The Effect of Water on Bacteria and Protozoa Densities, and The Nitrogen levels in the Soil

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Abstract

Nitrogen levels depend on the density of Bacteria and Protozoa while water positively affects the relationship between Bacteria and Protozoa. This relationship in turn affects the balance between Nitrate, Nitrite, and Ammonia. The levels of Nitrate, Nitrite, and Ammonia and the Bacteria and Protozoa densities were inconsistent In the Annual 2011 Biota Survey (E.S.S.R.E., 2011). Based on this, we hypothesized that the Bacteria's and Protozoa's unpredictable behavior was due to the lack of water in site 4. We started by measuring 3 plots that had 5 samples 2.5 cm wide and 15 cm deep were taken from each plot; one from each plot corner and one in the middle of the plot. Chemical tests for Nitrite, Nitrate and Ammonia were conducted on all soil samples using the LaMotte Model STH-14 soil test kit. Serial dilutions were performed using sterile water to 10^{-4} on all soil samples. Then we repeated the process after adding various amounts of water to each plot. Water is an important contributor to maintaining both of these relationships, thus we expected that as the amount of water increased, the relationship would go back to their natural states.

Introduction

Nitrogen is essential to the growth and development of plants and other organisms in the environment (Argonne National Laboratory, 2005). However this colorless, odorless gas cannot be used by plants or any other complex life in its neutral form (Killpack and Buchholz, 1993). Specialized soil bacteria add H_20 to the nitrogen, converting it into ammonia (NH_3) in a process called nitrogen fixation. Other soil bacteria then convert the ammonia into nitrites ($N0_2$) which nitrites are then transformed into nitrates ($N0_3$) in a process called Nitrification. The plants soak up the nitrate into their roots to create amino acids and nucleic acids, the vital building blocks of all life and these chemicals then move through the food chain where they are recycled through decomposition and the process repeats itself (Brewer, Kelley-Brown, Moats, and Wiltgen, n.d). Hence the nitrogen cycle is critical to the well-being of plants and soil.

The flow of the nitrogen cycle is greatly influenced by bacteria. Since bacteria play such a major role in the nitrogen cycle, the productiveness of bacteria influence the various levels of nitrite, nitrate and ammonia. The efficacy of bacteria significantly depends on the behavior of protozoa. Protozoa are the natural predators of bacteria, and so have an inverse relationship (meaning that as protozoa numbers increase the amount of bacteria decreases) (Valentino and Ingham, 2011) Furthermore the amount of water in the soil affects the relationship between protozoa and bacteria (Ingham, 2011). Since more water in the soil increases protozoa mobility, the protozoa can hunt the bacteria much faster. As the protozoa consume the bacteria they also force them to reproduce much faster (Ingham, 2011). Water therefore significantly changes the levels of bacteria and protozoa which in turn influence the amount of fixed nitrogen available to all organisms in the ecosystem.

In the past several years, Maryland has undergone a serious drought (USGS, 2011), and consequently, the soil has gone through serious changes due to the lack of water. In a recent biota survey (E.S.S.R.E 2011), unexpected levels of nitrite, nitrate and ammonia and unusual correlations between the density of bacteria and protozoa in the soil were observed. Since water affects bacteria activity, which in turn affects the nitrogen cycle, we hypothesized that increases in water in the soil would return bacteria and protozoa densities to their normal inverse relationship, and return normal chemical activity to the soil.

Methods

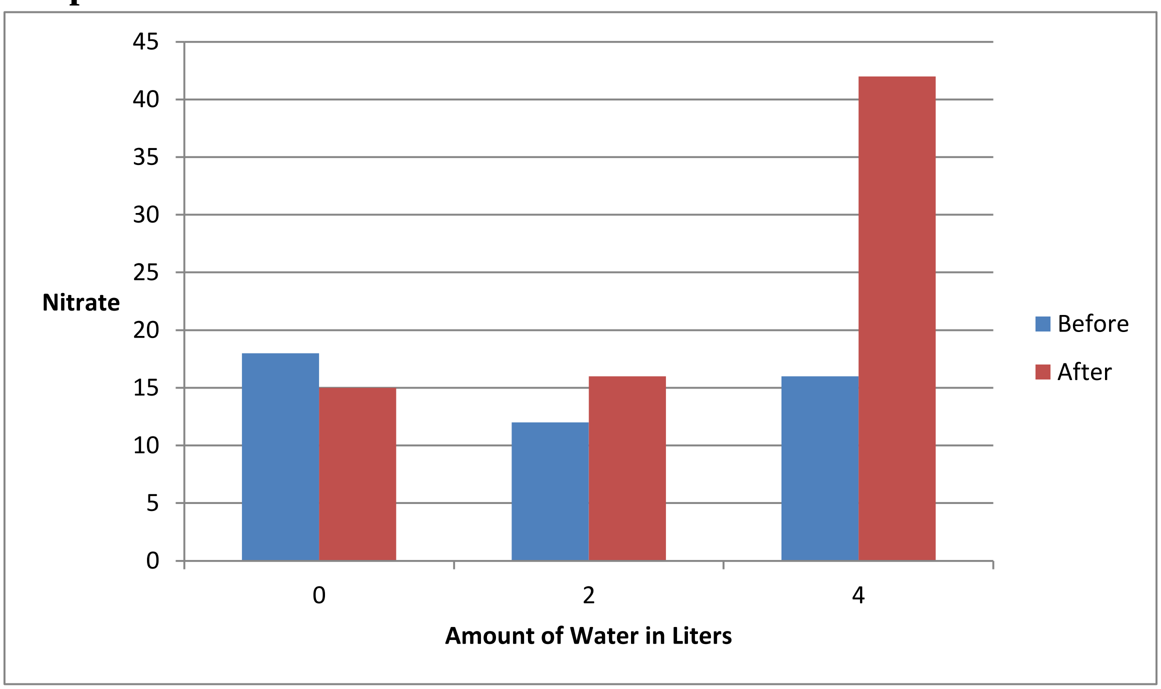
In the quadrant 3 of E.S.S.R.E Site 4 (N 39° 21.458, W 76° 38.331), 3 adjacent ½ m by ½ m plots were delineated. Flags were placed at the corners of each of the plots and labeled with their corresponding experimental variable (Plot 1, Plot 2, and Plot 3 respectively). All ground cover was cleared and 5 samples 2.5 cm wide and 15 cm deep were taken from each plot; one from each plot corner and one in the middle of the plot. Chemical tests for Nitrite, Nitrate and

Ammonia were conducted on all soil samples using the LaMotte Model STH-14 soil test kit. Serial dilutions were performed using sterile water to 10^{-4} on all soil samples. The 100 µl of 10^{-1} through 10^{-4} dilutions for each sample were plated on 3M™ Petrifilm™ Aerobic Count Plates and were allowed to grow for 3 days before counting. A modified Foissner-Uhlig extraction process (Brockmeyer, 2008) was performed on all samples to determine protozoa density.

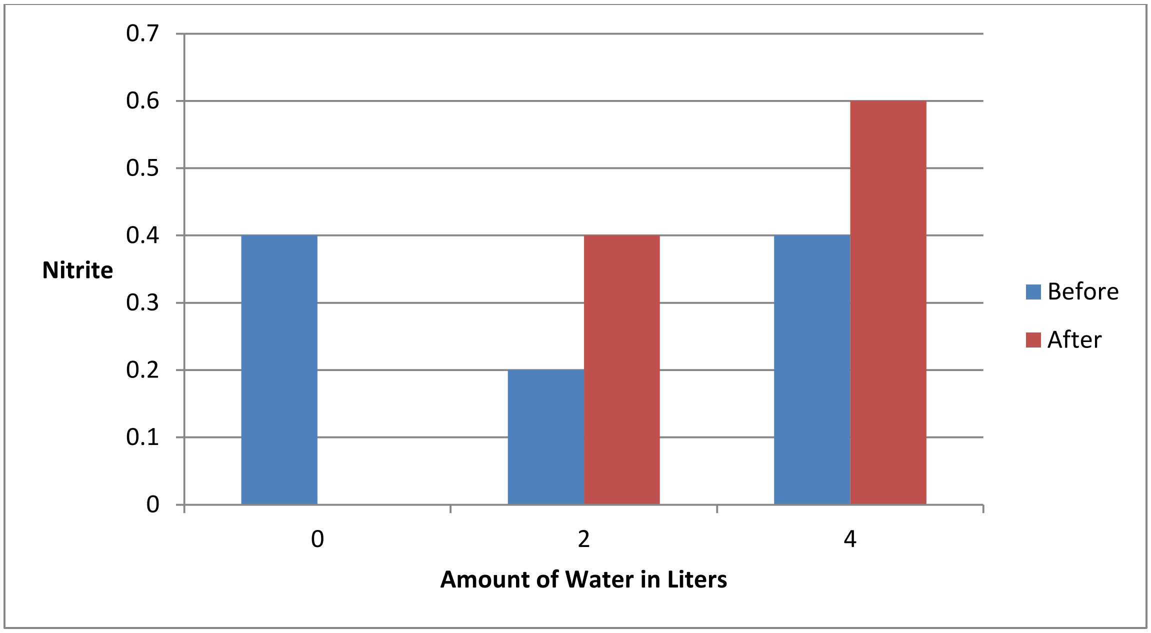
Following the extraction of the positive control samples, Plot 1 received no additional water. Plot 2 received 2 liters of water. Plot 3 received 4 liters of water. The plots were allowed to soak with their respective water treatments for 24 hours. The next day, 5 additional soil extractions 2.5 cm wide and 15 cm deep were taken from each plot adjacent to the holes left from the previous extractions. The same tests for nitrite, nitrate, ammonia, bacteria and protozoa were then performed on the second set of samples that were performed on the first set.

Results

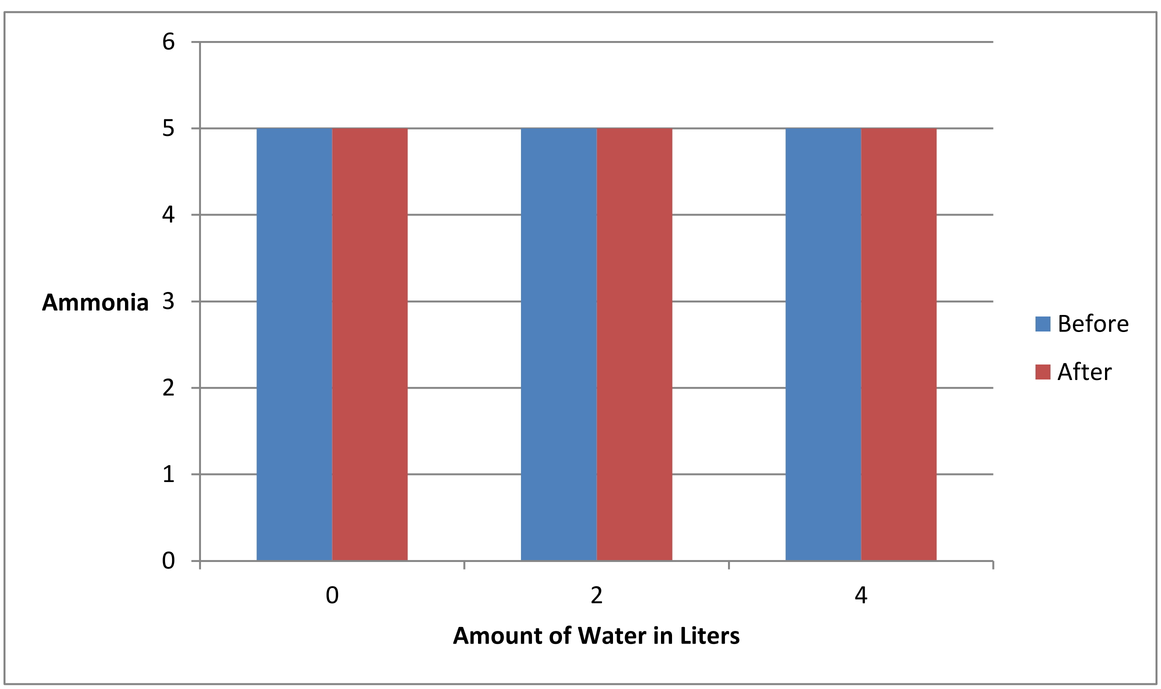
Graph 1: Nitrate Before and After



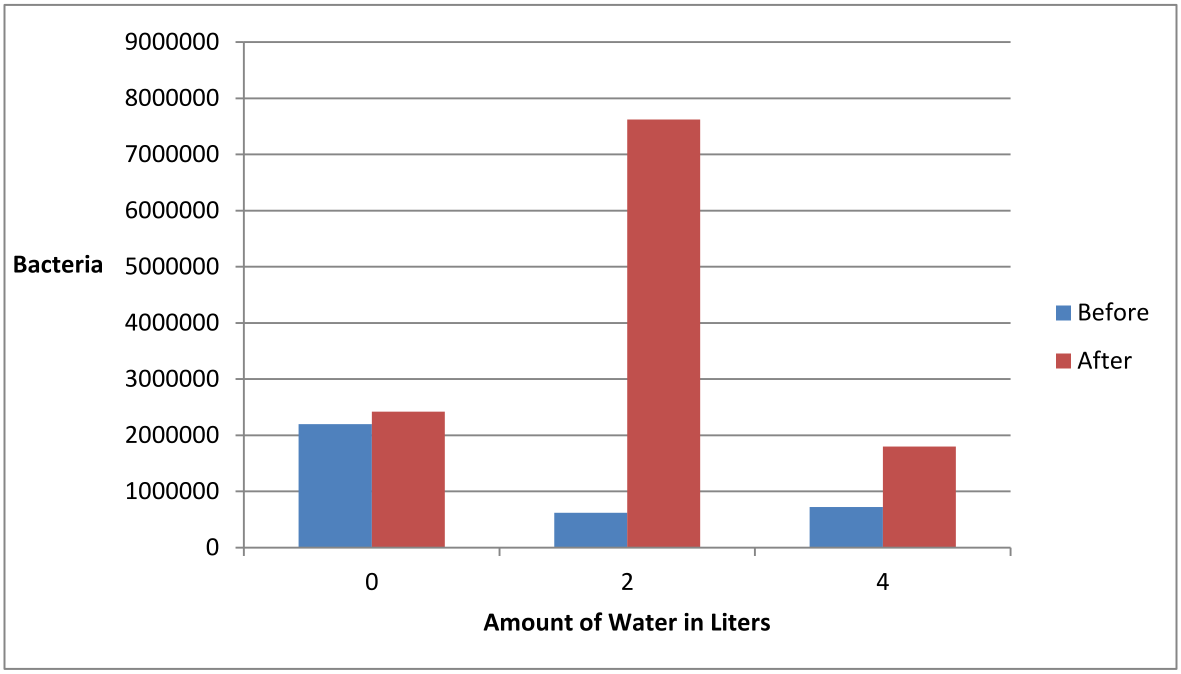
Graph 2: Nitrite Before and After



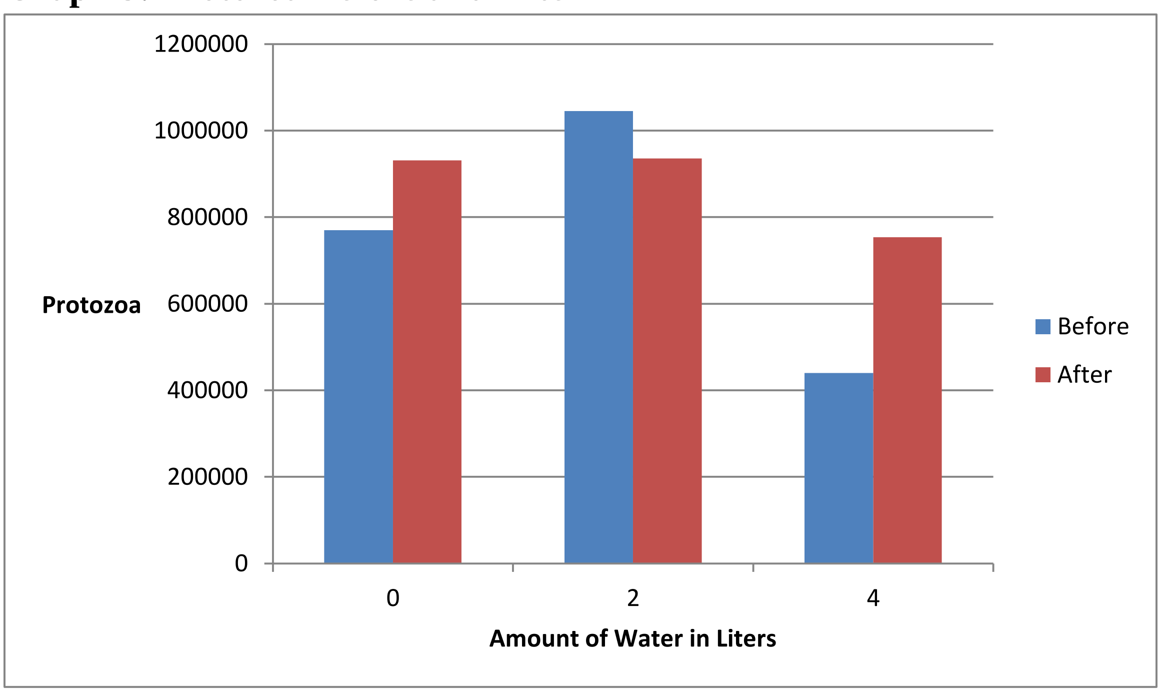
Graph 3: Ammonia Before and After



Graph 4: Bacteria Before and After



Graph 5: Protozoa Before and After



Discussion

In this experiment we hypothesized that adding water to our plots would help stabilize the nitrogen cycle which the 2011 E.S.S.R.E. biota survey had shown was being disrupted (E.S.S.R.E. 2011). Our hypothesis proved to be correct. As shown in graphs 1-2, the increased availability of water in plots 2 and 3 caused the levels of both nitrate and nitrite to increase, while increasing the rate at which ammonia was being converted in these plots (as indicated by the stable low levels of ammonia present in these plots) (see graph 3). The return of this normal relationship to the soil in plots 2 and 3 was further confirmed by the densities of bacteria and protozoa found after the water was added. As graphs 4 and 5 illustrate, after the water was added, the levels of both bacteria and protozoa dramatically increased.

Therefore the addition of water to the experimental plots helped to return the relationship between soil bacteria and protozoa back to its normal, inverse one, with healthy levels of bacteria and protozoa cycling around 500,000-1,000,000 bacteria per cm^3 and protozoa per grams of soil. Since the nitrate levels were considerably higher and the ammonia levels were lower (consistently 5 ppm across all samples), the data shows that the conversion of the ammonia into nitrate by bacteria had returned to its normal activity levels after the water was added. Based on our research findings, the next logical step we would take for further research would be to go out and take samples from many different areas. In particular because our plots were in close proximity to a stream, we would repeat our experiment in areas away from the stream where the ground was drier.

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