowl (Schloesser, D.W., Nalepa, T.F., 2013). Since 1990, they have established a presence in all five Great Lakes, reaching densities of one million individuals per square meter in some areas (Davis, A., Hanewish, S.M., et al. 1990).

Despite their brief history in the Great Lakes, zebra mussels have had huge abiotic, biotic, direct, and indirect impacts (MacIsaac, H, J., 1996). They have negatively affected the environment by clogging intake pipes, damaging boat hulls and engines, outcompeting fish and other mussels for food, and harming endangered species (MacIsaac, H, J., 1996). They alter energy flow, community structure, trophic interactions, and populations of native species across multiple trophic levels (Whitledge, G., Weber, M.M., et al. 2015). As filter feeders found in large numbers, zebra mussels have important impacts on nutrient concentrations in aquatic systems (Qualls, T.M., et al. 2007).

They impact nutrient dynamics when they use particulate nutrients for growth and excrete the nutrients in a dissolved form. The particles excreted as feces and pseudofeces by zebra mussels are returned to the water column and become deposited on lake bottoms or are resuspended and transported to deeper areas (Qualls, T.M., et al. 2007). Zebra mussels indirectly affect cycling processes by changing the composition and dynamics of organisms such as phytoplankton, protozoans, and small zooplankton that normally recycle nutrients in the Great lakes foodweb (Gardner, W.S., Cavaletto, J.F., et al. 1995).

Until the present day, only a few models have been developed that can predict the effects of zebra mussels on water quality and nutrient levels (Lindim, C. 2015). Studies across the Great

Lakes have used long-term data sets to determine the impacts of zebra mussels in aquatic systems; however, their impacts on smaller water systems such as inland lakes have not been thoroughly investigated (Qualls, T.M., et al. 2007). Their activity is speculated to affect nutrient cycling to a greater extent with greater mussel density, but this has also not been studied completely (Lindim, C. 2015).

We designed a short-term study to investigate the biogeochemical effects on nutrients that zebra mussels have on Douglas Lake, an inland lake in northern Michigan. The nutrients we analyzed were: chloride, fluoride, bromide, nitrite, nitrate, phosphate, chlorophyll-a, silicate, sulfate, sodium, potassium, magnesium, calcium, and ammonium. Our null hypothesis was that there would be no significant change in nutrient levels over time. Also, that there would be no significant change in water nutrient levels between tanks containing different densities of zebra mussels. We predicted that a greater density of zebra mussels would have a greater impact on the rate of change in nutrient levels and on ecosystems when they are present for a longer period of time.

Methods

This study took place on August 7, 2016 at the University of Michigan Biological Station (UMBS) located in Pellston, Michigan. We placed zebra mussels from the shoreline of Douglas Lake (35° 57, 40" N, 83° 32, 20" W) into tanks at the Alfred H. Stockard Lakeside Laboratory. Our five tanks were each filled with 5 gallons of water from Douglas Lake and air stones were used to maintain a steady influx of oxygen.

We organized the zebra mussels into groups of 0, 200, 400, 600, and 800 individuals. However, the actual amount of zebra mussels in each tank was a random sample of 0, 199, 398, 588, and 806 individuals, but these counts might have been off by as much as 50 zebra mussels

because counting each individual in a group was difficult. The tank with 0 zebra mussels was our control group for this experiment.

We took an initial 50 mL water sample, then sampled every hour for eight hours to test water nutrient levels. The chemical analyses for the nutrient cations and anions were run using a ThermoFisher/Dionex Integrion HPIC; silicate was run using the automated molybdenum blue method on an Alpkem FS3000 rapid flow analyzer; and the chlorophyll-a was analyzed using a TD-3100 laboratory fluorometer by Turner Designs.

Using SPSS, we performed a multiple comparison tukey test to determine which nutrients contained significant differences for densities and set our significance level at 0.05 for each. The nutrient concentrations that significantly changed with an increase in zebra mussel density and/or over the period of 8 hours were nitrate, ammonium, and calcium. R and R commander version 3.2.3. with linear regression analyses were used to test for the significance of time and density.

Results

Figure 1: Analysis of nitrite (NO_2) concentration (mg/L) over a period of 8 hours with 5 densities of zebra mussels (0, 200, 400, 600, and 800). The concentration of nitrite was not significant for time ($R^2 = 0.017$) or density ($R^2 = 0.046$).

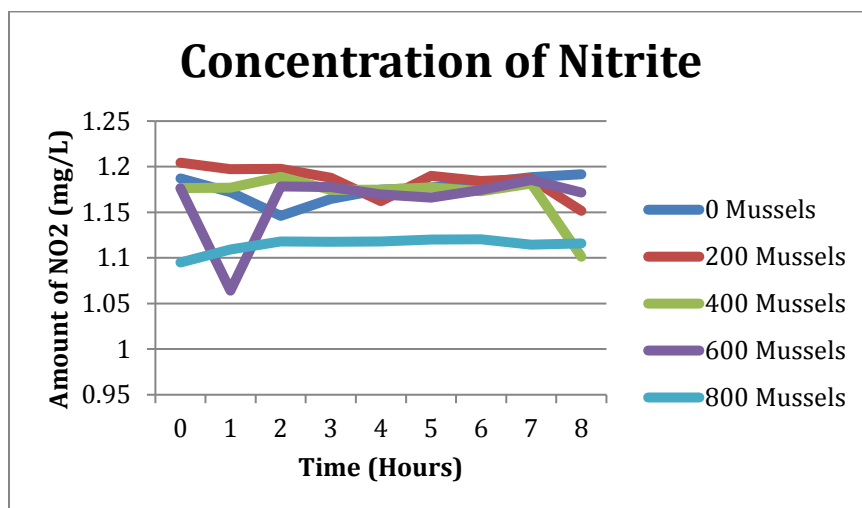


Figure 2: Analysis of nitrate (NO_3) concentration (mg/L) over a period of 8 hours with 5 densities of zebra mussels (0, 200, 400, 600, 800). The concentration of nitrate was not significant for time ($R^2 = 0.010$), but it was significant for density ($R^2 = 0.486$).

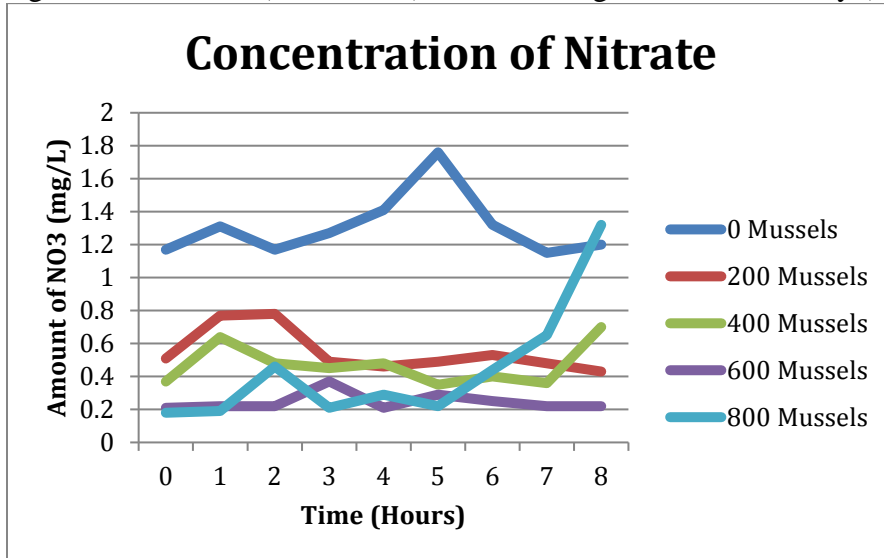


Figure 3: Linear regression analysis comparing the density of zebra mussels to the concentration of nitrate (NO_3) (mg/L) in the tanks. As the density of the zebra mussels in the tanks increased, the amount of nitrate decreased.

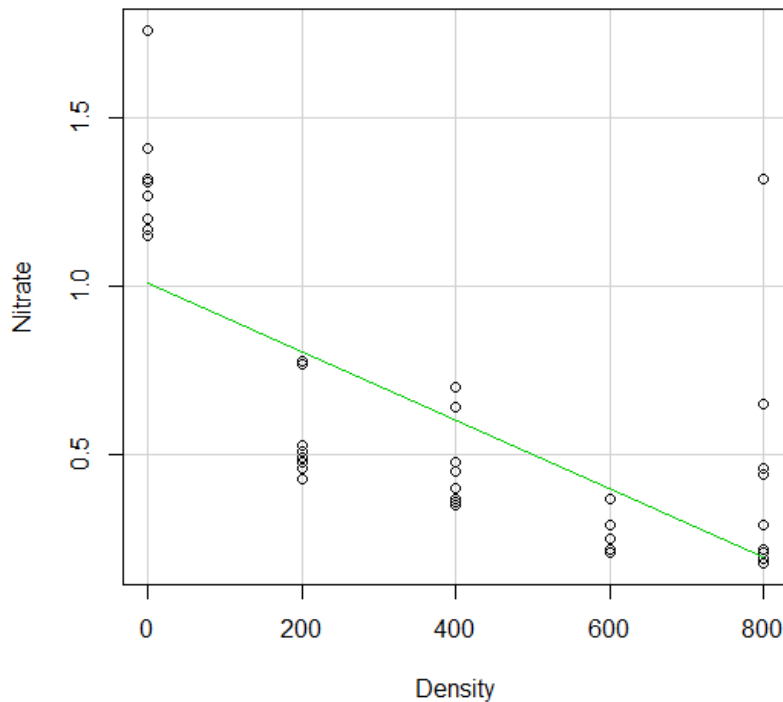


Figure 4: Analysis of ammonium (NH₄-N) concentration (ug/L) over a period of 8 hours with 5 densities of zebra mussels (0, 200, 400, 600, 800). The concentration of ammonium was significant for time ($R^2 = 0.39$) and density ($R^2 = 0.343$)

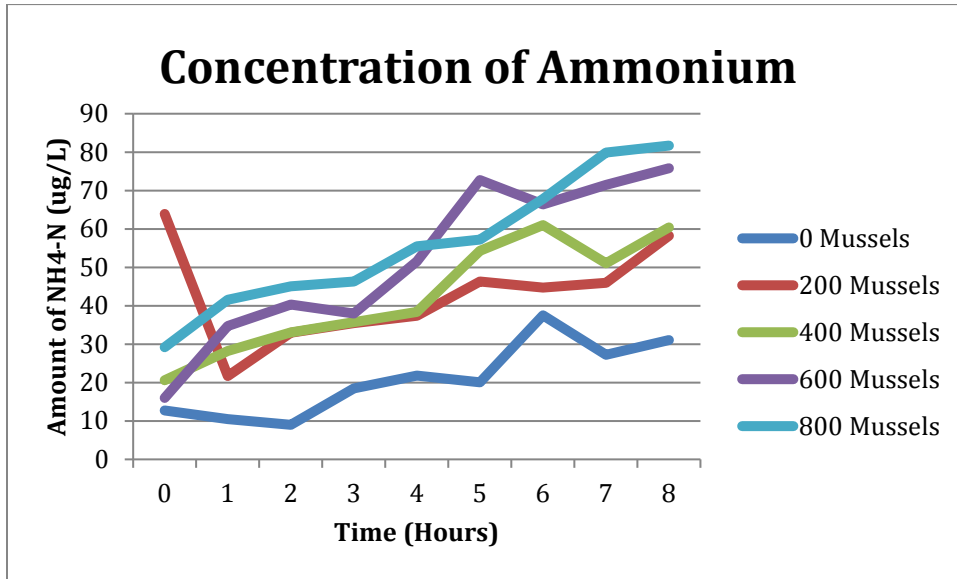


Figure 5: Linear regression analysis comparing the concentration of ammonium (NH₄-N) (ug/L) over the 8 hours that the zebra mussels were left in the tanks. The concentration of ammonium was found to increase over time.

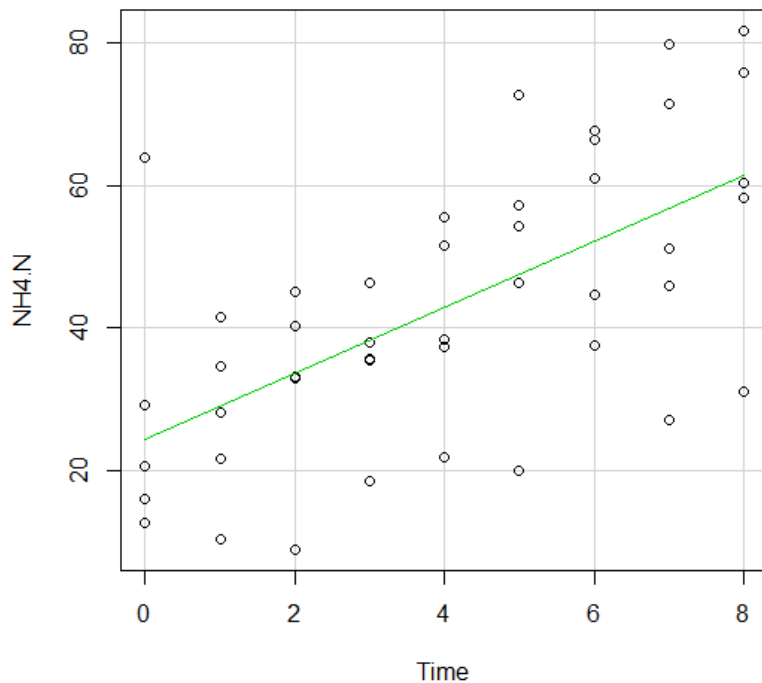


Figure 6: Linear regression analysis comparing the density of zebra mussels to the concentration of ammonium (NH₄-N)(ug/L) in the tanks. The concentration of ammonium was found to increase as the density of zebra mussels increased.

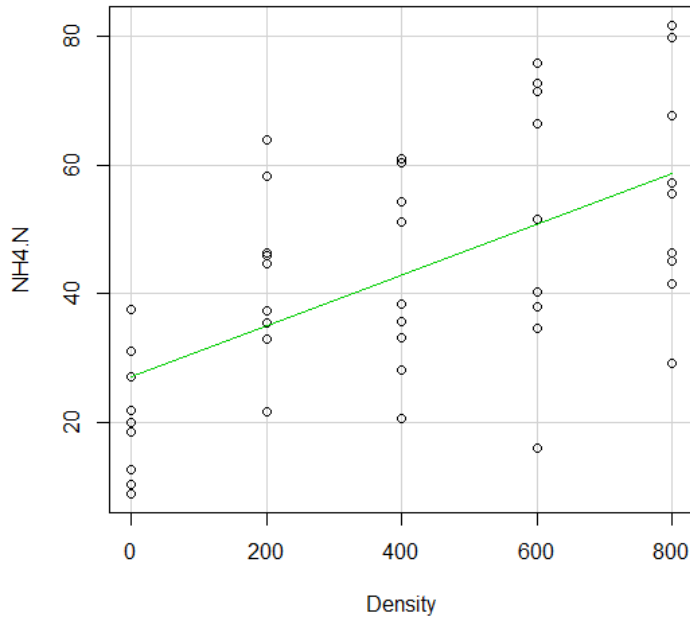


Figure 7: Analysis of calcium (Ca) concentration (mg/L) over a period of 8 hours with 5 densities of zebra mussels (0, 200, 400, 600, and 800). The concentration of calcium was not significant for time ($R^2 = 0.018$), but it was significant for density ($R^2 = 0.452$).

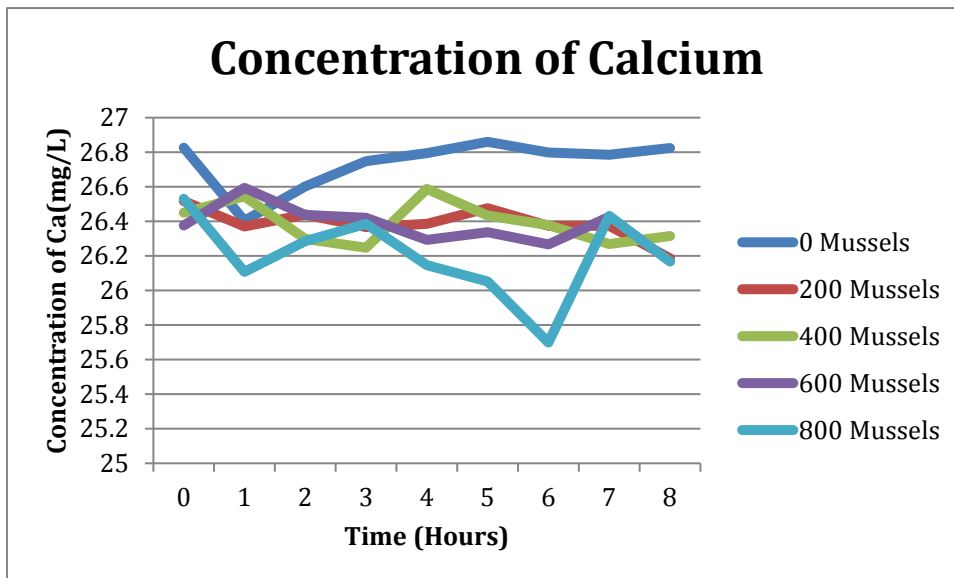
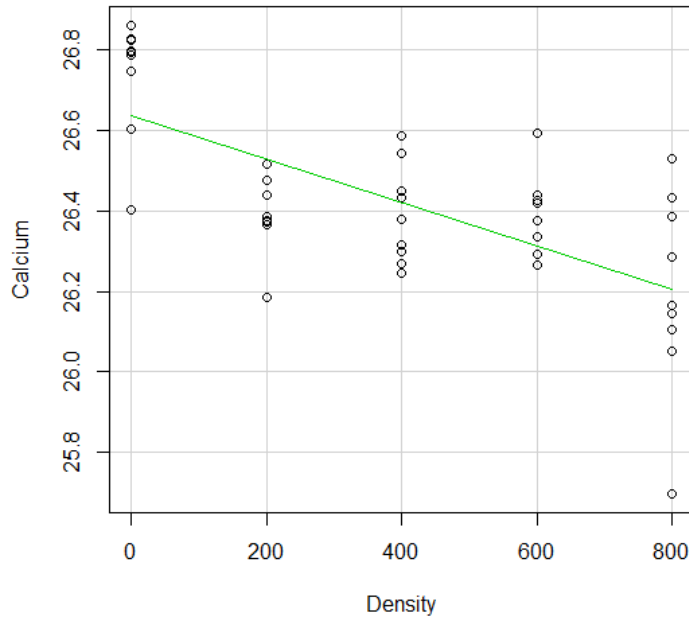


Figure 8: Linear regression analysis comparing the density of zebra mussels to the concentration of calcium (Ca) (mg/L). The concentration of calcium was found to decrease at the density of zebra mussels increased.



Our study showed that only 3 of the 14 nutrients we tested significantly changed over time or as zebra mussel density increased. The nutrients that changed in concentration were nitrate, ammonium, and calcium. The results showed that the change in nitrite concentration (mg/L) was not significant for time ($R^2 = 0.017$) or density ($R^2 = 0.046$) (Fig.1); however, nitrate concentration was significant for density ($R^2 = 0.486$), but not for time ($R^2 = 0.010$) (Fig. 2). The concentration of nitrate (mg/L) increased as the density of the zebra mussels in the tanks increased (Fig. 3). We found that the concentration of ammonium (ug/L) was significant for both time ($R^2 = 0.39$) and density ($R^2 = 0.343$) (Fig. 4) and had increased over time (Fig. 5) and with an increase in density (Fig. 6). The concentration of calcium (mg/L) was not significant for time ($R^2 = 0.018$), but it was significant for density ($R^2 = 0.452$) (Fig. 7) and was found to decrease with when zebra mussel density increased (Fig. 8). Thus, nitrate, calcium, and ammonium

concentrations increased significantly with an increase in zebra mussel density, but only the concentration of ammonium increased over time.

Discussion

The zebra mussels were alive and actively filtering throughout the duration of our experiment. We found that there was a more significant difference in nutrient levels between the tanks with different zebra mussel densities than nutrient levels over time. The difference in nutrient levels for chloride, fluoride, bromide, chlorophyll-a, silicate, sulfate, phosphate, sodium, potassium, nitrite and magnesium were not significant, so they were not important to this study. These nutrients may have increased or decreased in water bodies since the introduction of zebra mussels; however, they did not change significantly over a short period of 8 hours, unlike nitrate, ammonium, and calcium. We only rejected our null hypothesis that there will be no significant change in water nutrient levels over time for ammonium. Thus, there was a significant increase in the amount of ammonium (ug/L) over time. We rejected our null hypothesis that there would be no significant change in water nutrient levels between tanks containing different densities of zebra mussels for nitrate, ammonium, and calcium. Thus, the concentration of calcium (mg/L) and nitrate (mg/L) decreased significantly, but ammonium (ug/L) concentrations increased as zebra mussel densities increased.

The changes in nitrate and ammonium were potentially connected through nitrogen cycling in this system. Zebra mussels may significantly affect nitrogen transformation rates by excreting ammonium and altering the microbial food web structure. The cascading trophic effects caused by zebra mussels seem to be important to the transformations and fluxes of nitrogen in aquatic environments at regional and global scales (Lavrentyev, P.G., Gardner, W.S., Yang, L. 2000).

The change in nitrite concentration was not statistically significant, but the change in nitrate concentration was statistically significant as zebra mussel density increased. This is most likely because nitrate is the oxidized form of nitrite that zebra mussels may use; however, there is a lack of explanation for this in scientific literature. Decreasing nitrogen levels in water bodies due to the introduction of zebra mussels can affect aquatic plants. Nitrate is used by aquatic plants as their main nitrogen source for plant growth. Limiting the growth of primary producers can impact an aquatic ecosystems food chain (Stephen, L.J., Anthony, D.J., 2011).

Our results that indicated an increase in ammonium concentration are similar to a study showing that mean net ammonium production rate was significantly higher in zebra mussel treatments than control treatments (Lavrentyev, P.G., Gardner, W.S., Yang, L., 2000). The most important effect of zebra mussels on community nitrogen dynamics appears to be their direct excretion of ammonium (Gardner, W.S., Cavaletto, J.F., et al. 1995). Although we did not represent all of the complex processes that occur through the nitrogen cycle involving zebra mussels, we believe that the data offer insight into the effects that zebra mussels have on nitrogen dynamics in nature. For example, an increase in ammonium can cause eutrophication and acidification of the lake water. In turn, lakes infested with large amounts of zebra mussels may experience dead zones or a decline in aquatic organisms (Domingues, R.B., Barbosa, A.B., Sommer, Ulrich, Galvao, H.M., 2011).

It is not surprising that the concentration of calcium decreased significantly as zebra mussel density increased. Calcium is involved in muscular contractions, cellular cohesion, nervous functions, the maintenance of pH, and shell growth in the form of calcium carbonate for zebra mussels (Wojtal-Frankiewics, A., Frankiewicz, P., 2010). When they use a large amount of calcium, this leads to a decline in other organisms that need it for survival.

It's important to note that there was no significant change in chlorophyll-a levels in our tanks. A possible reason could be that zebra mussels are found to remove a large fraction of chlorophyll-a from inland lakes in October, but had a negligible effect on chlorophyll levels in June, July, August, and September when cyanophytes were abundant (Gardner, W.S., Cavaletto, J.F., et al. 1995). Thus, the seasonality of changes in nutrients caused by zebra mussels could be further explored not only with chlorophyll-a, but also with all the other nutrients tested in this study.

The results in this study were from our short-term experiment, thus the changing patterns in nutrient levels based on time and density may be more significant in a long-term study. However, if the zebra mussels were able to significantly impact the nutrient levels of our tanks with only a handful of zebra mussels placed in each, then the affects they have on larger bodies over a long period of time may be much greater. In future studies, we could also run more samples in the natural habitat of the mussels rather than in a laboratory setting.

Further, we focused on the negative impacts of zebra mussels on aquatic communities, but there are also some benefits. These include the ability of zebra mussels to filter out particles making the water clearer, providing refugia for macroinvertebrates, and moving energy through the food web (Ram, J.L., Palazzolo, S.M., 2008). More complete biological information on zebra mussel interactions with lower food web organisms and associated nutrient dynamics is needed to completely interpret their direct and indirect effects on nutrient cycling in Great lakes ecosystems (Gardner, W.S., Cavaletto, J.F., et al. 1995).

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