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Comparing The Microbial Diversity of Campus Cultivated Flowers Vs EEC Wildflowers At Longwood University

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Background Information

- Environmental microbiomes are microbial communities found in an ecosystem. (Ushio, M., et al. 2015)
- Flowers are directly linked reproductive output (Aleklett K., et al. 2014)
- Bacteria dominate the environment around us (Pace, N. R. et al. 1997)
- The microbiomes collected came from the surface of flowers at Longwood University.
- One high human interaction area, the Rotunda flower bed
- One less human Interaction area behind the Environmental Education Center(EEC)

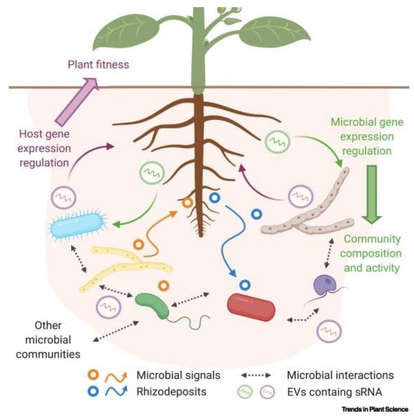
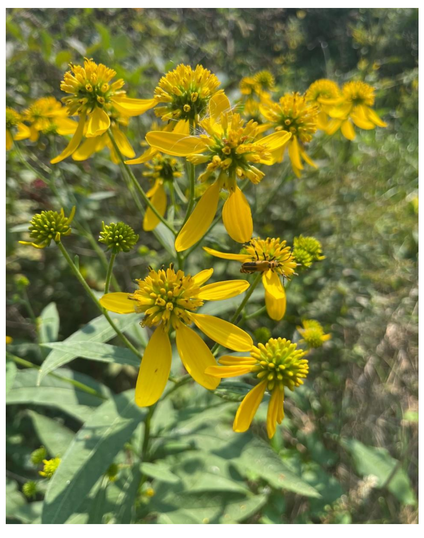


Figure 1. Microbiome types. This photo shows an example of different types of microbial communities found in plants

Specific Aim

Question: What is the microbial diversity among flowers in an area of heavy human interaction as opposed to a flower in an area of lower human interaction on the Longwood University campus?

Hypothesis: The flower in a heavy human interacted area would have more diverse microbiomes and the flower in a lower human interacted area would have less diverse microbiomes.



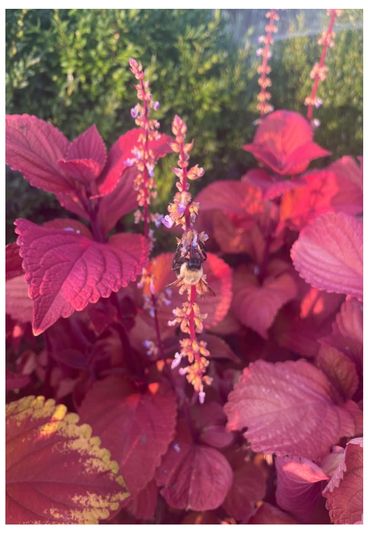


Figure 2. Flowers samples were taken from. . They were found behind the EEC. The red flowers are from Rotunda flower bed. The yellow flowers also known as Brassica Rapa

Methods

- **Collection**
  - Collection of samples from behind EEC and Rotunda Fountain
- **Growth of Bacterial Colonies**
  - Bacteria was incubated for 48hrs at room temperture.
  - Growth was observed over these hours.
  - Analyze and Figure out specific bacterial colonies
- **DNA Amplification**
  - 16s rRNA sequences were amplified using PCR
  - 16srRNA were cleaned and sent for DNA sequencing.
- **Identifcation of colonies**
  - The DNA sequence was analyzed and searched using the NCBI BLAST database.

Results

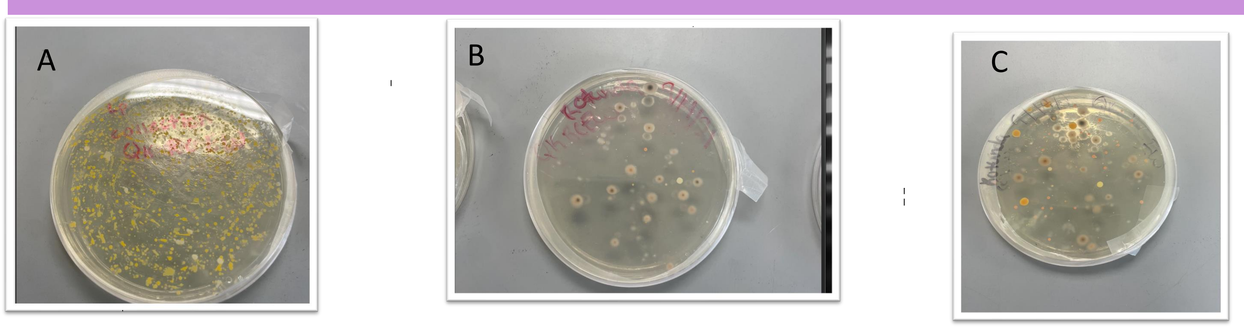


Figure 3. Agar Plates After 48 hour incubation.
A.EEC Bacterial Diversity. There was more bacterial abundance on flower swabbed behind the EEC.
B. Rotunda Fountain Flower, test 2. We have some fungal colonies here.
C. Rotunda Fountain Flower, test 1. We have more bacterial diversity as seen by different bacterial colony colors.

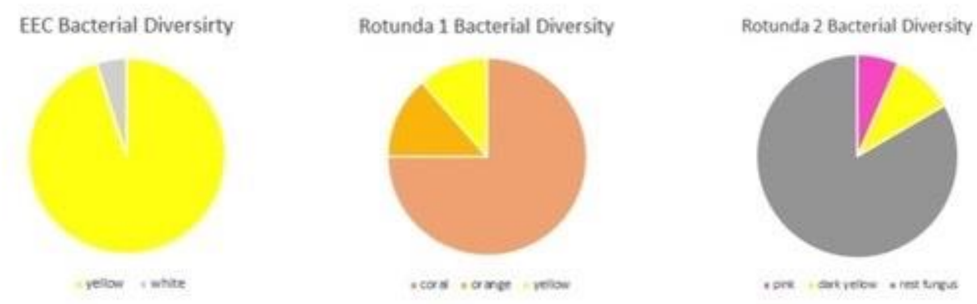


Figure 4. Bacterial Diversity Chart. These charts represent the proportions of colors found at each sampling site. (A left, B middle, C, right)

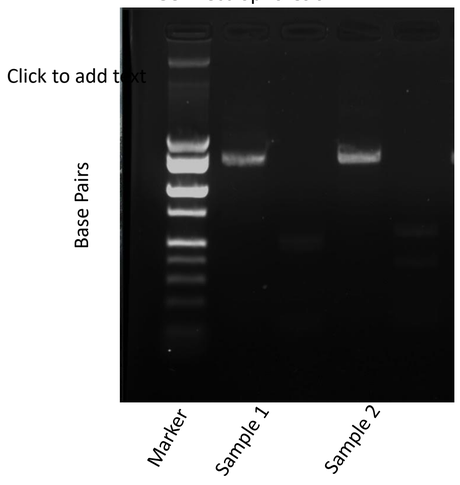


Figure 5. Gel Electrophoresis. This photo shows our product with and without MSP to see if there was any kind of electric charge.

Name of Bacteria	Percent Match	Nucleotide Gap Percentage
Pseudomonas trivialis	98.49%	4/955 0%
Pseudomonas poa	98.49%	4/955 0%
Pseudomonas tonstantinii	98.29%	4/955 0%
Pseudomonas marginalis	98.29%	4/955 0%

Figure 6. Percent Identity/Gap Percent.
This graph shows which bacterial group is more closely related to our test. The higher the percent identity, the closer it is a match, same goes for the gaps.

Conclusions

- The hypothesis was supported, there was more diversity in the highly human interacted regions of Longwood University.
- P*seudomonas marganalis* was found to be the closest match for both RNA sequences inserted in the BLAST data base.
- The highly human interaction region of the Longwood University campus had more bacterial diversity than the lower human interacted regions of Longwood University

Future Directions

- Collect more of an abundance of samples from more sampling sites
- Identify the bacterial DNA from the Rotunda samples to see if a match appears

References

Ushio, M., Yamasaki, E., Takasu, H., Nagano, A. J., Fujinaga, S., Honjo, M. N., Ikemoto, M., Sakai, S., & Kudoh, H. (2015, March 3). *Microbial communities on flower surfaces act as signatures of pollinator visitation*. Nature News. Retrieved September 13, 2021, from
Pace, N. R. & Norman R. PaceThe author is in the Departments of Plant and Microbial Biology and Molecular and Cell Biology, U. of C. A molecular view of microbial diversity and the biosphere. *Science* (1997).
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