Plastic-Decomposing Bacteria

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Program / Major: Running Start

Abstract

As plastic production is increasing, so is plastic pollution. The purpose of this experiment is to determine if there are any plastic-decomposing bacteria to help with reversing plastic pollution in bodies of water. Plastic samples from Lake Union were collected and swabbed bacteria samples were transferred to Luria broth and water media. Test tubes are prepared to observe different conditions of bacteria growth in LB and water media. In each of the media, algaecide, PET-1 and biodegradable plastic were introduced, as well as a control sample is prepared with only media and no algaecide. All conditions were incubated at room temperature. Overgrowth of bacteria was observed in LB media, while minor to no growth was observed in water media. LB samples were switched to a new tube with a no-carbon salt media to promote growth of bacteria that could break down hydrocarbon from the plastics. At this time, the plastic in the tubes were weighed every two weeks to monitor the change in weight. After two months, bacteria from each tube were plated on agar plates to isolate colonies. PCR was conducted to identify the potential bacteria that possess plastic decomposing qualities. These results will give us insight into types of bacteria that may breakdown various types of plastics by determining the mass change and detection of possible metabolites, such as enzymes.

Objectives

The increase of plastic production and consumption results in an increase of plastic pollution. When plastic waste enter ocean, they create a waste of 5.25 trillion pieces and add up to 269,000 tons of waste.
They cause a threat for marine animals as they mistakenly consume plastic as food.
They cause a threat for oceans as they physically degrade into smaller pieces of microplastics, making it harder to get out of the ecosystem.
They cause chemical pollution as they release harmful chemicals into soil, water and finally through larger ecosystems.
The objective of this experiment is to find if there is a natural way to combat the plastic pollution through bacterial decomposition in nature.

References

Wang, Xin, et al. “Growth Strategy of Microbes on Mixed Carbon Sources.” *Nature News*, 20 Mar. 2019, www.nature.com/articles/s41467-019-09261-3.
Yoshida, Shosuke. “A Bacterium That Degrades and Assimilates Poly(Ethylene ... - Science.” *Science*, 11 Mar. 2016, www.science.org/doi/10.1126/science.aad6359.

Collaborated with Puget Soundkeeper (plastic waste collection) and SoundBio Lab (use of their makerspace)

Methods

- Various plastic samples were collected from Lake Union and swabbed using a 1x1 cm grid.
- Swabs were introduced to Luria broth and water. Each of these media received PET-1 and biodegradable plastic with algaecide. There were two control tubes: one with media and another with media and algaecide.
- After incubation in LB, bacteria were transferred to a no-carbon salt media.
- The plastic samples in the no-carbon salt media were weighed every two weeks and the media was replenished.
- Plastic samples were washed with DI water, and ethanol; obtained dry weight, washed in ethanol and reintroduced to their tubes.
- After two months, T-streaks were done on LB Agar to isolate bacteria. From each sample, one isolated bacteria sample was selected for PCR.
- After gel electrophoresis, the samples were purified and sent to Eurofin for sequencing.
- Sequence results were analyzed using BLAST analysis with 16srRNA database.
- The weighing experiments continue every two weeks.

Plastic collected with bacteria samples

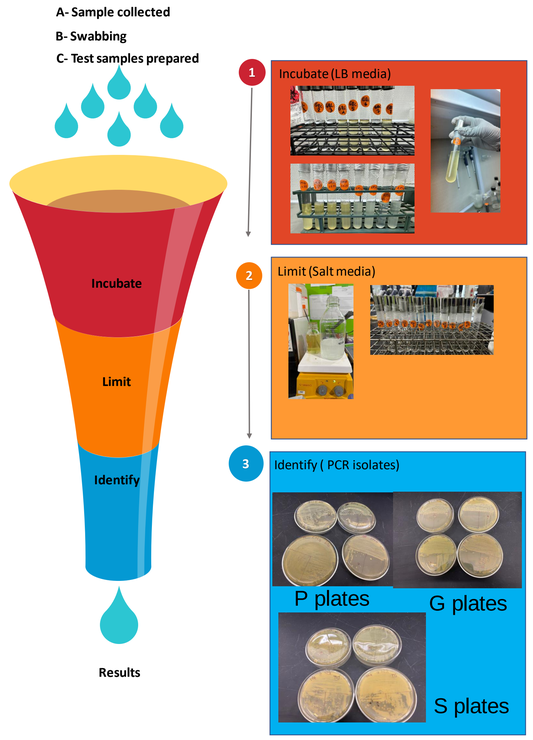
Origin of S, P, and G plates bacteria

Plastic samples Decomposition

PET-1

Biodegradable Plastic

Methods

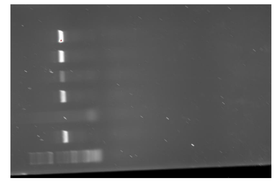


Discussion

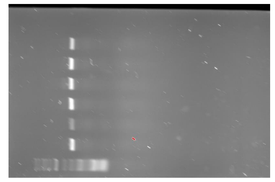
To determine bacteria with plastic decomposing qualities, the bacteria from the plastic waste had to be selected. First, the bacteria swabbed from the plastic samples were incubated in nutrient media, resulting in overgrowth of bacteria. To limit bacteria strains, the media is changed to a salt media which allows the carbon from the plastic to be the only nutrient for bacteria. This change in media led to a growth of selected bacteria which are potential plastic decomposers. To determine the extent of decomposition, the weight of the plastic samples (PET-1 and Biodegradable plastic) were tracked over a two-month period (the changes in weight overall weight measurements are presented in the graphs). Overall, biodegradable plastic showed an overall net decrease in weight, displaying a stronger potential of decomposition. PET plastic, however, showed a net increase in weight. This increase is expected to be caused by the creation of a layer of biofilm which may have colonized next to the plastic but has not been able to decompose it. So far, biodegradable plastic is expected to be decomposable by bacteria in nature (specific bacteria listed in results section). It is also worth noting that a new scale was used as of April 22nd which was calibrated so it is not expected to have any impacts on measurements.

Results

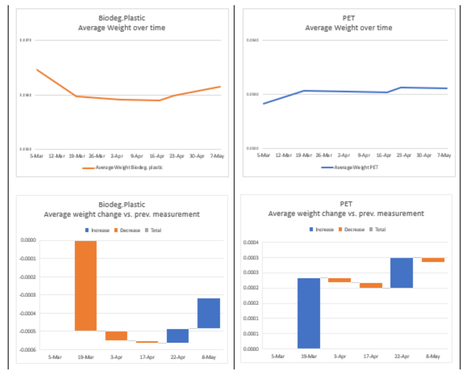
These are the bacteria identified from various tubes:
G4- *Janthinobacterium rivuli strain FR68W*
G5- *Pseudomonas urethralis strain BML-PP042*
G6- *Pseudomonas allii strain MAFF 301514*
G8- *Pseudomonas allii strain MAFF 301514*
P5- *Pseudomonas brenneri strain CFML 97-391*
P6- *Pseudomonas gessardii strain CIP 105469*
P8- *Pseudomonas palmensis strain BBB001*
S4- *Pseudomonas brenneri strain CFML 97-391*
S5- *Pseudomonas brenneri strain CFML 97-391*
S6A- *Pseudomonas brenneri strain CFML 97-391*
S6B- *Pseudomonas oryzihabitans strain L-1*
S8- *Pseudomonas palmensis strain BBB001*



G4 – P6



P8-S8



Conclusion

Biodegradable plastic showed an overall net decrease in weight by -0.9%, displaying a stronger potential of decomposition by bacteria. PET-1 plastic showed a net increase in weight by 0.5% expected to be caused by the creation of a layer of biofilm which may have colonized next to the plastic but has not been able to decompose it. Biodegradable plastic is expected to be decomposable by bacteria in nature (specific bacteria listed in results section).

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