Microbial Air Sampling in and around NYC

Laura Andersen, Nushera Nahia and Dr. Bryan Wilkins

Manhattan Center for Science and Mathematics, DeWitt Clinton High School

URBAN BARCODE RESEARCH PROGRAM
Supported by the Pinkerton Foundation & Science Sandbox

CSH Cold Spring Harbor Laboratory DNA Learning Center

Abstract

Microorganisms are a vital part of nature, human health, and to an ecosystem. They live in water, in soil, on the skin, and in digestive tracts. There are several types of microorganisms, including bacteria, fungi, protozoa, algae, and viruses. However, the types of microorganisms in the air can depend on their location and their environment. Our project is aimed at identifying microorganisms in three different areas in, and surrounding, New York City: The Bronx, Manhattan, and Westchester County. We used passive agar plate air sampling to collect and identify common microorganism colonies.Using DNA extraction, PCR amplification, and DNA Barcoding we identified several colonies that settled on our plates. This project helped us understand which microorganisms are common within one city.

Introduction

Microorganisms, also known as microbes, are microscopic organisms and make up a significant part of the planet's living material. They are crucial in maintaining Earth's ecosystem and can be multicellular, unicellular or cell clusters. The most common microorganisms are bacteria, fungi, protozoa, archaea, algae, and viruses and they are found in nearly every environment. Contrary to popular belief, most microbes are not harmful to humans and perform many important roles such as producing oxygen, providing plants with nutrients, decomposing dead material and aiding in digestive systems. Certain types of microbes, however, can be pathogenic or harmful to human health and many of these microbes may also be present in the air. Identifying and studying the different microorganisms that exist in the air, especially microbes that may be harmful, allow for the discovery of critical links between airborne microbes and health and illness or disease. Passive sampling, using agar plates to capture and grow microorganisms, is a simple method for collecting samples. Each microbe species can be identified using DNA barcoding. This approach can help us find specific microbes that may be more prevalent in a particular areas of the city.

Materials & Methods

We used agar plates to collect airborne microorganisms in three different locations, the Bronx, Manhattan, and Westchester county, to identify and compare the difference between these locations. We sampled with three plates per location and the plates were exposed to the environment for 15, 30 and 45 minutes, and then immediately closed and sealed with parafilm. The plates were incubated at room temperature for 4 days and then imaged. Microbial colonies were chosen for genomic DNA isolation using Chelex kits that were provided by the UBRP. We then performed PCR amplification of the 16S rRNA gene using universal 16S rRNA primers. The PCR products were gel purified, sequenced, and analyzed using the DNA subway bioinformatic tool.

Results

From the collection plates (fig. 1) we observed a variety of different types of microbes based on size, color, and form, across the environmental areas we sampled. We chose several different microbes for barcoding from our Westchester and Manhattan plates (fig. 2A and 3A). Genomic DNA was isolated the organisms and verified via agarose gel electrophoresis (fig. 1B and 2B). 16s rRNA and Internal Transcribed Spacer (ITS) region primers were used for amplification of each genomic region. The PCR products were verified via agarose gel electrophoresis (fig. 2C and 3C), and then sequenced. Sequencing results were BLASTed using DNA Subway. These results are summarized in table 1.

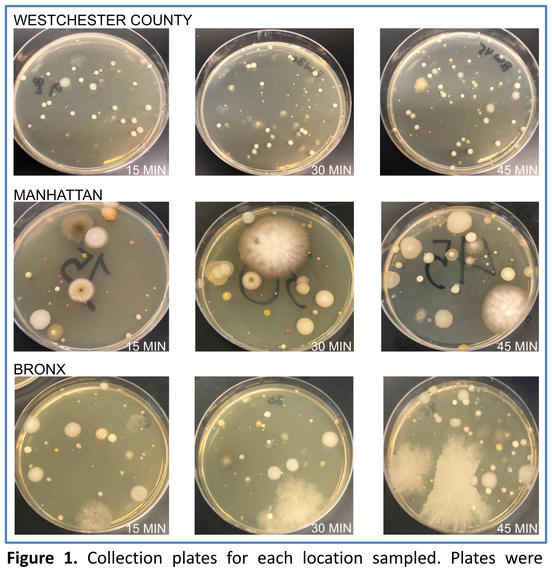

Figure 1. Collection plates for each location sampled. Plates were exposed to the air for 15, 30, and 45 minutes.

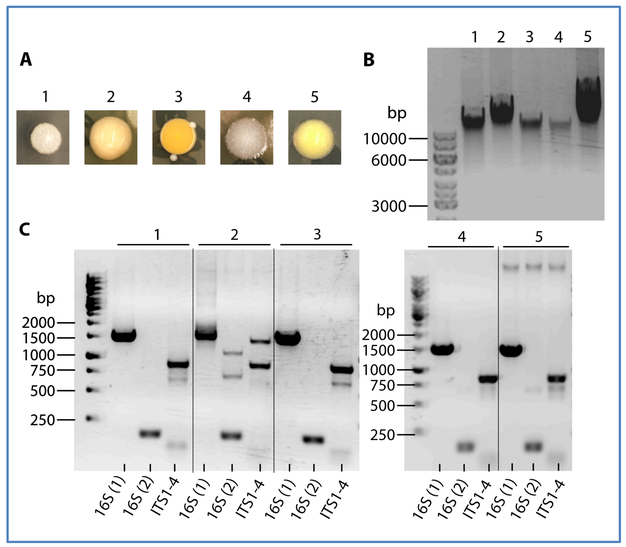

Figure 2. A) Microbe samples collected in Westchester County, NY. **B)** Agarose gel stain for genomic DNA isolation of the microbes sampled. **C)** Agarose gel stain for the PCR barcoding products using 2 different sets of 16s rRNA primers and the ITS region primers.

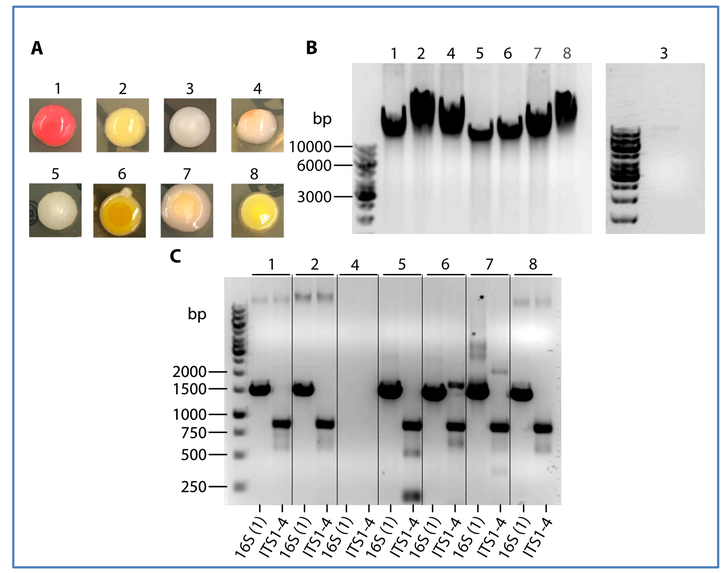

Figure 3. A) Microbe samples collected in Manhattan, NY. **B)** Agarose gel stain for genomic DNA isolation of the microbes sampled. **C)** Agarose gel stain for the PCR barcoding products using 16s rRNA primers and the ITS region primers.

Westchester County	Manhattan
Bacillus Megaterium	Arthrobacter agilis
Pseudomonas lini	Labedella endophytica
Exiguobacterium artemiae	Bacillus sp.
Bacillus sp.	Chryseobacterium sp.
Pseudomonas donghuensis	Psychrobacter sp.
	Okibacterium fritillariae

Table 1. Microbe image and species identification via DNA BLAST

Discussion

Overall, we were successful in identifying airborne microorganisms from different locations in New York such as Manhattan and Westchester County. Our primers worked in the range expected (1500 and 800 bp). We were unable to analyze all the samples that we intended to, due to complications resulting from remote work, and time limitations, but we were able to analyze enough samples that highlighted most of the aims of our research. Using the 16s rRNA barcode to blast for species identification, we successfully learned how to use DNA sequencing and bioinformatics as a way of identifying and classifying a living organism.

References

1. "What Are Microbes?" InformedHealth.org [Internet]., U.S. National Library of Medicine, 29 Aug. 2019, www.ncbi.nlm.nih.gov/books/NBK279387/.
2. Libretexts. "1.2A Types of Microorganisms." Biology LibreTexts, 3 Jan. 2021, bio.libretexts.org/Bookshelves/Microbiology/Book:_Microbiology_(Boundless)/1:_Introduction_to_Microbiology/1.2:_Microbes_and_the_World/1.2A_Types_of_Microorganisms.
3. OpenStax. "Microbiology." Lumen, courses.lumenlearning.com/microbiology/chapter/types-of-microorganisms/.