Microbiomes Around us: Exploring Bacterial Biodiversity of Oysters at polluted and conserved regions on the East Coast

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Abstract

It is crucial to understand the effect different environmental conditions within an urban area have on ecosystems. One way to manage this relationship is to compare the amount of biodiversity of any given organism within these regions. In this project, we decided to explore the effect on bacterial biodiversity of oysters within different regions of the East Coast, where urbanization varies based on the waters where the organisms were supplied. Since oysters are considered filter feeders and purchasing commercial oysters is frequent by consumers, we can analyze the bacterial population within the oysters located at Blue Point, Long Island, and Plymouth Rock Massachusetts. The urban location of Blue Point causes the water quality to be increasingly contaminated, whereas the waters at Plymouth Rock generally tended to be cleaned by the company that would provide the oysters. In order to analyze the bacterial biodiversity within both regions, we will first gather the oysters and plate the bacteria by swabbing their gills carefully. Once the bacteria has developed and grown some colonies, we then extracted the DNA using the silica method. And after, we performed gel electrophoresis on this sample in order to discern if they worked. We then sent off the samples for sequencing. Finally, we then input the sequences into DNA Subway to find any possible identifications of the bacterial colonies.

Research Aim

Analyzing bacterial biodiversity within commercial oysters at Blue Point, NY and Wellfleet, Massachusetts.

Hypothesis

The bacteria within the microbiomes of Blue Point oysters would have higher biodiversity due to higher pollution, than the biodiversity of Wellfleet oysters.

Introduction

Biodiversity within urban areas plays a crucial role for both the people and the ecosystems that reside within them. Compared to suburban and rural locations, urban areas tend to have higher rates of disturbances due to increasing environmental factors (Bierwagen, 2006). This continuous declination of biodiversity creates a surge to discover new sustainable ways to allow ecosystems to thrive (Martínez-Ramos et al., 2016). There have been many attempts and studies that focus on ways to preserve and expand biodiversity within a wide range of urban locations (Blicharska et al., 2011). For example, in New York City, multiple green spaces have been created in order to maintain the growing biodiversity within the city, carrying multiple functions (García Sánchez et al., 2018). Other forms of monitoring biodiversity within urban areas tend to target organisms within polluted areas of water (Parmar et al., 2016). Similar to larger organisms, bacteria can also be an indicator when analyzing the effects of urbanization within ecosystems (Oljira et al., 2018). There can be a biodiversity of bacterial colonies that are created due to urbanization that can gradually affect other organisms within the urban ecosystem (Krishna et al., 2009). Filter feeders can be used for this exact purpose since they tend to capture bacteria from surrounding waters within their glands. Thus, these organisms would allow the collection of these bacteria in order to analyze their biodiversity, depending on the location (Ostroumov).

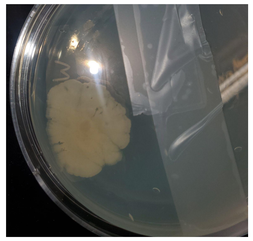
In this project, we will be working with oysters. Oysters play a crucial role within the ecosystem since they clean out surrounding water and provide food and shelter for certain organisms. They are considered filter feeders since they tend to filter out any bacteria that reside within the algae or phytoplankton that they consume (Jeamsripong et al., 2018). This filtering method would lead to different forms of bacteria accumulating within these regions of the oysters. By analyzing these bacteria, there may be potential to reveal the effects of their environments on ecosystems. In this study, we will be comparing the biodiversity of bacteria within oysters due to their functions in marine life (Hines et al., 2023). This will be accomplished by targeting oysters from two different markets located on the East Coast: Blue Point, Long Island in New York, and Plymouth Rock Oysters in Massachusetts. The waters at Blue Point are prone to poor water quality, due to the urban residues that tend to accumulate (Bosworth, 2019). On the other hand, Plymouth Rock waters are tended for: The company that would provide oysters maintains the oysters to supply consumers with specimens from conserved environments.

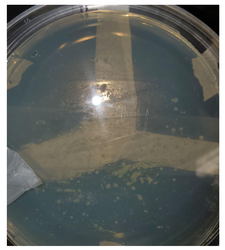
Methodology

1) Shucking and swabbing the gills of the oysters.

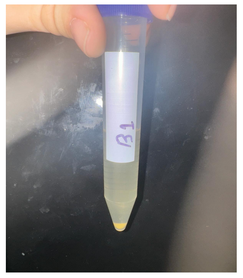
2) Plating and streaking the swabs onto designated nutrient agar plates.

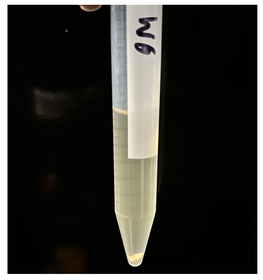
3) Allowing the growth of the bacterial colonies.





4) Allowing singular colony growth in LB broth for pelleting and DNA extractions





Results

Once our samples were returned from being sequenced, we BLASTED the samples that were successful and we received the following results in figure 1. Only three of our samples were able to be identified, though these also had poor quality of DNA.

Conserved Area Oysters: Wellfleet, Massachusetts
Bacteria Identification
Sample:

KEM-014-R	**Moraxellaceae bacterium or staphylococcus**
KEM-016-R	**Bacillus thuringiensis**
KEM-017-R	**Agrobacterium tumefaciens**

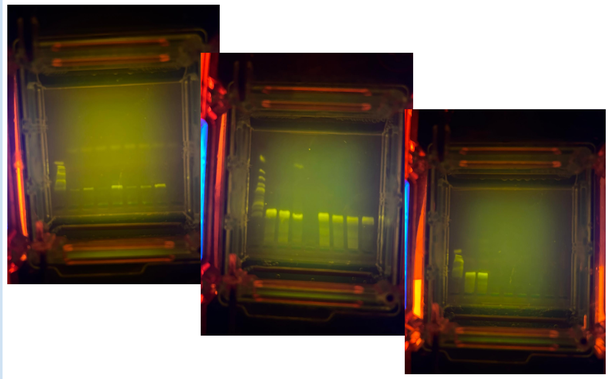
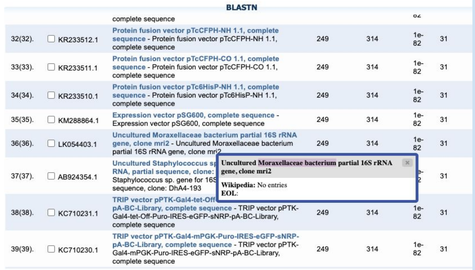


Figure 1: The following gels illustrate the DNA fragments from the samples. There are eight samples in each of the gels: Gel no.1 has the samples 1-8 of Blue Point Oysters, and gel no.2-3 has the samples 9-18 of Wellfleet oysters (reading from left to right). Compared to the ladder, there were predicted to be a large number of successful sequences due to the bands present on the gels.

Even though were able to identify only three of the 18 bacterial samples, our identifications would be considered inaccurate due to the busy DNA sequences. Majority of our samples were unsuccessfully sequences. When the BLASTS were conducted, there was no clear identification of a possible bacteria species. Therefore, we had to pick and choose the possible bacterial species.



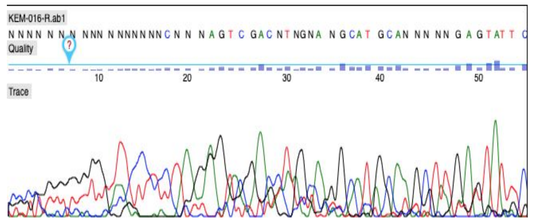


Figure 3: The sample conditions of KEM-016-R is illustrated, which is consistent with all three of the samples that were successfully sequenced. There is no clear DNA sequence due to the high amounts of disturbances that are present.

Discussion

Out of 18 of the possible samples, only three were able to be identified even though there were poor sequences. The bacteria that was most likely present in sample KEM-014 appeared to be Moraxellaceae bacterium, which is a species of psychotropic bacteria. This species commonly thrive within a wide range of habitats ranging from soils to within respiratory tracts of animals (Yang, X. 2014). Another possible identification of sample KEM-014, is staphylococci, which are commonly found on the human skin, which can lead to infections. Thus, they tend to thrive in environments within poor conditions such as contaminated waters and soil. Sample KEM-016 was identified to be possibly Bacillus thuringiensis, which are a form of bacteria found in plants that are used within insecticides (Ibrahim MA et al. 2010). This was found by researching one of the BLAST results with minimal differences, “Chloroplast transformation vector pN-IC101.” There most likely was a strain of this bacterium residing within the oyster that was swabbed, due to there being a transformation occurring within this sample. Sample KEM-017 was identified to possible be Agrobacterium tumefaciens, which is an invasive species of bacteria that thrives in infested soils, which can form tumors on the roots of a variety of different types of plants (Meyer T, et al. 2019). This was found by researching one of the BLAST results with minimal differences, “Tobacco plastid transformation vector pSS33.”

The identifications for sample KEM-014 may be a contaminant due to this species commonly being found on the human skin. However, this can also most likely be the species that was originally found from the oyster that was swabbed. There can not be a definite origin point of this sample since there were too many disturbances within their sequences. On the other hand, the identifications for samples KEM-016 and KEM-017 tend to thrive on a variety of plants and insecticides, which can yield to the conclusion that they were present within the oysters found at Wellfleet due to there being a consistency of human activity of tending to the environment.

Next Steps

Based on other observations and studies, bacteria like Arcobacter, Spirochaeta, Pseudoalteromonas, Marinomonas, Fusobacterium, Psychrobacter, Psychromonas, and Oceanisphaera are expected to be found. Notably, Psychrobacter and Psychromonas are the most found bacteria in oysters situated in a polluted environment. The reasons why our results are limited could be due to the following: improper handling of oysters, contamination in equipment, long periods of the oysters being exposed to freezing temperatures, and/ or oysters being thawed several times, small number of samples, not properly following procedures, and running too much DNA in the gel electrophoresis. By analyzing the results of other research studies, we conclude that if oysters are located in an unclean environment, bacteria like the ones previously mentioned are guaranteed to be found in the organism, particularly in the gills. Hence, it is vital to maintain specific and healthy environments for oysters to ensure their safety and quality for human consumption. In order to yield accurate and clear results in the future, we will follow this same protocol and be particularly careful with our bacteria samples to prevent minimal contamination and degradation. Since now we have a clear set of methodology and an idea of external factors that skew our data, we can repeat this analysis with further experience in order to have viable data to use.

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