THE ROLE OF BRYOPHYTES IN STREAM ECOSYSTEM FUNCTIONS

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Abstract. Nitrogen is an important chemical nutrient for primary producers in streams. These primary producers, such as bryophytes and algae, regulate nutrient cycling. In the streams of the Hubbard Brook Experimental Forest, New Hampshire, the distribution of bryophytes and their role in regulating nitrate were examined. Across four watersheds, surveys were conducted to study the distribution and abundance of bryophytes. Various factors were examined, but substrate size was found to be the most important in influencing their distribution. The streams of the HBEF valley experience annual increases of discharge, causing small substrates to overturn. This suggests that bryophytes were more abundant on larger substrates because they overturn less.

Nitrate releases were conducted to study the effect of the abundance of the bryophyte, *Scapania undulata* on the rate of nitrate uptake in experimental channels. Two high discharge releases and two low discharge releases were performed to represent the different discharges in the streams throughout the year. A significant difference between the uptake of nitrate at low discharge and high discharge releases occurred, indicating a relationship between nitrate uptake and discharge. The bryophytes in the low discharge releases affected nutrient concentration more than in the high discharge releases. In slower water flow, there is more contact time between a nitrogen molecule and a bryophyte mat. Looking at the distribution of bryophytes and their role in nitrogen cycling, their location could be an important component of stream ecosystems.

INTRODUCTION

Nitrogen is an important chemical nutrient for primary producers in streams. In many streams, N increases as a result of human activities, such as increases in fertilizers, pollution, and deforestation. Biological transformations (algal and microbial uptake) of nitrogenous compounds in streams can alter both the form and timing of export to downstream ecosystems (Richey et al., 1985). This increase in nitrogen can affect concentrations downstream impacting algal growth and denitrification.

Aquatic primary producers (algae and bryophytes) and terrestrial detritus are primary sources of organic matter available to consumers in streams. These producers, such as detritus, during decomposition, provide nutrients for microbes and invertebrates. The epiphytic microbes provide food for invertebrates as well. The shade from heavy forests can cause light to be limiting to primary production and it is anticipated that the majority of biological activity could be due to microbial communities (Richey et al., 1985). However, relative to algae, little work has been done on bryophytes.

A number of studies suggest that bryophytes can be important contributors to total stream metabolism, nutrient cycling, and food web interactions in streams (cited in Arscott and Bowden, 1998). Bryophytes may serve as a direct food source for some invertebrates. Bryophytes also serve as a substrate for epiphyton and periphyton attachment and growth (Steinman and Boston, 1993); this is important because bryophytes may play an indirect role in nitrogen uptake by providing more surface area for algae that use nitrogen for metabolic purposes.

The distribution and abundance of bryophytes can be another important factor in nutrient cycling. The abundance of bryophytes may contribute to nutrient regulation in streams. There are many important factors that can control the distribution and abundance of bryophytes. Annual fluctuations in rainfall can cause sporadic increases in the water level and flow. Smaller rocks can be easily pushed over from increased water flow preventing them from being a suitable substrate for bryophytes to colonize. Suren and Ormerod (1998) studied bryophytes at different elevations in Nepal and concluded that substrate stability was the dominant factor in the distribution of bryophytes in streams. The annual fluctuations in water flow can increase or decrease the concentration of nitrogen molecules interacting with the bryophytes.

The purpose of this study was to 1) understand the factors that regulated the distribution and abundance of bryophytes and 2) determine the role of bryophytes in nitrate uptake. The study consisted of two parts: 1) a study of the effect of bryophyte abundance on the rate of nitrate uptake in experimental channels and 2) a survey of the distribution of bryophytes in four streams. The results of the nitrate uptake and the survey then were combined to make predictions about where bryophytes have the greatest potential effect on stream nitrate concentrations.

MATERIALS AND METHODS

Study Site

The study was conducted at the Hubbard Brook Experimental Forest (HBEF), which is in the White Mountains of New Hampshire and is part of the Long-Term Ecological Research network. The forest at HBEF was logged in about 1920 and, since that time, several small watersheds have been experimentally cut. Streams draining Watersheds 2, 4, 5, and Bear Brook were surveyed for bryophyte abundance and distribution. Watershed 2 was experimentally clear-cut 35 years ago and treated with herbicide for three years following. Watershed 4 was experimentally cut 25 years ago and Watershed 5 was cut 15 years ago. The Bear Brook watershed has not been altered since 1920.

Surveys

Between June 27 and July 3, 2000 each of the four streams in the watersheds were surveyed for habitat, substrate, bryophyte absence or presence, and canopy cover. In each stream, three longitudinal (i.e., situated along the thalweg) point transects were surveyed. Transects in each stream were 30m long and were separated by 50m. Every 0.5m, a finger was placed in the water and habitat type, substrate (Table 1), and the presence or absence of bryophytes was recorded when touched. Canopy cover over the stream was measured every 3m using a spherical densiometer.

Nitrate Releases

Artificial, flow-through channels, provided by Kate MacNeale, were used to examine the effect of bryophyte abundance on the rate of nitrate uptake. The channels were constructed of plastic rain gutters approximately 10 feet in length and located adjacent to Bear Brook. Stream water flowed from Bear Brook by gravity into a tub and then was distributed to the channels. Nitrate was added to one set of five channels and another five channels served as nutrient control channels. Above the head of the control channels, stream water dripped via tubing connected from the tub of stream water. Similarly, nitrate was dripped above the head of the treatment channels via tubing from a tub filled with nitrate and stream water.

The liverwort, *Scapania undulata* was the bryophyte utilized in the releases. The bryophytes collected for the channels remained attached to rocks to prevent damage to the bryophyte community and subsequent leaching of nutrients. All bryophyte samples were collected from watershed 3 for consistency. All rocks without bryophytes were collected from Bear Brook. Within each set of channels, different abundances of *Scapania undulata* were used for the nitrate releases. In the treatment channels, eight rocks of similar sizes were placed in every channel with the exception of the control channels. In this set, the control channel contained no rocks or bryophytes. One channel contained rocks only, while the other channels contained different abundances of bryophytes on the rocks. Channels with bryophytes contained a high abundance of bryophytes on the rocks, a medium abundance of bryophytes on the rocks, or a low abundance of bryophytes on the rocks. The percent cover of bryophytes on the individual rocks and within the channels was estimated visually. The set of control channels located adjacent to the treatment channels were set up the exact same way as the treatment channels. Stream water alone dripped into the set of control channels.

The concentration of nitrate in the water samples of Bear Brook was measured using an ion chromatograph. For the treatment channels, the target concentration of nitrate released (initially 120 ug/l) was five times the background concentration of Bear Brook. The concentration was chosen to be high enough to detect changes in nitrate, but not so high as to be above levels that occurred during extreme natural conditions (Likens *et al.*, 1977). The nitrate was mixed in a tub of stream water and dripped via tubing into the treatment channels. Four releases were conducted between July 18 and August 5, 2000. Two releases were at low discharge (approximately 2.5 ml/s) and two releases were at high discharge (approximately 27.5 ml/s). Each release was conducted for 30 minutes and 10 water samples (1 for each channel) were taken just below the enrichment site and 10 water samples (1 for each channel) were taken at the downstream end of each channel. Nitrate concentrations of the water samples were determined using an Ion Chromatograph. For each release, new rocks were collected from Bear Brook and new bryophytes were collected from watershed 3. Following each release, the rocks and bryophytes were removed and the channels were scrubbed with a sponge to decrease algal growth between the releases. All releases were conducted approximately one week apart.

To ensure that there was similarity in rock size and a difference in bryophyte abundance between the channels, all of the bryophytes and rocks from the channels were measured. Bryophytes were scraped from rocks and were dried in an oven at 60°C for 12-16 hours; samples were then weighed to the nearest gram. Weighed samples were combusted at 500^oC and were re-weighed for the determination of ash-free dry mass. After the removal of bryophytes, the surface of each rock was covered in aluminum foil. The weight of the aluminum foil was measured and the surface area of the rocks was estimated by regression, according to the following relationship (Equation 1):

$$X = (Y - 0.001576)/(0.004453) \qquad (1)$$

where X is the area (cm^2) and Y is the mass (g) of the aluminum foil.

RESULTS

Bryophyte Surveys

Bryophyte abundance was similar across the four streams. Bear Brook contained a higher frequency of occurrence of approximately 31% compared to watersheds 2 and 5, which contained a frequency of approximately 17% (Figure 2). Figure 3 represents bryophyte frequency of occurrence verses substrate size in watershed 4 (refer to Table 1). This watershed was fairly representative of the other watersheds surveyed. The frequency of bryophyte occurrence increased as substrate size increased. Bryophytes were found on approximately 48% of the bedrock while only 5% of the gravel substrates sampled contained bryophytes. However, bedrock only constituted approximately 12% of the sampled streambed. Boulder, large cobbles and medium cobbles occurred more frequently (Figure 4). Within each of the ten transects spread over the four streams, bryophyte cover was significantly related to percentage of large substrates in each transect. There was ~40% bryophyte cover on 78% of the larger substrates, while only 35% of the large substrates contained 7% bryophyte cover (Figure 5). A greater percentage of bryophytes inhabited the larger substrates.

Nitrate Releases

Bryophyte densities between the channels were represented in release 4 data, which was characteristic of all the releases. The channel with high bryophyte density contained approximately twice the amount of bryophytes as the medium bryophyte abundance channel. The medium bryophyte abundance channel contained approximately twice the amount of bryophyte abundance as the low bryophyte abundance channel (Figure 6).

Rock sizes were also measured to ensure consistency in sizes between the channels. The total surface area of the rocks in each channel of the treatment set and control set for all of the releases conducted were measured (Figure 7). The rock surface areas were fairly similar ranging from approximately 1300 cm^2 to 2050 cm^2.

In the first release at low discharge, there was a greater uptake of nitrate at higher bryophyte densities. As bryophyte densities increased, nitrate uptake increased (Figure 8). There was less nitrate uptake in the control channels as well as in the channels of lower bryophyte density. However, there was an uptake of nitrate in the control channels. The control channels with rocks had a higher nitrate uptake than the control channel without rocks.

The second release at low discharge did not produce similar results as the first release. The control channels were consistent with increasing nitrate uptake by the rocks but as bryophyte density increased, nitrate uptake decreased (Figure 8).

Two releases were also conducted at higher discharges of approximately 30 ml/sec. Compared to the uptake of nitrate in the low discharge releases, uptake was significantly lower in each case. There was no correlation between bryophyte abundance and nitrate uptake. The nitrate uptake in all bryophyte channels was significantly lower. However, there was greater uptake of nitrate in the rock and empty control channels. In the first release at high discharge, the rock channel had a higher nitrate uptake than the empty channel, similar to that in low discharge releases. But in the second high discharge release, the empty channel had a higher nitrate uptake than the rock channel (Figure 9).

DISCUSSION

Watershed 4 data, which was fairly representative of the other watersheds surveyed, displayed a positive correlation between increased bryophyte abundance and large substrates (Figure 3). This was attributed to their stability, which helped minimize biomass losses from sloughing and abrasion. Steinman and Boston (1993) studied the ecological role of aquatic bryophytes in a woodland stream and found that with respect to substrate type, bryophyte biomass was greatest on boulder and bedrock substrates and intermediate on cobble, wood, and gravel. This increased bryophyte frequency of occurrence verses type of substrate may also be related to seasonal patterns of discharge in the streams. Watershed 3 annual discharge data indicates that these streams have high variability of flow due to episodic rainfall with sporadic periods of increased discharge (Figure 10). The annual fluctuations can easily cause small substrates to overturn which helps support our results that bryophytes are more abundant on larger substrates. Various factors in the surveys were examined to reveal that substrate size and stability were most important in influencing bryophyte abundance and distribution. Although canopy cover was measured, it did not show a significant effect on bryophyte distribution (Figure 11). Larger substrates were associated with greater light availability.

For the nitrate releases, discharge seemed to have a significant influence on the bryophyte's ability to take up nitrate. At slower flow rates, the nitrate molecules in the water have a longer amount of time to come into contact with the bryophyte mats. Richey et al. (1985) found that during extended periods of high discharge, the contact time between a nitrogen molecule and in the water column and the streambed will be reduced, while during periods of low flow, contact time, and hence uptake and transformation, will be enhanced. Bryophytes may act like sponges taking in nutrients that flow over them. Soares and Pearson (1997) inferred that bryophytes growing in low N areas take rapid advantage of exogenous N sources, whereas bryophytes already growing in areas with high available N will incorporate less N in proportion to availability. At higher discharge rates, the nitrate molecules flow over the bryophytes too quickly. This supports our data of the increase in nitrate uptake at lower discharges. However, in the control channels there was a greater than zero nitrate uptake. Even after the control channels with no rocks or bryophytes were scrubbed down before each release, algae and biofilm grew on the surface of the channels rapidly, within a day. Algae and biofilm formed on the rocks, which could have contributed to the increase in nitrate uptake at lower discharges. In Bear Brook, nitrate concentrations provide a ready source of nitrogen for autotrophic organisms, which aid in nitrogen transformations (Richey. et al, 1985). The rocks without bryophytes were collected from Bear Brook, whereas the bryophytes were collected from watershed 3. This could have caused a discrepancy in the data because the watersheds contained different concentrations of nitrate, which can affect the algae and biofilm that inhabit them. The samples collected for the releases were not uniform between the channels.

The inconsistency of the data between the two low discharges and the two high discharges hinders the formation of a definitive conclusion. However, it is clear that rates of nitrate uptake were higher at low discharge than at high discharge. Since only four releases were conducted, it is not conclusive that bryophytes alone remove nitrate from the streams. Although it was evident that these channels took up nitrate, the destination of the nitrate is unknown. Under certain conditions, some bacteria found in bryophyte mats can cause denitrification. At lower discharges, water slows down resulting in less mixing enabling bacteria to easily form.

Relative to the streambed, the channels were less variable in size and substrate. This must be taken into account when comparing the nutrient cycling in the channels to the nutrient cycling in the streams. The channels were only 10ft long and in the time it took for a nitrate molecule to pass over the bryophyte mats and dump back into the streambed, it may not have had as significant of an impact as it would in the streambed. The nitrate molecules were also limited to where they could go. In the streambed, nitrogen can sink into the soil, be taken up by bryophytes, consumed by algae, or be released into the atmosphere through denitrification. In the channels, soil was absent and the channels were too short for denitrification to occur. The distance between the bryophytes was controlled in the channels. This would not be the case in a stream, where bryophytes usually occur in clumped masses on large substrates and their distribution is influenced by the separation of these large substrates. However, flow rates were controlled to model flow rates of annual discharge. This seemed to have a significant effect on the nitrogen cycling in the channels. In relation to the surveys, bryophytes were more abundant on larger substrates. But in the channels, the only substrates that could fit were small cobbles. However, the ratio of substrate size to stream size was fairly similar to the ratio of substrate size to channel size.

Nitrogen cycling in streams can be influenced by the distribution and abundance of bryophytes. Bryophytes were found to be more abundant on larger, more stable substrates. Larger substrates were also more abundant in the watersheds. However, in periods of low discharge, bryophytes may not be involved in regulating nutrient cycling because water is not flowing over them. In this case, bryophytes may only be part of the cycling if they are located close to the water flow, such as in ponds or on smaller substrates. However, even in periods of increased water flow, bryophytes are more abundant on larger substrates where cascades and riffles caused faster water flow. Since high discharge in the channels resulted in less nitrate uptake, it can be indicative that bryophytes do not contribute significantly to the uptake of nitrate from the streams.

Time constraints prevented us from performing repetitive nitrate releases. However, this study is a useful pilot study. This experiment and past research did show strong correlations between bryophyte abundance and substrate size. For the channel releases, a significant difference between the uptake of nitrate in low discharge and high discharge occurred indicating a relationship between discharge and nutrient uptake in streams. Looking at distribution of bryophytes and their role in nitrogen cycling, their location could be important in streams with harmful nitrogen levels. With this study, bryophytes are found to be an important component of stream ecosystems, but their role in stream nutrient cycles remains largely unexplored.

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APPENDIX

TABLE 1. Substrate Size (mm) and Frequency of Occurrence (%) averaged over the 3 transects for each watershed.

SUBSTRATE	DIAMETER (mm)	SUBSTRATE FREQUENCY OF OCCURRENCE (%)			
		Bear Brook	Watershed 2	Watershed 4	Watershed 5
Sand	0.25-2.00	1.09	0	0	4.84
Gravel	2.00-50.0	9.84	17.74	12.6	53.23
Small Cobble	50.0-100	10.93	22.58	12.57	59.68
Medium Cobble	100-150	10.38	6.45	13.11	46.77
Large Cobble	150-200	26.23	6.45	16.94	32.26
Boulder	200-2000	32.24	11.29	26.23	56.45
Bedrock	>2000	8.74	32.26	12.57	29.03

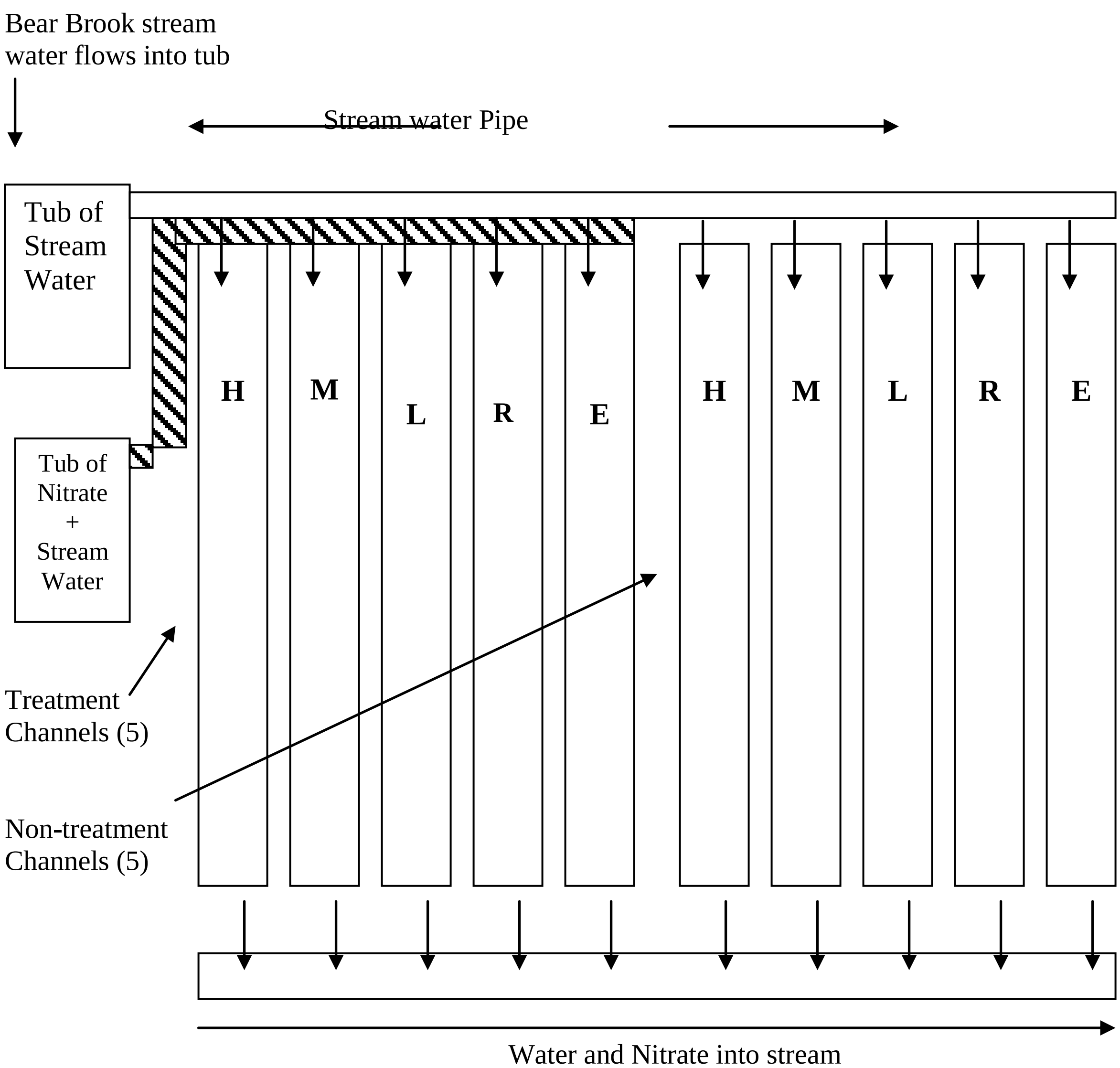

FIGURE 1. Overhead view of artificial channels utilized for nitrate releases.
H: High bryophyte abundance
M: Medium bryophyte abundance
L: Low bryophyte abundance
R: Rocks only
E: Empty

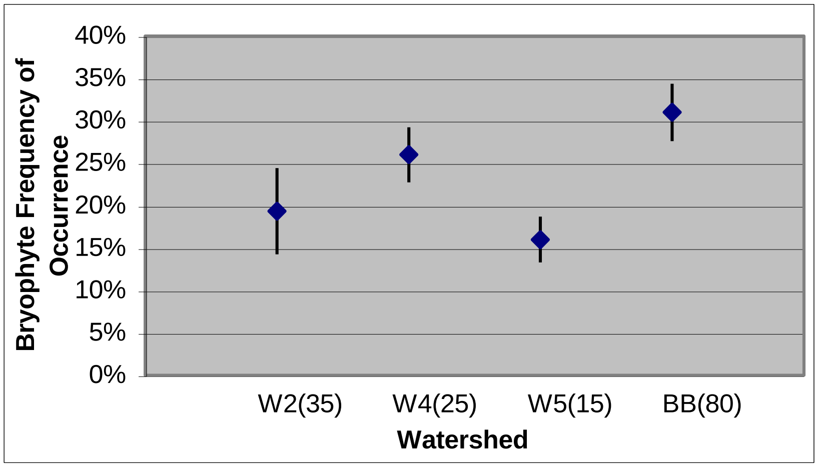

FIGURE 2. Total Bryophyte Abundance in Each Stream

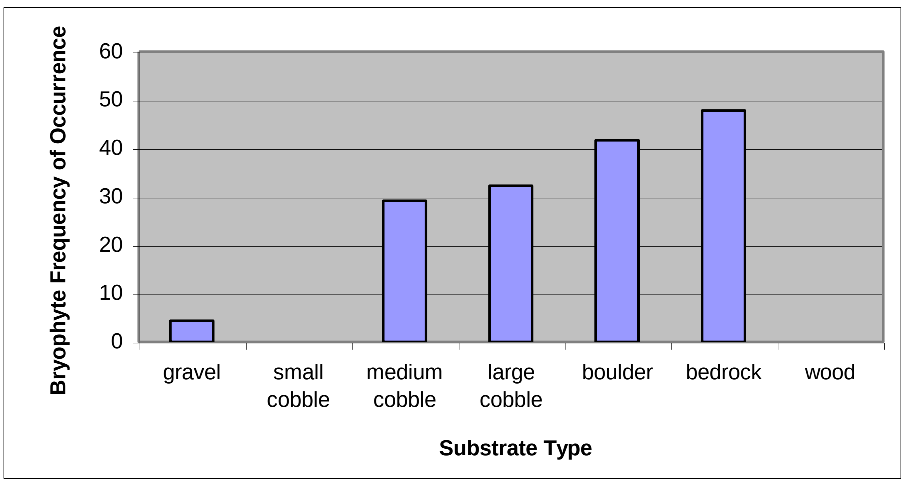

FIGURE 3. Watershed 4: Bryophyte Frequency of Occurrence vs. Substrate Type

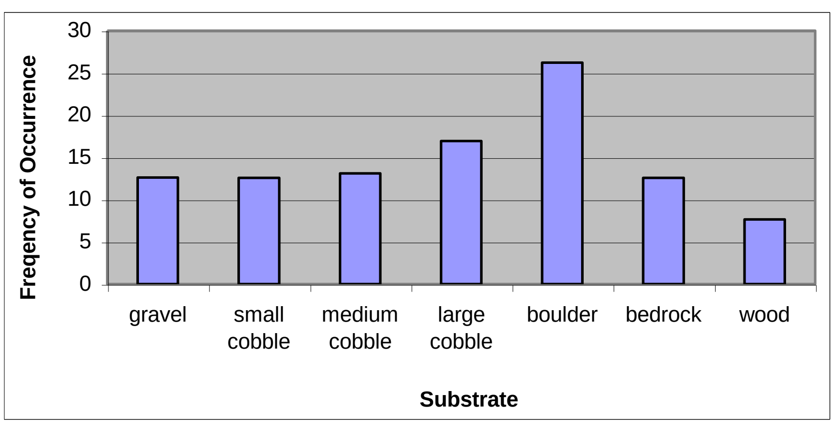

FIGURE 4. Watershed 4: Frequency of Substrate Occurrence

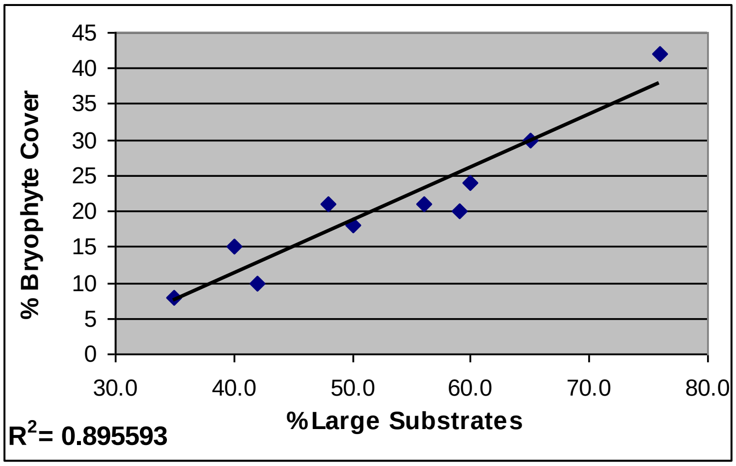

FIGURE 5. Percentage of Bryophytes Inhabiting Large Substrates vs. Frequency of Occurrence of Large Substrate (Cobble, Boulder, and Bedrock)

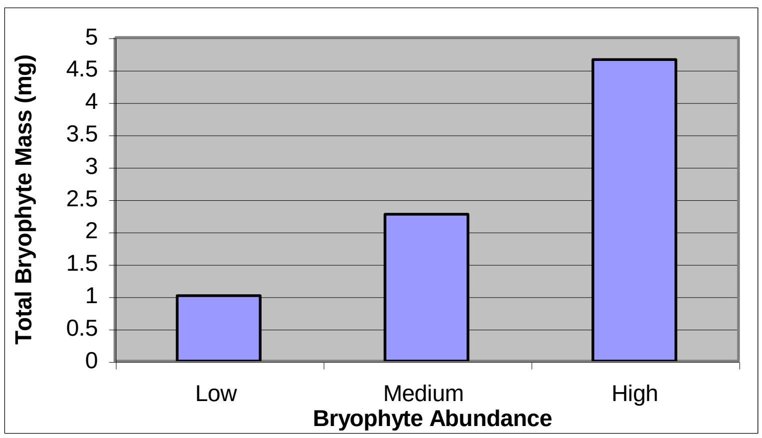

FIGURE 6. Release 4: Ash-free Dry Mass of Bryophytes

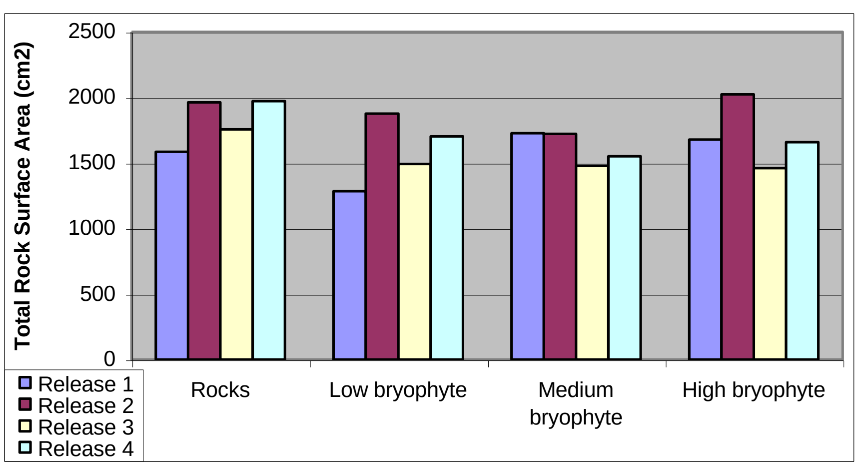

FIGURE 7. All Nitrate Releases: Rock Measurements

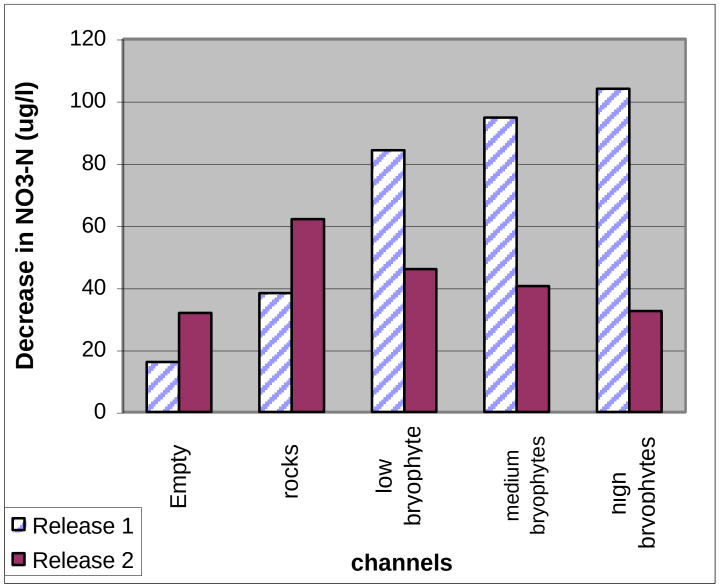

FIGURE 8. Low Discharge Release 1 and 2: Decrease in Nitrate Concentrations by Different Abundances of Bryophytes (initial concentration=118.5 ug/l)

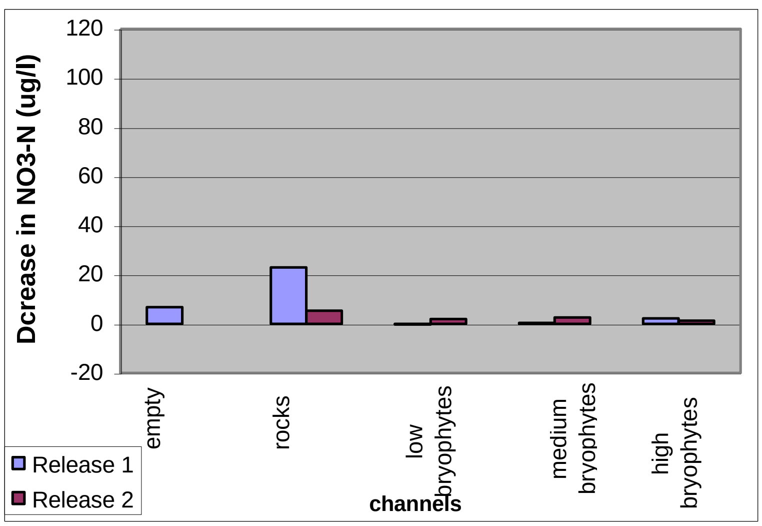

FIGURE 9. High Discharge Release 1 and 2: Decrease in Nitrate Concentrations by Different Abundances of Bryophytes (initial concentration=121.6 ug/l)

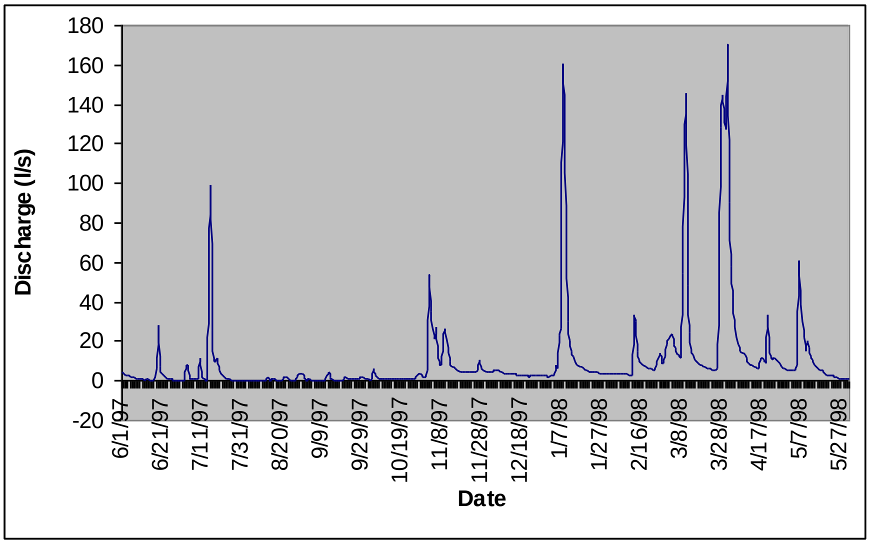

FIGURE 10. Watershed 3 Discharge Data

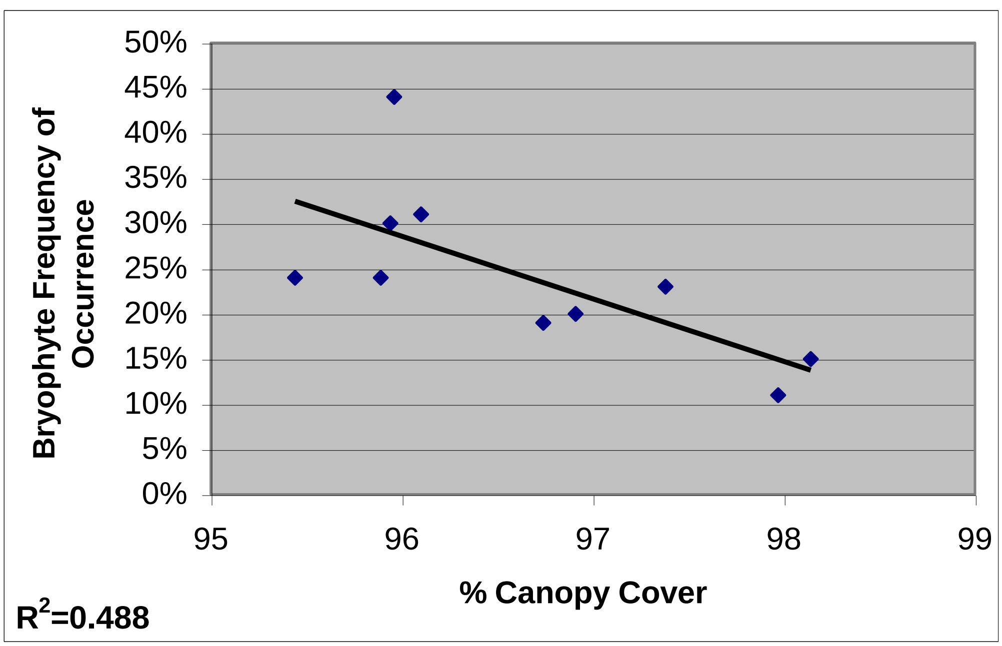

FIGURE 11. Canopy Cover vs. Bryophyte Frequency of Occurrence