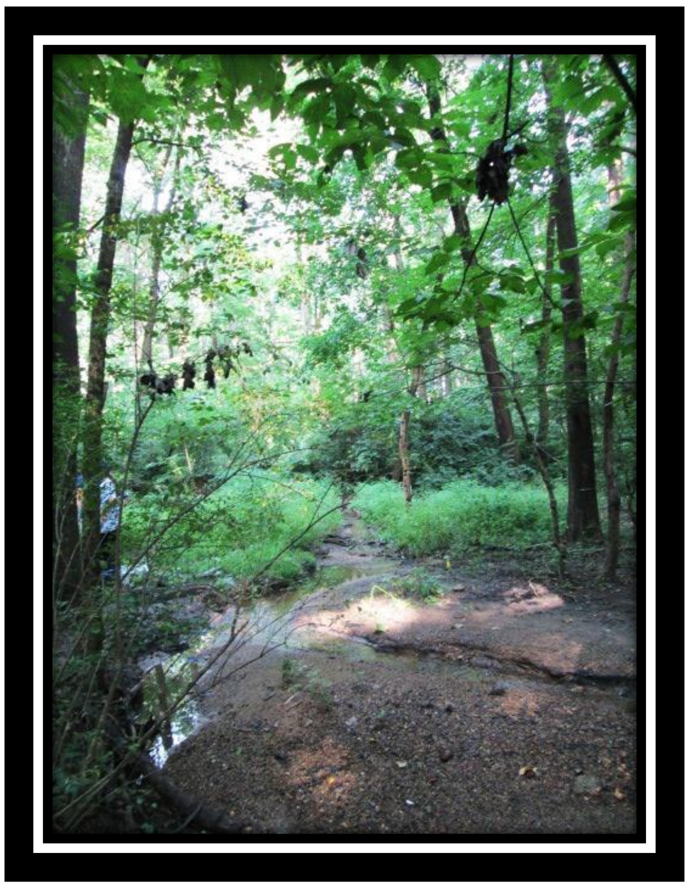
The Effect of White Grass and Orange Jewelweed on Nitrate Levels and Bacteria Populations in the Soil



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Abstract

A disruption in the nitrogen cycle in ESSRE Site 4 was discovered during a general biota survey in 2012. Bacteria levels were high, as expected, but nitrate levels were still very low in comparison to other sites. The survey also revealed two species that together composed more than 70% of the plant life in Site 4: white grass and orange jewelweed. Grasses and annuals consume large quantities of nitrogen, so we hypothesized that these two plants were absorbing disproportionate amounts of nitrate. We located seven plots with varying amounts of grass and nitrogen, and tested soil core samples from each of the sites once a day for three days for bacteria density and nitrate. We found that the soil from sites with both a greater density of white grass and jewelweed contained less nitrate, but that white grass was slightly more strongly correlated with low nitrate. We also discovered that jewelweed density was associated with higher bacteria counts, while invasive white grass did not help attract more bacteria. This was concerning because white grass' presence has been growing over the past five years in ESSRE Site 4, and it appears that white grass has been absorbing disproportionate quantities of nitrate while failing to foster the bacteria that fixes it from the air. Further research on the nitrogen cycle and white grass is strongly recommended.

Introduction

Within the nitrogen cycle, many organisms are involved in converting nitrogen gas into a form that can be utilized by plants. Some groups of bacteria in the soil, often located within the roots of plants, take the nitrogen from the air and convert it into the organic form ammonia (NH_3). This form is later released in its inorganic form ammonium (NH_4^+), which plants can use to make nucleic acids and proteins. Other groups of bacteria then transform any excess released ammonium into nitrite ($N0_2^-$), which a third group of bacteria convert into the other form of nitrogen plants can utilize, nitrate (NO_3^-) (Ophardt, C. 2003; Brencas-Moreno, 2009; Carey, Cirelli, Laria & McCormack, 2012).

During the ESSRE biota survey (ESSRE, 2012) we found that the soil in ESSRE Site 4 (ESSRE, 2012) contained the densest population of heterotrophic bacteria (633,000/cc of soil) in comparison to other sites in the ESSRE program (with p-values ranging from 0.0515 to 0.1027). This confirmed prior research completed at ESSRE Site 4, which traditionally has tested for high levels of both bacteria and nitrogen (Rogers, Torres, Campochiaro & Sick, 2002; Adoga, Dinger, Searles & Thomas, 2011; ESSRE, 2012). However, the anticipated levels of nitrogen (ESSRE, 2012; Rogers et al., 2002; Adoga et al., 2011) did not appear in this year's survey and in fact ESSRE Site 4 contained the least amount of nitrate (6.96 ppm) compared to each of the other ESSRE sites tested this year (with p-values ranging from 0.005 to 0.04)

Yet the 2012 biota survey of ESSRE Site 4 also revealed that the site was 43% composed of white grass (*leersia virginica*) and 35% composed of orange jewelweed (*impatiens capensis*), both annual plants known to consume large amounts of nitrogen (Voight, Fermanian & Wehner, 1998; Carl, Reeder, Whiting & Wilson, 2011). These overwhelmingly dominant species in ESSRE Site 4 were not found in any other ESSRE sites, and indeed the population of invasive white grass has grown steadily from nonexistence in 2007 to its current level. Jewelweed populations have been strong every year, except for a brief respite in 2005 and 2006 (ESSRE, 2002-2012). While all plants require nitrogen, some types of plants need more than others. Members of the grass family are well documented to require large amounts of nitrate to reach peak health. Nitrogen, a key macronutrient, makes up three to six percent of a grass's mineral composition, proving that "it is present in larger percentages than other chemicals" (Voigt, Fermanian & Wehner, 1998). In addition to grasses, many other annual plants such as the jewelweed found in ESSRE Site 4 are also among those that require large amounts of nitrogen to survive, which is why nitrogen fertilizers are often used to supplement natural levels on grass lawns and in gardens (Carl, Reeder, Whiting & Wilson, 2011).

Therefore, when looking at the apparent anomaly in the nitrogen cycle discussed earlier, we realized that a possible cause for this anomaly could be the dominant kind of plant life found in ESSRE Site 4. We hypothesized that, despite the statistically higher density of bacteria combined with the unexpectedly statistically lower amounts of nitrate found in the soil this year in ESSRE Site 4, the source of the lack of nitrogen was due to the large population of white grass and orange jewelweed located there.

Methods

Seven plots 40 cm in diameter were chosen in ESSRE Site 4 (N 39.35733; W 076.63840) located three meters from the streambed. One plot with no plants south of the stream served as the negative control. From that point, five more plots containing a mixture of white grass and jewelweed were chosen with centers one meter apart on a line parallel to the streambed. One plot containing neither white grass nor orange jewelweed was also chosen north of the streambed as a positive control.

For each plot, the number of orange jewelweed and white grass specimens were counted and recorded. Each day between 9:00-11:00 am for three consecutive days one 2.5 cm wide by 15 cm deep cylindrical sample of soil was taken at the same time from each of the seven plots for a total of 21 samples. A different random location within each plot was chosen for sampling each day.

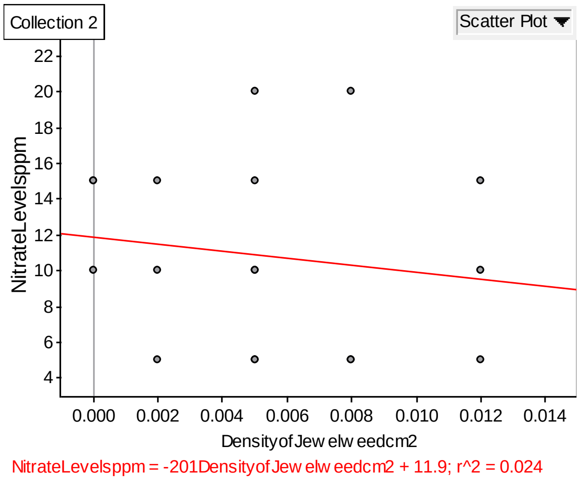
The samples were extracted and tested for nitrate (ppm) using the LaMotte™ Model STH-14 (code 5010-01) Combination Soil Test Kit. Simultaneously, the same samples were also serial diluted with sterile water to 10^{-3}. 100 µl of all four dilutions for each sample were then plated on their own individual 3M Petrifilm™ Aerobic Count Plates. The plates were left to grow for three days and then counted on the lowest dilution containing at least five bacteria colonies to calculate the number of bacteria per cc of soil. Nitrate data and bacteria data were examined to determine if there were any correlations between the density of orange jewelweed and white grass and the amounts of bacteria and nitrogen.

Results

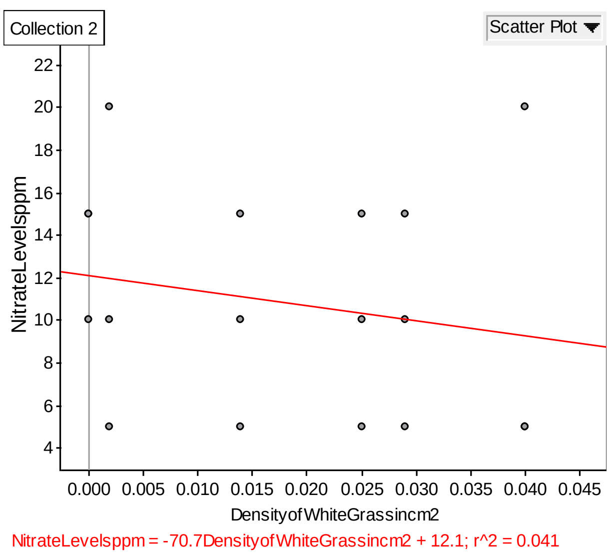
As shown in Graph 6, bacteria levels fluctuated but remained ultimately relatively consistent, in accordance with our hypothesis. However, Graph 3 shows that an increase in jewelweed density led to an average increase in soil bacteria, with an r^2 value of 0.046. Interestingly, white grass did not affect the bacterial levels so, as depicted in Graph 4, no matter the density, there was about the same quantity of bacteria, averaging around 1,500,000.

In Graph 5, the positive correlation between greater soil bacteria quantities and more nitrogen is clear, with an r^2 value of 0.13. In accordance with our hypothesis, both jewelweed (Graph 1) and white grass (Graph 2) are negatively correlated with the amount of nitrogen. The white grass' effect is stronger, however, as the data shows, an r^2 value of 0.041 as compared to 0.024

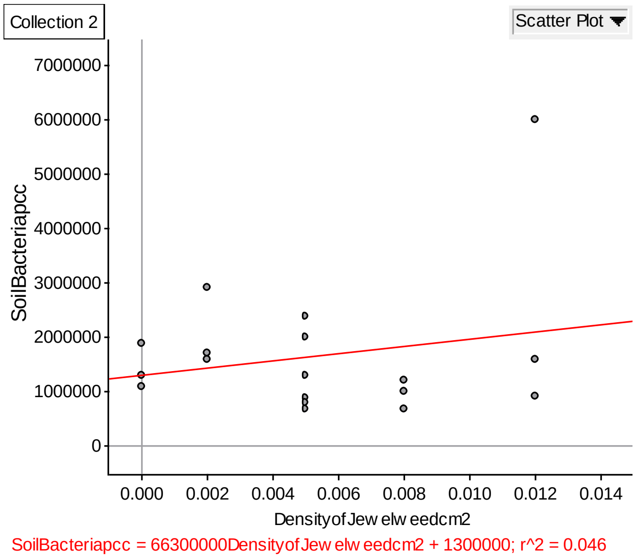
Graph 1. Density of Jewelweed vs. Nitrate Levels



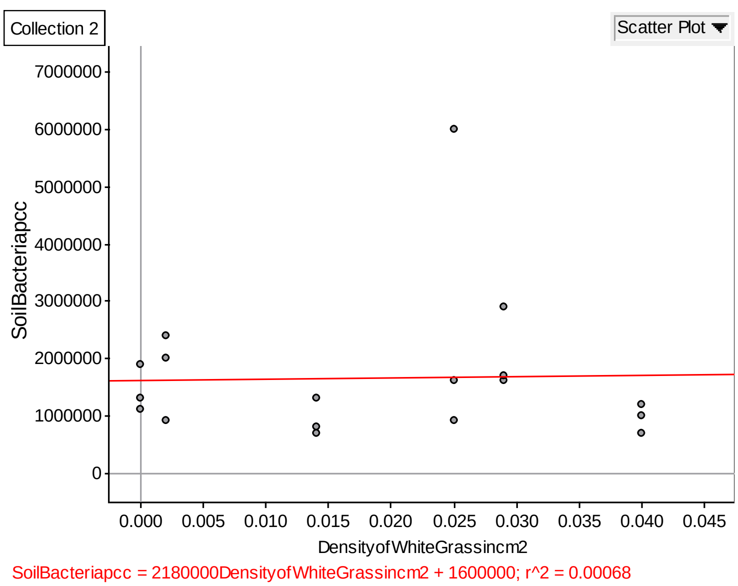
Graph 2. Density of White Grass vs. Nitrate Levels



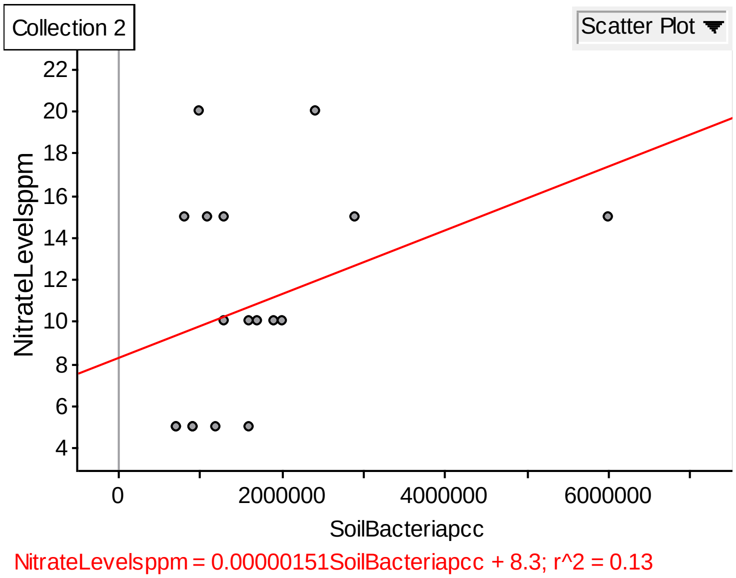
Graph 3. Density of Jewelweed vs. Soil Bacteria



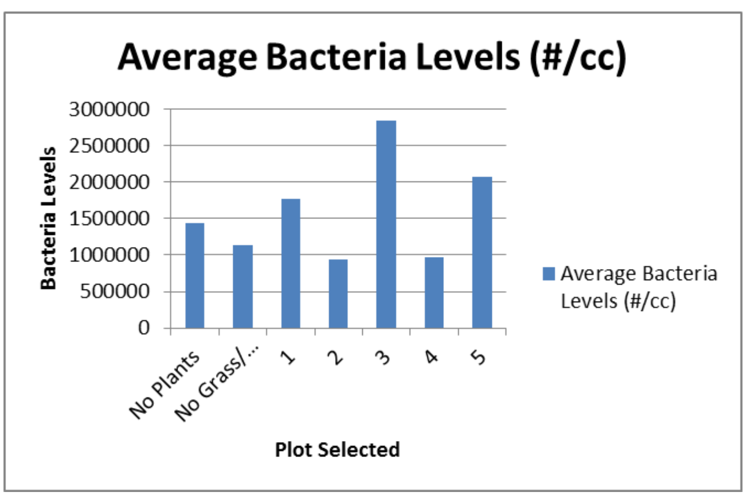
Graph 4. Density of White Grass vs. Soil Bacteria



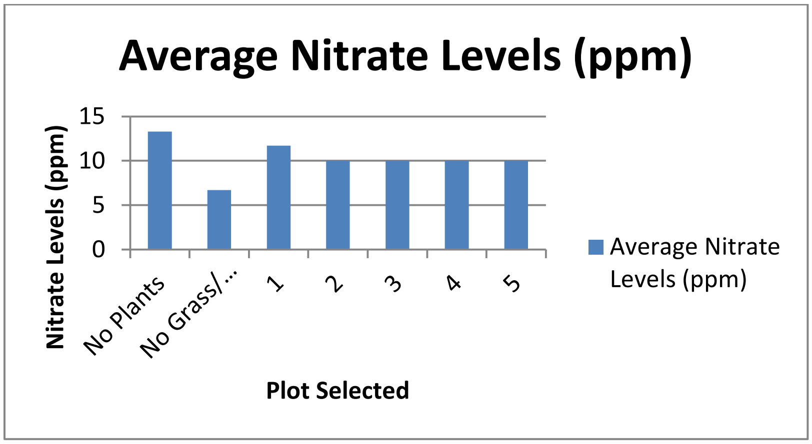
Graph 5: Soil Bacteria vs. Nitrate Levels



Graph 6. Average Bacteria Levels



Graph 7. Nitrate Levels



Discussion

Before performing this experiment, this year's survey data (ESSRE, 2012) led us to wonder whether the nitrogen cycle itself in ESSRE Site 4 was disrupted. We believed that, due to very small amounts of nitrate (6.96 ppm) in spite of a dense bacteria population (633,000/cc of soil), the relationship between the two might have been upset. However, as Graph 5 clearly shows the expected, healthy relationship between bacteria density and nitrate levels (Rogers et al., 2002; Adoga et al., 2011; Ophardt, C. 2003; Brencas-Moreno, 2009; Carey et al., 2012). Hence, it is clear that the source of the original anomaly was not due to an overall disruption in the nitrogen cycle.

Therefore, as we hypothesized, the source of the low nitrate levels was due to the presence of the plant life there. As shown in Graph 1 and Graph 2, both jewelweed and white grass are consuming great quantities of nitrogen. Hence, while the relationship between bacteria and nitrate (i.e. denser populations of bacteria yield higher levels of nitrate) is still intact, the presence of jewelweed and white grass in ESSRE Site 4 appears to be causing the low levels of nitrate in the soil there by absorbing it for their own biological purposes.

While yet it might seem that both white grass and jewelweed are having a negative impact on the environment by reducing the amount of nitrate in the soil, the data clearly shows that the jewelweed are providing a favored habitat for bacteria. Graph 3 shows that, as the number of jewelweed increases, the density of bacteria populations increases as well, and since jewelweed is a native plant in the area, it would make sense that the bacteria are well adapted to their root systems. In a symbiotic relationship, the jewelweed's roots provide habitat for bacteria, and the bacteria in turn produce nitrogen, which jewelweed consumes in large quantities. Therefore, despite causing decreased nitrate levels, the jewelweed appears to have a positive impact on the environment by maintaining a give-and-take relationship with the soil bacteria surrounding its roots.

In contrast, the white grass appears to be a threat within its ecosystem by “stealing” nitrate from the native jewelweed. Graph 4 shows no positive correlation between white grass density on bacteria levels at all (r^2= 0.00068). White grass is clearly not beneficial to bacteria in this region and this is why it is especially concerning that white grass seems to be consuming nitrate at a greater rate than the jewelweed, as shown by a higher correlation between the density of white grass and the amount of nitrate in the soil shown in Graph 2 (r^2 =0.041). Therefore, white grass appears to be consuming that which it is not helping to produce..

Further research is recommended to confirm this correlation; however, these preliminary findings suggest that white grass, as opposed to jewelweed, is negatively impacting the ecosystem, and there is a longitudinal data from previous ESSRE biota surveys (ESSRE, 2012) to support this claim.

White grass is an invasive species whose presence has grown from nonexistence in ESSRE Site 4 since 2006 (ESSRE, 2002-2012). Jewelweed reached levels of almost 80% dominance in the first few years of the ESSRE program, but populations have been declining in favor of white grass in recent years (ESSRE, 2002-2012). This invasion may not only explain recent nitrate deficiencies, but also why plant populations have been so consistently unstable since ESSRE’s inception.

Further research on the impact of white grass is strongly recommended for the health of the ecosystem at ESSRE Site 4. In future studies, it may be beneficial to investigate more aspects of the nitrogen cycle, such as protozoa, ammonia and nitrite to discover whether white grass has a negative impact on those as well. Continuing research on whether white grass does, in fact, have no impact on bacterial levels is also recommended. One deficiency of this research is that plots with both jewelweed and white grass were investigated. As white grass continues to overtake the ecosystem, it should be easier in future studies to isolate areas of just white grass for study.

Conclusion

Overall, white grass and jewelweed both were found to have an impact on the nitrogen cycle. Our original hypothesis was proven correct. However, the impact of orange jewelweed is positive, while white grass’ impact is negative. White grass is an invasive species that does not contribute to the health of bacteria and nitrogen in the ecosystem, and further researchers should monitor the condition of the nitrogen cycle in future years.

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