The Relationship Between Mold and Sulfate Levels in Soil

Payton Bennett, Abby Miller, Emilia Pierce, Sophia Peltzer

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Abstract

Fungi and sulfate are both very important in soil ecosystems. Fungi play a significant role in the process of decomposition, during which they convert organic matter into forms such as nitrate and sulfate that are accessible for other organisms to use for their own metabolic needs. One such nutrient released, sulfate, is vital for plants and animals because it is needed to manufacture enzymes and other proteins as well as some vitamins. The 2017 E.S.S.R.E. Biota Survey revealed statistically significant differences in sulfate levels (ppm) and fungi densities (#/cc) between the soils of Microclimate 1 and Microclimate 2. It was hypothesized that where there were increased fungi densities (#/cc), a greater rate of decay was taking place that would explain this discrepancy. Therefore, 24 samples of soil 15.5 cm deep with a diameter of 2 cm were collected from the 2 different microclimates and were tested for fungi density (#/cc), sulfate (ppm), and humus (ordinal scale) as a test indicator for decomposition rates. It was found that although the rate of decomposition was significantly higher in Microclimate 2, fungi density (#/cc) was statistically the same throughout both microclimates. Additionally, although it was expected that Microclimate 2 would have higher levels of sulfate (ppm), the data showed that the levels were actually higher in Microclimate 1. Therefore, our hypothesis was incorrect. Further research revealed that mold takes sulfate from the soil and converts it to certain amino acids which act as nutrients for plants, and Microclimate 2 data fit this expected pattern but Microclimate 1 data. We believe this could be due to difference in moisture levels between the two sites impacting the other major decomposer, bacteria. For further research, bacteria and its relationship to levels of moisture in the soil could be tested.

Introduction

One of the most important roles that fungi play in any ecosystem is the process of decomposition, which is especially true in soils. Through this process, fungi convert organic matter that is trapped in unusable forms, such as dead leaves, into forms that are easier for organisms to use, such as nitrate or sulfur (Ingham, 2107). Fungi are able to do this by releasing digestive enzymes that turn different organic compounds into simple carbohydrates such as glucose, xylose, sucrose, and fructose (which the fungi can absorb and metabolize for their own needs) along with other simple organic molecules and inorganic salts. Some fungi can also use proteins as their source of carbon and nitrogen (Moore et al., 2017). Through fungi and the process of decomposition, nutrients containing the biological elements are metabolized and replenished into the soil where plants are able to access them through their roots and use these nutrients to manufacture their own organic material (Campbell, 2017). Thus, plants rely on decomposers to provide nutrients crucial to them as well as the rest of ecosystem.

Sulfur is one of these necessary nutrients for plants and animals. Sulfur is needed to form plant enzymes and other as well as some vitamins. Hence, it is essential to the production of chlorophyll and for the process of photosynthesis (Tucker 1999). Soil temperature and moisture are big factors that determine how much sulfate is available to plants, and cold weather as well as water level extremes (both high and low) reduce sulfate's mobility and therefore its availability. In most soils, Sulfate is relatively mobile, however it is easily leached out from sandy soils (Camberato et al., 2017).

While analyzing the 2017 E.S.S.R.E. Biota Survey (E.S.S.R.E., 2017), we observed in E.S.S.R.E. Microclimate 1 (N 39.37789, W 076.6409) high amounts of both sulfate (687.5 ppm) and mold (37,383/cm^3), and in Microclimate 2 (N 39.21485, W 076.38389) we observed statistically significantly lower amounts of both sulfate (1.25 ppm) and mold (10,154/cm^3). Given that both these microclimates share common temperature and moisture levels, we were puzzled by this anomaly. However, there is significantly more dead plant material in Microclimate 1; therefore we hypothesised that there may be more decomposition taking place in Microclimate 1, correlating with the increase in amounts of sulfur found in the soil there.

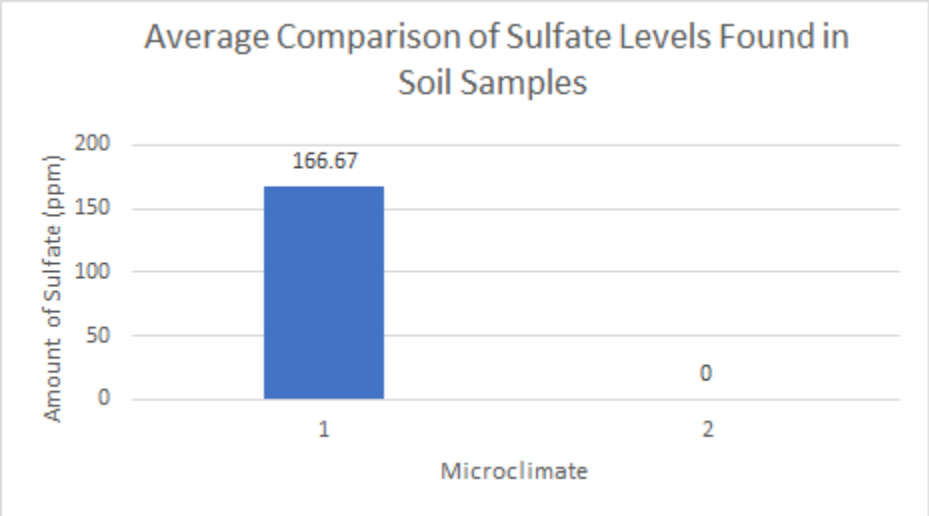
Methods

In E.S.S.R.E. Microclimates 1 (N 39.37789, W 076.6409) and 2 (N 39.21485, W 076.38389), a total of 6 soil core samples 15.5 cm deep with a diameter of 2 cm were extracted daily for 4 days for a total of 24 samples. Each day, 3 of these samples were collected from one of the corners of Quadrant 1 in Microclimate 1 and 3 from Quadrant 4 in Microclimate 2. Sampling was eventually done over the course of the 4 days in order to sample one corner per day. Samples were collected in areas with little to no herbaceous or woody plant life, other than ground ivy to control for the impact of plant life. Samples in each corner were taken approximately 30 cm apart from one another.

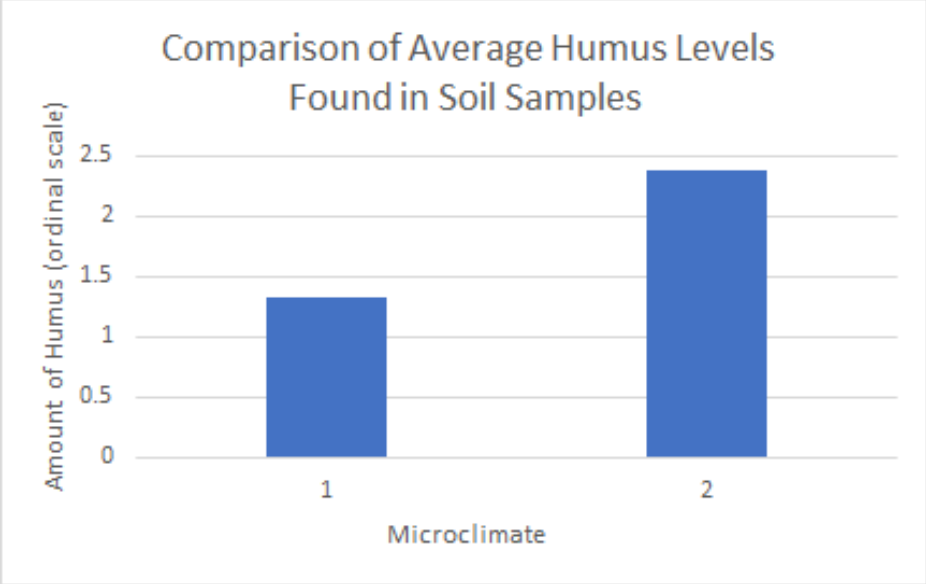
Following collection each day, all 6 samples were individually serially diluted using sterile water to the 10^{-2} degree with 100 µl of each dilution plated onto its own individual 3M Petrifilm™ Yeast and Mold Count plate and grown for 72 hours. Simultaneous with the serial dilutions, all 6 samples were tested for sulfate (ppm) and humus (ordinal scale) using the LaMotte model STH-14 test kit. 72 hours following plating, the number of mold and yeast per cm^3 of soil was determined from each plated sample. All data was collected over the course of the following dates: July 19th, 20th, 21st, and 24th of 2017.

Results

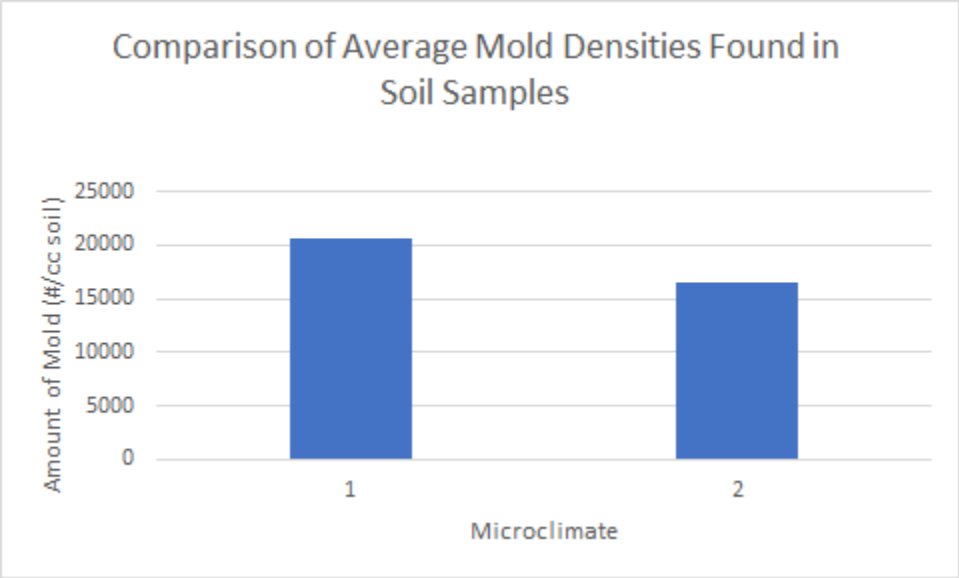
Graph 1:



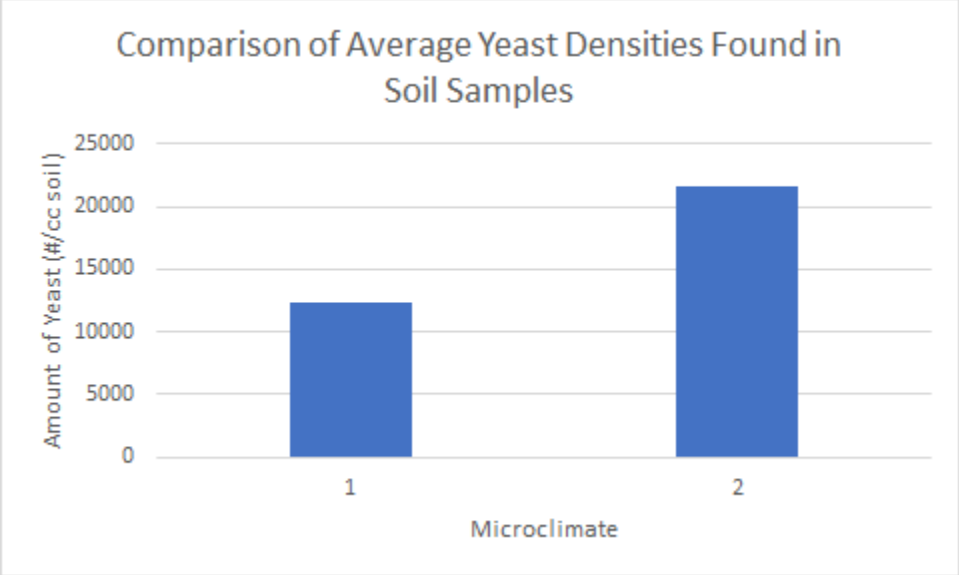
Graph 2:



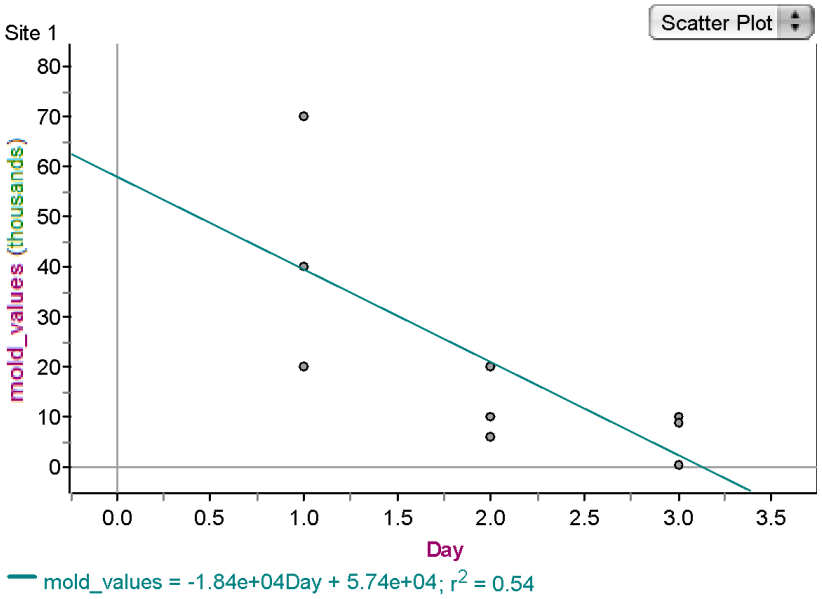
Graph 3:



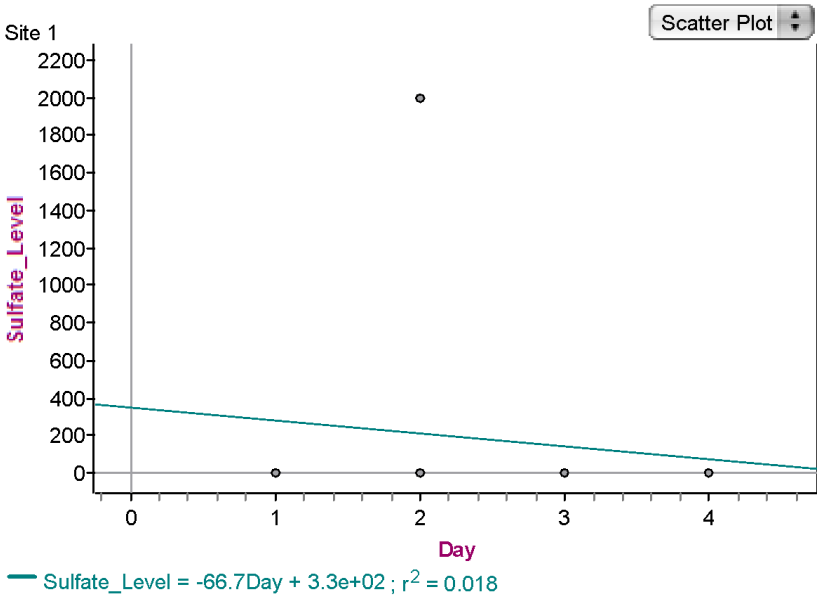
Graph 4:



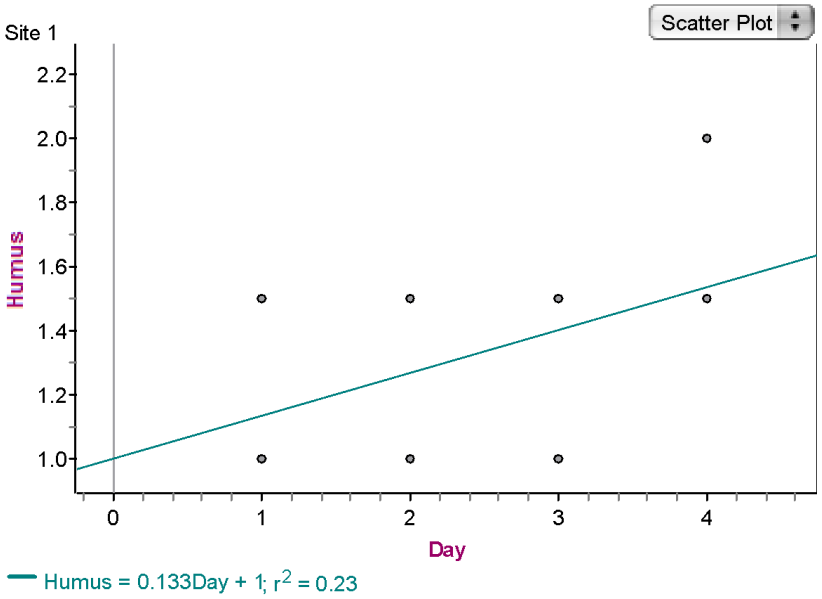
Graph 5: Shows the correlation between time and the densities of mold (# / cc) in the soil found in Microclimate 1 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.



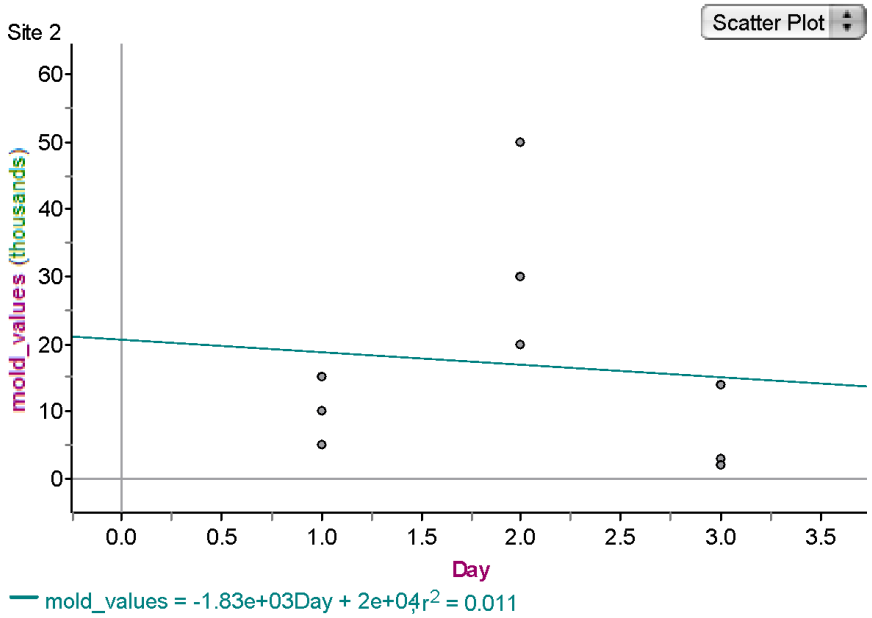
Graph 6: Shows the correlation between time and the level of sulfate (ppm) in the soil found in Microclimate 1 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.



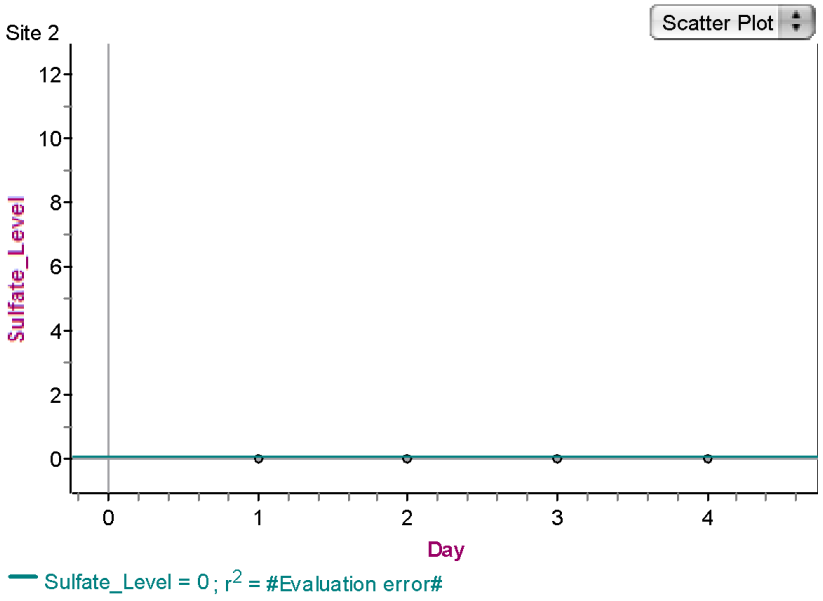
Graph 7: Shows the correlation between time and the level of humus (ordinal scale) in the soil found in Microclimate 1 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.



Graph 8: Shows the correlation between time and the densities of mold (# / cc) in the soil found in Microclimate 2 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.



Graph 9: Shows the correlation between time and the level of sulfate (ppm) in the soil found in Microclimate 2 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.



Graph 10: Shows the correlation between time and the level of humus based on the Ordinal Scale in the soil found in Microclimate 2 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.

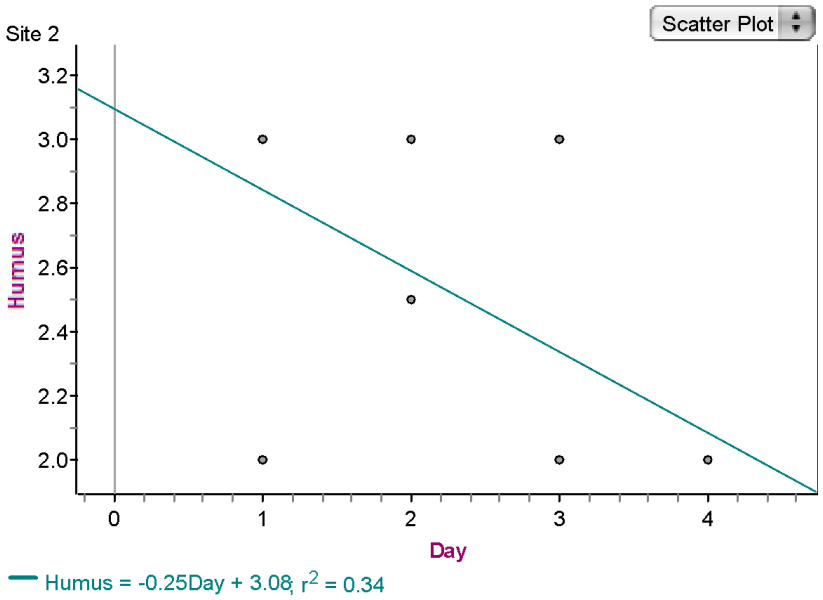


Table 1: Shows the results of t-tests between all data collected in Microclimate 1 and Microclimate 2 over the course of 4 days, July 19th, 20th, 21st, and 24th of 2017.
P-Values

Site 1 vs. Site 2	p-Value
Humus	6.1974×10^{-6}
Sulfate	.347
Mold	0.655
Yeast	.284

Discussion

As Graph 1 shows, there was a significant difference in the decay rates between Microclimate 1 and Microclimate 2 ($p= 6.1974x10^{-6}$), indicating that there was a higher rate of decay in Microclimate 2 during the course of our experiment. However, as Graph 3 shows, the densities of the mold in both microclimates were effectively the same (p=0.655). Furthermore, because the rate of decay was significantly greater in Microclimate 2 than in Microclimate 1, we would have anticipated the sulfate levels in Microclimate 2 to be higher than in Microclimate 1, given that most sulfur in the soil is stored in organic matter, which is the result of decomposition. However, as Graph 2 clearly indicates, this was not the case. Instead, the sulfate level went from a value of 1.25 ppm in Microclimate 2 found in the original Biota survey to 0 ppm while the sulfate levels in Microclimate 1 fell from an original value of 687.5 ppm to 166.67 ppm (p=0.347). Hence, we can conclude that our original hypothesis was incorrect.

Further research, though, revealed that mold in the soil is responsible for taking the sulfate there and transforming it into the amino acids methionine and cysteine. Therefore, where there is more mold in the soil there should actually be less sulfate (Fitzgerald et al., 1988). As a consequence of this finding, we realized that the data from Microclimate 2 fits this expected pattern. As Graphs 8 and 9 show, the change in mold values over the course of the experiment was minimal ($r^2 = 0.011$), as was the corresponding

change in the sulfate levels (r^2=0). But most significantly, as Graph 10 shows, the *change* in the rate of decay over time dropped significantly in Microclimate 2, which would be expected given the results seen in Graph 8 as well as the significantly greater density of yeast found in Microclimate 2 (see Graph 4; p=.284). Since Fungi in their yeast morphology do not decompose very well (Brock, 2006), the decreasing change in rate of decomposition in Microclimate 2 is most probably due to the fact that more fungi have transitioned from their mold morphology to their yeast morphology in that location, slowing the rate of decay over the course of our experiment.

Although the results for Microclimate 2 fit the expected pattern, the results for Microclimate 1 did not. As shown in Graph 5, there was a statistically significant decrease in mold over time (r^2=.54) and therefore, one would expect to see a significant increase in the sulfate levels. However, as Graph 6 clearly shows, there was instead a significant decrease in the sulfate levels over time. Since the mold levels decreased over time, we would have expected the humus levels to decrease because the change in the rate of decomposition taking place should have been less. Instead Graph 7 shows that the rate of production of humus actually increased over time (r^2=0.34). Therefore, the change in rate of the production of sulfate levels should have increased because where there is more organic matter we would expect more sulfate. But, as graph 6 shows us, the rate of change in production barely altered (r^2=0.018).

In conclusion, there must be other factors than what we hypothesized that are affecting the decomposition and the relationship between mold density and humus and sulfate levels in Microclimate 1. One possible factor could be the amount of moisture in the soil. Microclimate 1 has a large amount of sand (63.3% according to the E.S.S.R.E. 2017 Biota survey) and therefore there may not be adequate water for the soil bacteria living there (which also contribute significantly to the decomposition process) to flourish. Indeed, low levels of bacteria were found in this year's survey (114,000 #/cc) which might account for the disparities seen in the

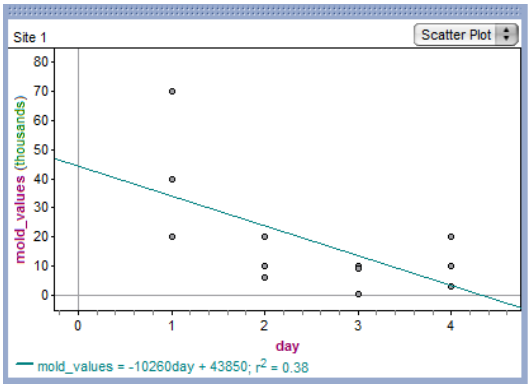


Figure 1

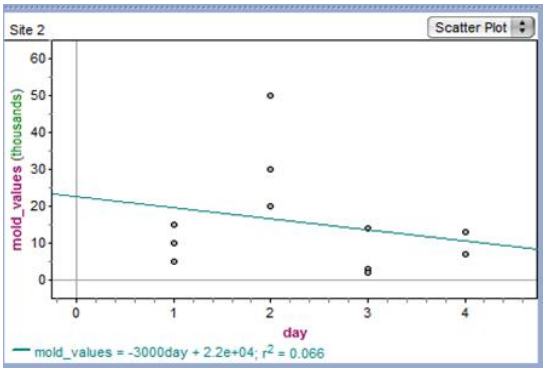


Figure 2

Microclimate 1 data. In addition, when our day 4 data was added to our preliminary results, the notion that the observed rates in decay may be due to moisture is supported. Between Day 3 and Day 4 and before Day 4 data was collected, there was significant rainfall in both Microclimates 1 and 2 (weather.gov, 2017), when the Day 4 data was added into the statistical analysis, the r^2 value of the mold scatterplot for Microclimate 1 decreased from 0.54 to .38 and the r^2 value for Microclimate 2 went up from a value of 0.011 to a value of 0.066. Because this is the expected change in rates of decay if the conditions for decomposition had improved, we are all the more confident that water may be the key to the disparities seen in the Microclimate 1 data. In the future, we would conduct an experiment in which we manipulate Microclimate 1 by adding different amounts of water to the soil and testing for the change in the density of the soil bacteria located there.

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