

Nitrogen Flow and Distribution in a Northern Michigan Dune System

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ABSTRACT

While the study of nutrient flow from terrestrial habitats to aquatic habitats is well known, little is known of the nutrient flow from aquatic to terrestrial habitats. The study of nitrogen flow from water to land in a northern Michigan dune system may provide an accurate measure of nutrient transfer. The purpose of this study was to determine the ratios of N-14 to N-15 in American beach grass in transects at various distances from the shore in hopes of explaining how aquatic nitrogen is distributed in a dune system. Samples of *Ammophila breviligulata* were taken from four transects running parallel to the shoreline at 0, 10, 20 and 30 meters from the shoreline. Washed up algae were also collected at random along the shoreline to act as a control. δ N-15 values, total percent nitrogen, and percentage of nitrogen obtained from aquatic or terrestrial sources were calculated for each distance. Results supported our hypothesis that the proportion of N-15 isotope would decrease as the distance from shore increased. Only samples taken at 0 meters showed a significant amount of aquatic nitrogen in proportion to terrestrial nitrogen. Physical and biological factors may account for the difference in distributions of the two nitrogen isotopes.

INTRODUCTION

The boundaries between terrestrial and aquatic ecosystems are often thought of in terms of how the terrestrial system mediates the transfer of nutrients and energy into the aquatic system (Helfield and Naiman, 2001). For example, the canopy of trees regulates the stream habitat by providing allocthonous organic matter via litterfall (Helfield and Naiman, 2001). However, these two systems, aquatic and terrestrial, work together in a cyclic process rather than a unidirectional process (VanderZanden Sanzone, 2004). Factors that influence aquatic inputs also contribute to terrestrial productivity and vice versa. Flooding, erosion and sediment deposition affect the soil composition and nutrient dynamics of the terrestrial zone, which in turn affect patterns of vegetation and species composition (Helfield and Naiman, 2001). In other words, the movement of energy and nutrients across habitat ecotones influences the receiving ecosystem, from population dynamics to ecosystem processes (VanderZanden and Sanzone, 2004). An observation of this process can be seen in the dune systems of northern Michigan.

Dune systems of northern Michigan are a part of the largest freshwater dune system in the world (Haas and Cline, 1972). Although they vary in their formation and topography, they all follow a general gradient from shore to land. As distance increases from the shoreline, the communities range from physically controlled (temperature, wind) to more biologically controlled (Brown and McLachlan, 1990). Vegetation that proliferates in this harsh environment is predominately aggressive pioneer plants such as *Ammophila*. *Ammophila*, American beach grass, exhibits rapid growth that can

outpace sand accumulation (Brown and McLachlan, 1990). The extensive horizontal and vertical rhizomes allow this plant to take advantage of the nutrients and water available underground (Hitchcock, 1935). *Ammophila* typically grows along coasts; however, it will grow on inland sites high in sand/saline content provided sufficient nitrogen (Voss, 1972).

Because nitrogen is a limiting factor for the growth and survival of life, its distribution across the dune system can portray nutrient transfer between systems. Stable isotope analysis can be used to quantify this transfer of nutrients across systems (Polis et al., 2004). For example, in a study testing the nitrogen isotope ratios in coastal mice, a high proportion of aquatic nitrogen, N-15, was found relative to the terrestrial nitrogen, N-14. This indicated that the mice were subsidized heavily by marine-based resources (Polis et al., 2004). Stable isotope analysis can show how allochthonous marine inputs ramify through the food web, from carnivorous insects to herbivores to plants (Polis et al., 2004).

In this study, we analyzed one aspect of the dune system food chain. Plants, in particular the American beach grass, represent the bottom of the food chain in dune systems; the first link exposed to aquatic nitrogen transfer. This transfer, whether by the insects, shore drift, or by windblown spray, is deposited in the soil and up taken by the plants (VanderZanden and Sandzone, 2004). The purpose of this study was to determine the ratios of N-14 to N-15 in American beach grass at various transects along the shore in hopes of explaining how aquatic nitrogen is distributed in a dune system. We suspected that an increase in the distance from shore would result in the decrease of

the N-15 isotope in proportion to the N-14 because more terrestrial nitrogen will be incorporated further from shore.

MATERIALS AND METHODS

The study site for this experiment was along the coastline of Sturgeon Bay in Emmet County, Michigan. This location has many dunes with much vegetation extending from the shoreline back into the forest. Four transects of dune grass running parallel to the shoreline were measured and marked. The first (groups A1-10) was directly along the shoreline, as close to the water as the dune grass had developed. The second (groups B1-10) was approximately 10 meters inland; the third (C1-10) approximately 20 meters inland; and the fourth (D1-10) approximately 30 meters inland, just beyond the first dune formation. Samples of dune grass were taken every 20 steps for 10 collections. The final range of dune grass sampled covered from 50-55 meters along the coast in all four transects.

The above ground stem material for five shoots of grass constituted each sample. The leaves were also taken in case additional analysis was needed. Each sample was cleaned and frozen in a -80 degree freezer. After frozen, all samples were freeze-dried in a Lyophilizer overnight where they underwent sublimation to rid the samples of water without losing any nitrogen. The samples were then crushed using the Spex 8000 Mixer/Mill. Each was put into a different vile and labeled. Samples were then prepped using the Thermo Finnigan Delta XP. This instantaneously combusted each sample with the help of pure oxygen at 1000 degrees C. This released the nitrogen into several

different types of organic molecules which were then placed in Costech EA 4010 where they underwent reduction via H_2 and Cu. The N_2 gases were then transferred to the Stable Isotope Mass Spectrometer (Inst. Precision for $N_2 = 0.3 \text{ ‰}$). This separated the isotopes by weight to determine the ratios of N-14 to N-15. Isodat 2.1 software was used for analysis.

Algae samples were collected as a control, representing the purely aquatic ratio. Washed up samples were collected at random along the shoreline, within the range of the dune grass transects. Each sample was cleaned and frozen in a -80 degree freezer. As with the dune grass samples, all were freeze-dried, crushed, and analyzed for ratios of N-14 to N-15. Calcium carbonate was digested in HCl.

In order to better understand the dune habitat including any physical factors that may affect the transfer of nitrogen, observations and measurements of our sample site were taken. We sampled vegetation characteristics and physical factors at 5 sites varying in successional status (foredune-front; foredune-crest; foredune-back; reardune near; reardune far). At each site we set out 1m X 1m quadrats, in which we made the following measurements: (1) soil temperature at 0, 5, and 40cm depth, (2) air temperature, wind velocity, and humidity at 0, 5, 40, and 100 cm above ground. To sample the vegetation present at each of these sites, we recorded in each of the 4 quadrats: (1) number of species present, and (2) % cover of each species.

Data Analysis

Once the ratios were obtained, a one-way ANOVA was applied to determine significant differences between all four transects. Independent t-tests were then applied

to determine differences between each transect. In order to understand the percentage of nitrogen contributed from the two sources, terrestrial and aquatic, to each sample, a mixing model of source partitioning was used

(www.epa.gov/wed/pages/models/stableisotopes/isotopes.htm). The aquatic source was the algae samples and the terrestrial source we used as the 30 meter transect.

Therefore, fractioning of samples at 0 meters, 10 meters, and 20 meters were obtained.

RESULTS

The one-way ANOVA test showed that the sigma values of the four transects were significantly different ($p=.003$, $F=7.283$). Independent t-tests showed that algal samples were significantly different from transects at 0m ($p<0.001$, $t=9.298$), 10m ($p<0.001$, $t=18.327$) and 20m ($p<0.001$, $t=15.469$). The transect at 0 meters was also significantly different from transects at 10m ($p=0.00798$, $t=3.574$), 20m ($p=0.00831$, $t=3.127$), and 30m ($p=0.0136$, $t=3.089$). Transects at 10, 20 and 30 meters were not significantly different from each other.

δ N-15 values were compared to the algae (a), the aquatic standard (Fig. 1). Because the δ value at 0m (b) is more positive than 10, 20, and 30m, that sample contains a higher amount of N-15 relative to N-14. The level of N-15 in samples taken at 10, 20, and 30 meters (c) contain statistically similar amounts as mentioned above.

The aquatic versus terrestrial nitrogen composition of each sample was calculated using mixed modeling analysis (Fig. 2). Samples taken at 0m contained about 23% of aquatic nitrogen (N-15) and 77% of terrestrial nitrogen (N-14). Samples

taken at 10m contained about -3% units of aquatic nitrogen and 103% units of terrestrial. Samples taken at 20m contained about -2% units of aquatic nitrogen and 102% units of terrestrial. The difference between nitrogen proportions at samples 10m and 20m were negligible. Only at sample 0m was a substantial difference observed.

A total percentage of nitrogen in *Ammophila breviligulata* was also calculated (Fig. 3). The highest levels of nitrogen were calculated at 30m ($\bar{x}=1.249$, $SD=0.118$), followed by 0m ($\bar{x}=1.207$, $SD=0.226$). A decrease was seen at the two samples in between these, 10m ($\bar{x}=0.97$, $SD=0.1$) and 20m ($\bar{x}=1.05$, $SD=0.081$).

Data collected on the micro-environmental factors of the dunes showed that just beyond the first dune, the vegetative diversity not only changed but also increased. Diversity changed from predominantly *E. arenensis*, *A. breviligulata*, and *Artemisia campestris* plants to *Salix cordata*, *Prunus pumila*, *Soldago spp.* and *Arctostaphylos uva-ursi*. Little change was seen in the air temperature and percent humidity at each portion of the dunes, although wind velocity was stronger closer to the shoreline.

DISCUSSION

Results from the dune system in Sturgeon Bay supported our hypothesis that the N-15 isotope would decrease in proportion to the N-14 as a result of an increase in distance from shore. According to our data, only the dune grass along the shoreline took up a significant amount of the aquatic nitrogen deposited in the soil by insects, shoredrift or windblown spray (VanderZanden and Sanzone, 2004). This supports the findings that increased δ N-15 values were found along the river corridors of streams

(Helfield and Naiman, 2001). The levels of N-15 at 10, 20 and 30 meters showed no variation and were apparently 100% terrestrial. This suggests that the movement of nitrogen from aquatic to terrestrial ecosystems occurs over a narrow ecotone.

The dunes closest to shore are characterized by strong winds, erosion, high rates of evaporation and low nutrient availability (Lichter, 1998). This can be observed in our data at 10 and 20 meters from shore, where the first dune is formed. A higher percentage of nitrogen was observed near the shoreline thanks to aquatic input. This aquatic input may therefore act as the stabilizing factor of this site. Without it, the shoreline would most likely be an extremely nutrient-poor area.

The accumulation of organic matter can improve the nutrient levels in the soil of a dune system, given a sufficient distance from shore (Lichter, 1998). At 30 meters from the shore, just beyond the first dune, the total percentage of nitrogen began to show a significant increase. Paralleled to this increase in nitrogen, the vegetative diversity increased to include plants such as *Salix cordata*, *Prunus pumila*, *Soldago spp.* and *Arctostaphylos uva-ursi*. The rise in nitrogen as well as the change in plant species may illustrate the steps of dune succession, changing from a physically controlled habitat to a more biologically controlled habitat (Brown and McLachlan, 1990). At 30m, accumulation of nutrients may be due to decreasing wind velocities as noted in our analysis and sand movement as well as decay and decomposition of terrestrial organisms.

These findings have potentially important implications for how we understand the interactions between aquatic and terrestrial ecosystems. Any action that would

deplete aquatic nutrient levels would in turn affect the nutrient levels along the shoreline. Although this study did not analyze higher trophic levels of herbivores and carnivores, depleting nutrient levels would most likely affect all links in the food web. Further studies needed to fully understand the cyclic flow of nitrogen in the dune system would include a more detailed study of the nitrogen levels between 0 and 10 meters. Because our study only tested at 0m and 10m, the composition of nitrogen between these two transects is unclear. Additionally, determining nitrogen isotope levels in the herbivores and carnivores present, and specifically in the aquatic insects whose decay may contribute substantially to the isotope levels found in beach grass.

Table 1. Summary of Isotope Analysis

Transects	n	Avg. d15N	std
algae	3	1.65033895	0.46838282
0m	5	-2.7680638	0.87376972
10m	5	-4.3063026	0.40314507
20m	5	-4.2516625	0.60183249
30m	5	-4.1131216	0.42962174

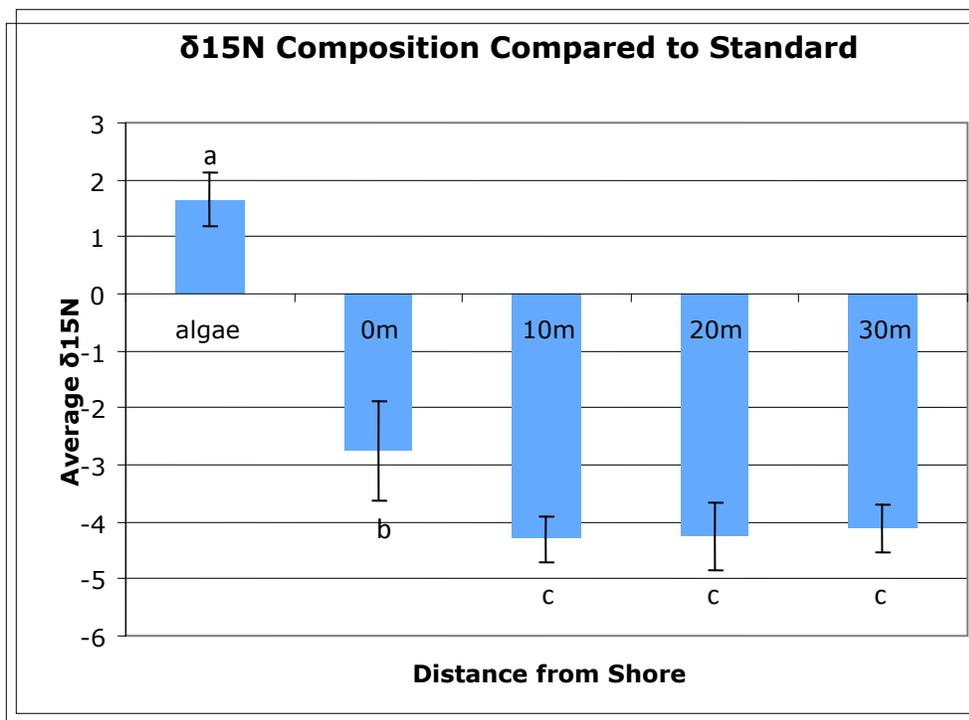


Fig. 1. Changes in δ N-15 values at varying distances from shore. Bars are labeled as a, b, and c to show the three statistically significant sigma values.

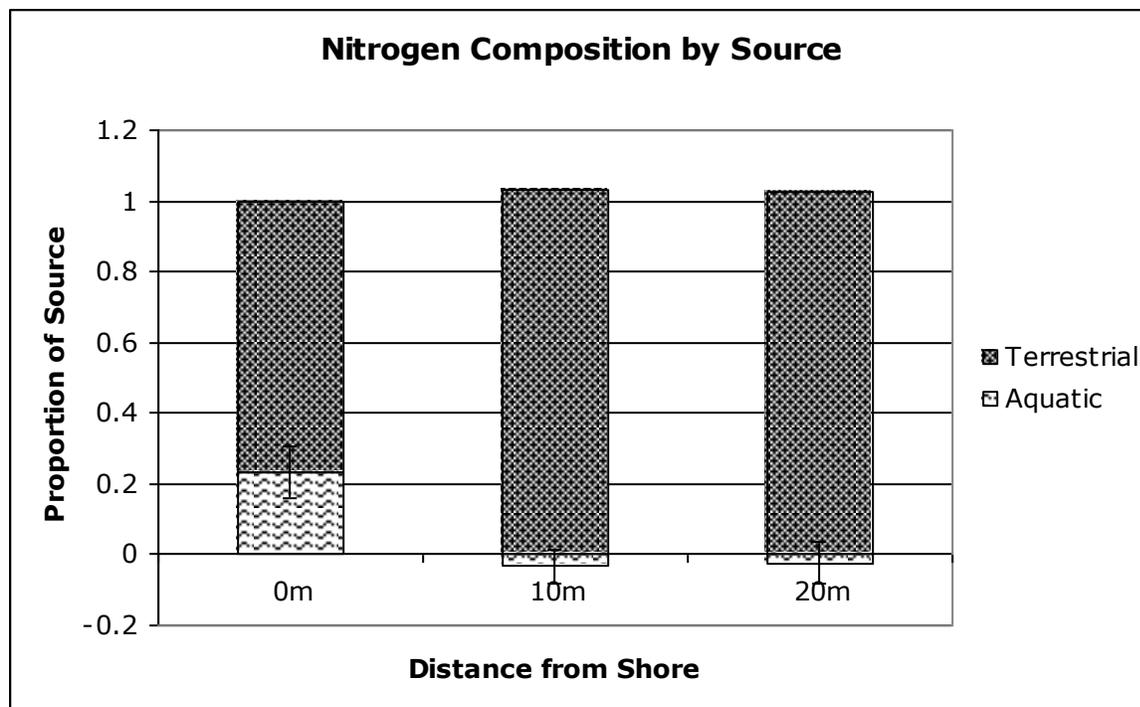


Fig. 2. Aquatic contribution of nitrogen to a specific sample to the terrestrial contribution of nitrogen to that same source.

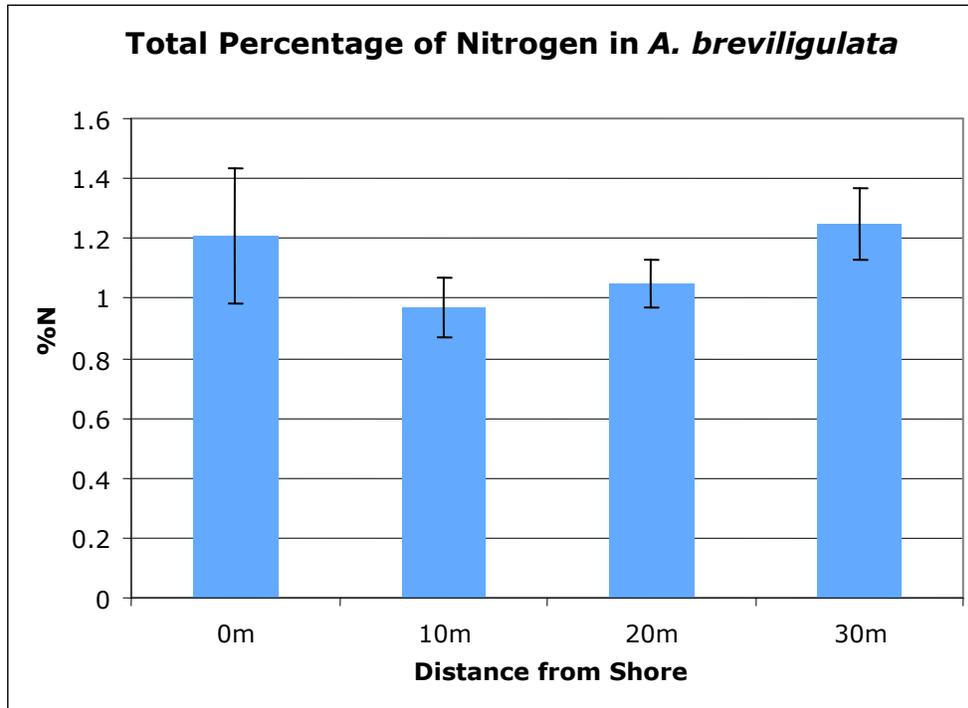


Fig. 3. The total percentage of nitrogen (all isotopes) found in *Ammophila breviligulata* at the four transects.

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