



# The Potential for Bivalve-Enhanced Denitrification in Long Island Sound

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## Introduction

One of the most common forms of environmental contamination is the addition to excessive amounts of nitrogen into an ecosystem by anthropogenic activities. The eutrophication of coastal waters and estuaries results in uncontrollable microalgae blooms, which result in hypoxic conditions when these blooms die.

The denitrification component of the nitrogen cycle provides a pathway for nitrogen species to leave an aquatic system and avoid the harmful effects of eutrophication. However, in many cases, the input of nitrogen from other sources far exceeds the capacity of the system to break it down. A proposed method to enhance denitrification is by utilizing the filtration and biodeposition of nutrients by bivalves. Bivalves filter nutrients out of the water column and deposit them on the surface of sediment as urine, feces, and pseudofeces. These biodeposited particles have been shown to stimulate microbial metabolism to promote coupled nitrification-denitrification. This study aims to examine the influence and potential for bivalve biodeposition on denitrification in Milford Harbor and other areas of Long Island Sound.

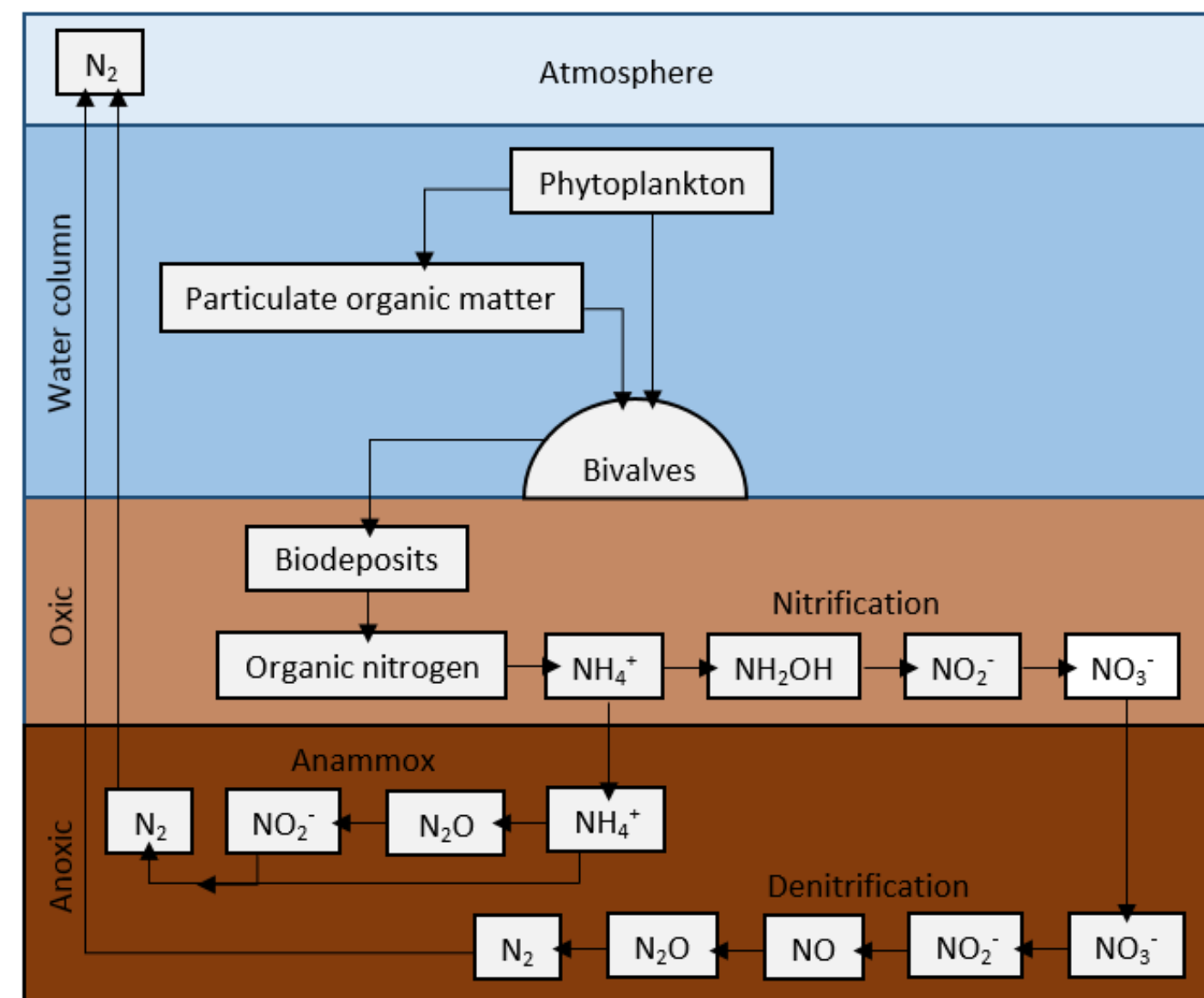


Figure 1. Shows the major processes of the nitrogen cycle, which include nitrification, denitrification, and anammox. Bivalves are believed to enhance denitrification by filtering nitrogen species out of the water column and depositing them on the surface of sediments as feces as pseudofeces (adapted from Kellogg et al. 2013).

## Materials and Methods

A series of experiments was conducted to study the influence of bivalves and biodeposition on the Milford Harbor ecosystem. In the first experiment, two raceway tanks housing scallops were allowed to accumulate biodeposits while measurements were taken every week for three weeks. The second experiment studied the influence of biodeposits alone, using a series of beakers containing sediments saturated with bivalve biodeposits. Four beakers were each incubated at 4°C, 10°C, and 25°C and exposed to both dark and light conditions. Measurements were taken several days apart over a span of four weeks.

Nitrogen species were measured using a nitrous oxide microsensor (Unisense A/S) and an autosampling nutrient analyzer to measure ammonia, nitrate, and nitrite (Seal Analytical, Inc.). The nitrous oxide microsensor is a Clark-type electrode which has been used in environmental studies of denitrification in lieu of other techniques due to its reliability and sensitivity. Once it is calibrated the microsensor requires no additional preparations, so real-time in situ measurements can be taken immediately and continuously.

## Results

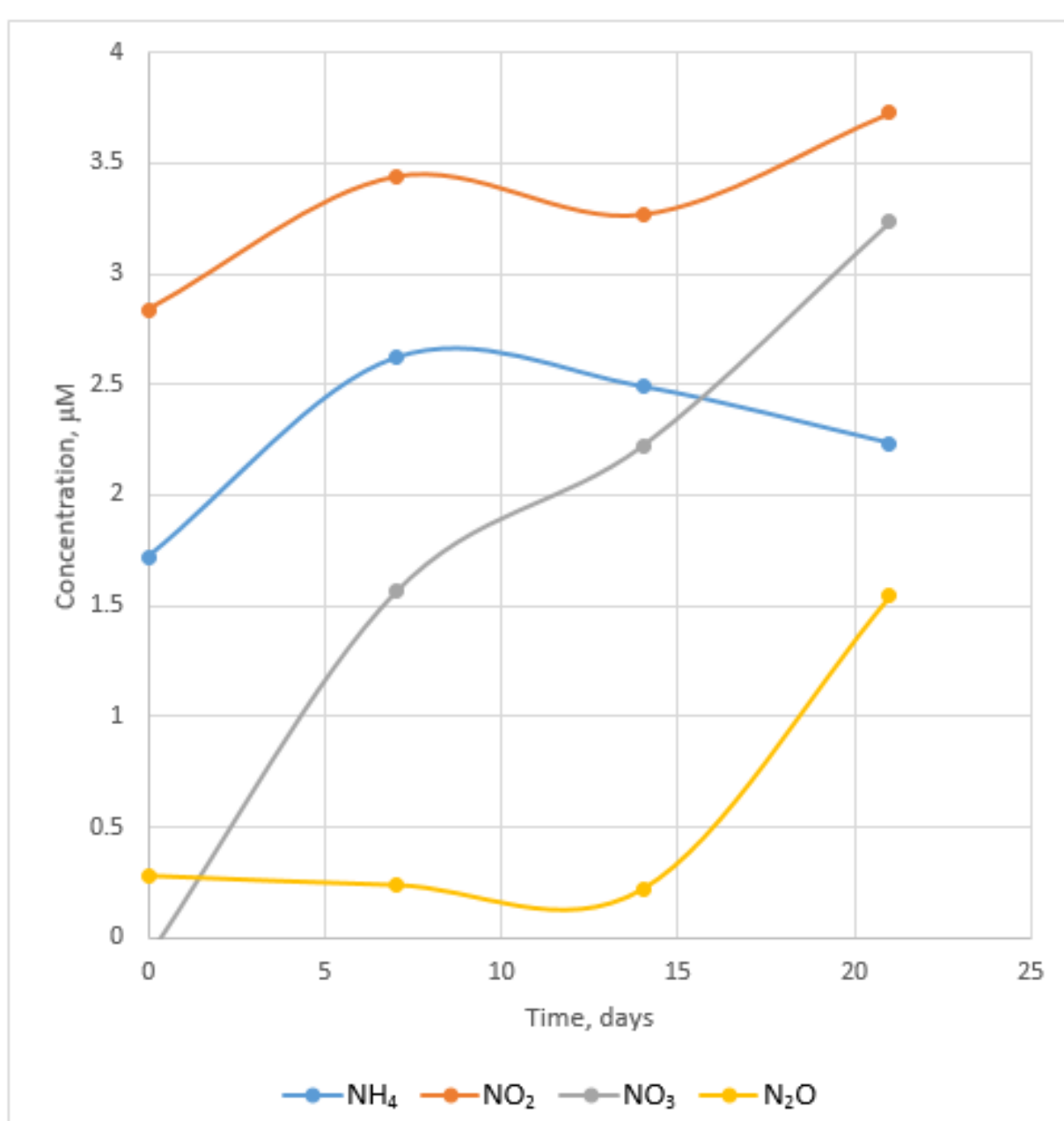


Figure 2. Shows the measurements of nitrogen species over time in outdoor tanks containing scallops that were allowed to accumulate biodeposits at 4-8°C.

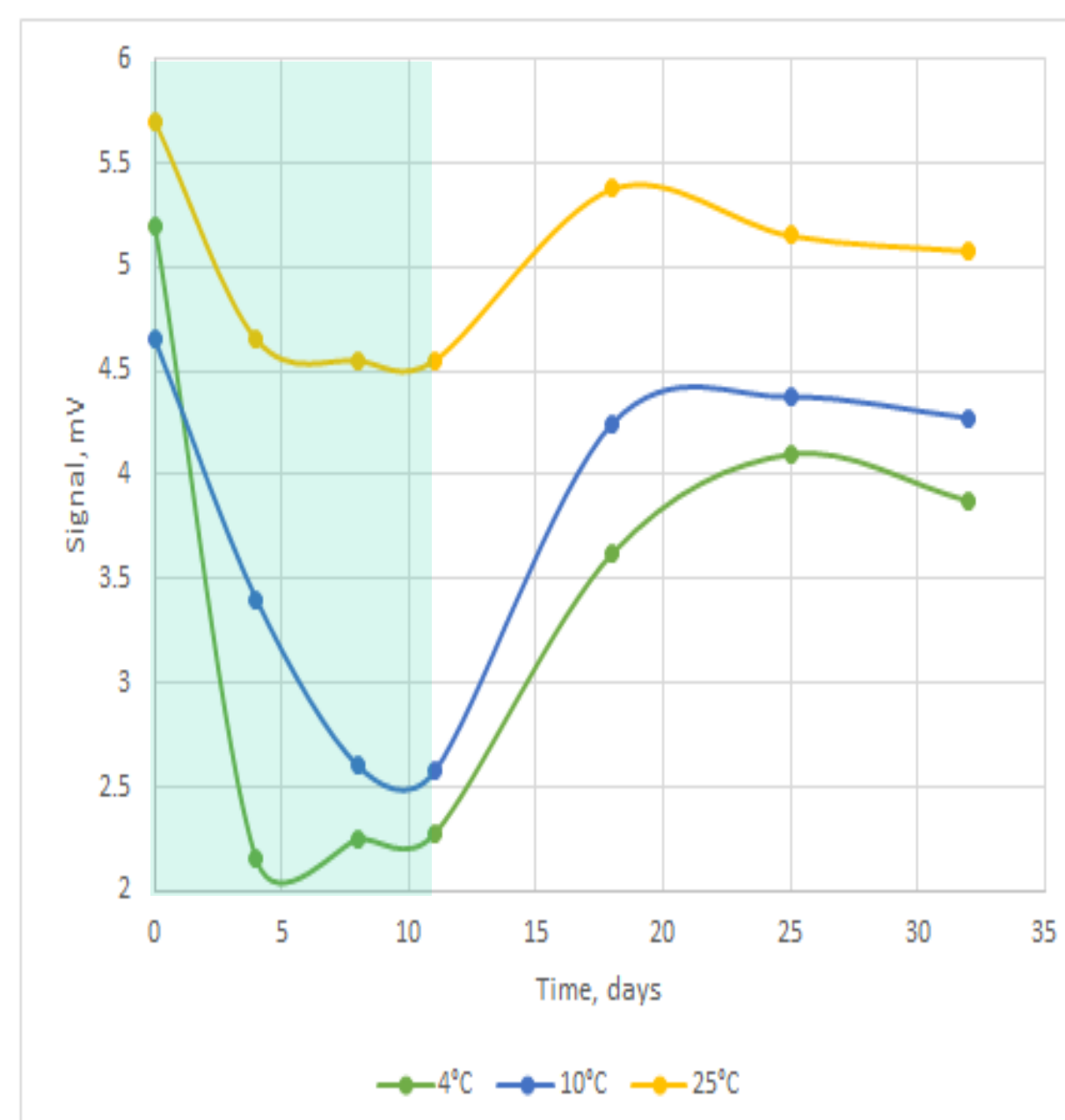


Figure 3. Shows the measurements of nitrous oxide over time in beakers containing sediment saturated with biodeposits at 4°C, 10°C, and 25°C. The shaded region denotes dark conditions. Measurements at 25°C were significantly different from those at other temperatures ( $p < 0.0001$ ).

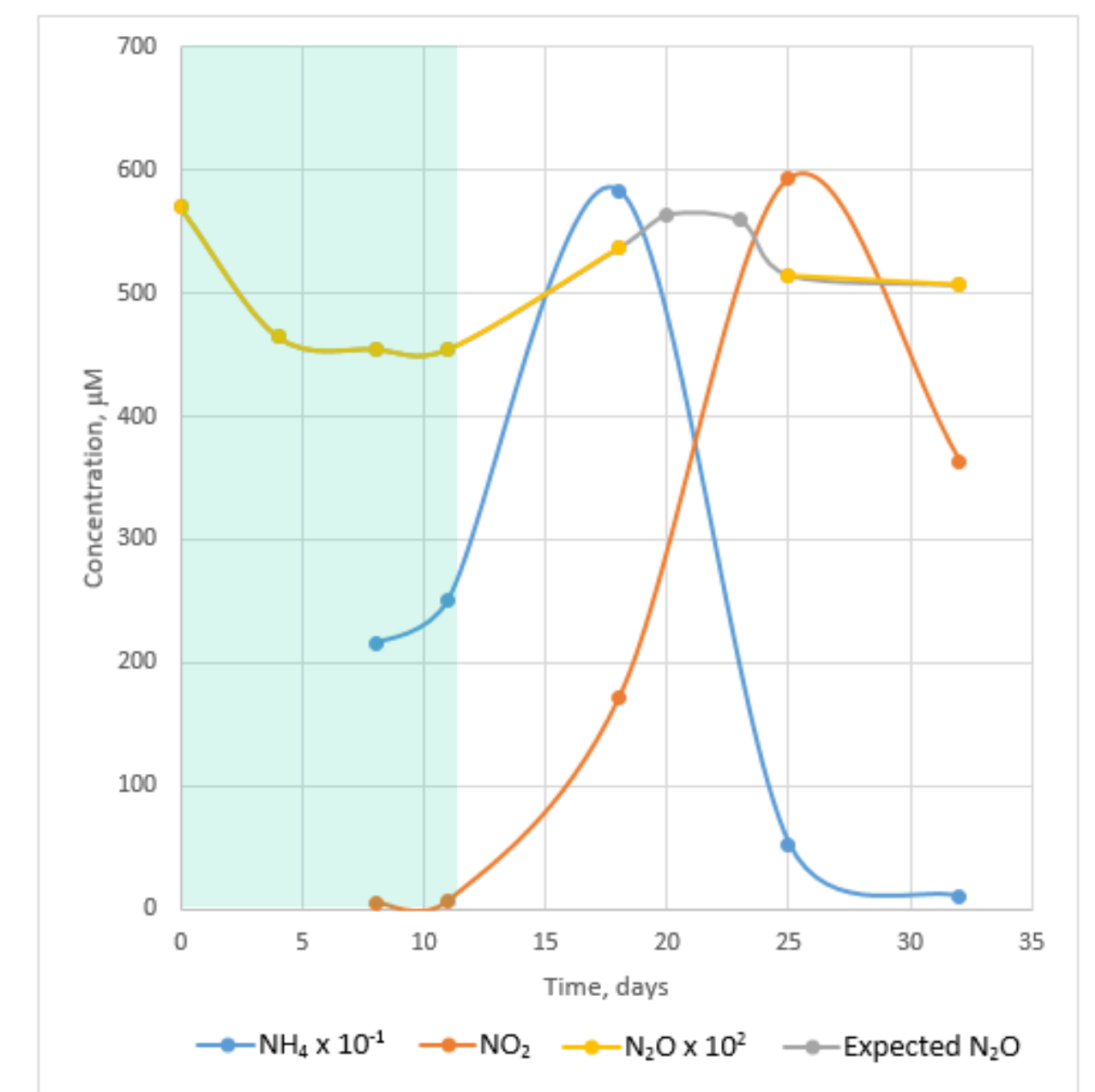


Figure 4. Shows the measurements of nitrogen species over time in beakers containing sediment saturated with biodeposits at 25°C. The shaded region denotes dark conditions. A calculated "expected N<sub>2</sub>O" has been added to extrapolate the measured data ( $R^2: 0.875$ ,  $p < 0.0001$ ).

## Discussion

Both the tank and beaker experiments showed the reduction of ammonia to nitrite, indicating that denitrification had occurred (figures 2 and 4). However the beaker experiments, which used sediment with a high concentration of biodeposits, saw a much higher turnover of nitrogen, with the average concentration of ammonia peaking at close to 6000µM. This finding suggests that the degree to which denitrification is enhanced by bivalves depends on the amount of biodeposits, and therefore on the amount of bivalves present. The discrepancies in studies assessing the promotion of denitrification may be able to be accounted for by the population density of the bivalves; if there are not enough bivalves present to deposit sufficient organic matter to drive denitrification processes, they will not appear to have any affect (Newell 2004, Kellogg 2013, Higgins et al. 2013).

The beaker experiments demonstrate the temperature dependence of denitrification (figure 3), which is likely related to the metabolic activity of microbial communities. The differences in the turnover of nitrogen species at varying temperatures of the same gross sample of sediment shows that microbial communities are in fact present, but they may not be metabolically active for a variety of reasons. This is one of several factors that affects nitrogen cycle processes which must be taken into consideration when assessing the influence of bivalves on denitrification. Additionally, the beaker experiments showed no significant difference in the concentration of nitrogen species when comparing the dark and the light phases of incubation. This supports the highly debated subject that light does not have a significant influence on denitrification (Risgaard-Petersen et al. 1994, An and Joye 2001).

The findings of this pilot study show that bivalve-enhanced denitrification is possible in the Milford harbor ecosystem and offers a potential solution to mitigate eutrophication in Long Island Sound.

## Acknowledgments

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