Effects of Flowing Water on Bacterial Growth

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Abstract

The East River is a salt water tidal strait, meaning it connects to the ocean at both ends, the New York harbor at one end and the Long Island Sound at the other. Due to this fact the East River is made up of brackish water and is also subject to the tides, meaning the direction the river flows can change 2-3 times a day, rising tide results in the river flowing north and east, while ebbing tide results in a south and west flowing river. The question we set to explore was how flowing water affects bacterial growth and diversity. We did this by comparing the microbiomes obtained through metagenomic analysis from two sites, Long Island City (LIC) ferry terminal, and 34thst ferry terminal. We ran a series of experiments on water samples from both sites testing various markers for water quality, including dissolved oxygen content through titration, salinity through evaporation and dialysis, dissolved carbon dioxide by titration; temperature and pH were measured at the time of collection as well. For the metagenomic analysis, we extracted DNA from our samples using an isolation kit, and samples were sent for sequencing and analysis using 16S rRNA to determine the bacteria population in samples from both locations. For the deep water sample from 34th st presented 91 classes of bacteria were detected, while the sample from LIC had 32 bacterial classes. Our intention was to compare the environment at both sites and using the data from our experiments determine whether the direction the water flows plays a role in bacterial growth. In the future we would expand the number of sites along the river to gain a wider variety of data and how it relates to tides.

Introduction

There are many factors that contribute to bacterial growth and diversity including salinity, dissolved oxygen and carbon dioxide, pH, temperature as well as chemical factors within their environment. Our goal with this research was to rule out as many of these factors as possible to determine if water flow has a demonstrable effect on bacterial growth and diversity. The body of water we examined was the East River, a tidal strait consisting of brackish water that changes direction of flow 4 times a day based on the tides (The RITE Project, 2019). We examined two sites along the East River, one at the 34th street ferry terminal, and the seconded located slightly north at the Long Island City ferry terminal. The sample from the 34th street was taken at 7:30am, while the Long Island City sample was taken at 7:50am, both samples were taken on 9/26/19 at a period of rising tide. The movement of bacteria in flowing water has been thoroughly researched, according to ESPCI Paris, an institute of higher learning in France, the flow of water can effect the movement of bacteria. In slow moving water bacteria has been found to simply rotate in a circle, as you increase the flow of water bacteria has been found to move against the stream (ESPCI, 2019). According to research done by the Massachusetts Institute of Technology if you increase the flow to extreme rates the bacteria are likely to attach themselves to surfaces (MIT, 2014). With these facts in mind we set out to compare the two sites. The two sites we compared had very similar results for dissolved oxygen, carbon dioxide, pH, and temperature. The one difference we found between the two sites were the salinity levels. With the two sites having very similar environments it presented us with a good opportunity to see how the flowing water effected the bacteria in each site. While the number of locations was limited it is the beginning of a process that could possibly lead us to methods that would allow us stop bacteria flow through water and create dams that may allow for the removal of dangerous strains of bacteria from flowing water. With bacteria adhering to surfaces at points of high water flow it may be possible to create surfaces for the bacteria to attach to that could then be removed with the bacteria still attached. Here we present our findings on the environmental factors and metagenomic analysis from the two sites.

Materials and Methods

Direct evaporation of 10 ml of each water sample from LIC deep and East River deep was used in determining the total dissolved solid. Additionally, salinity was also measured by constructing a standard curve regulated by dialysis bags with varied known percentages of NaCl solutions. Temperature and pH were determined at the time of water sample collection. Lauryl tryptose broth and TSA-agar plates were used to determine the presence of bacteria. Extraction process to isolate DNA as one type of organic molecule by purifying water samples was also performed using MoBio WaterPower kit. Concentrations of dissolved oxygen and carbon dioxide measurements were assayed by LaMotte dissolved Oxygen and Carbon Dioxide titration kit. The tidal data at the time of the sample collection was obtained from *World Wide Tide and Currents Predictor* (https://tides.mobilegeographics.com/).

Results

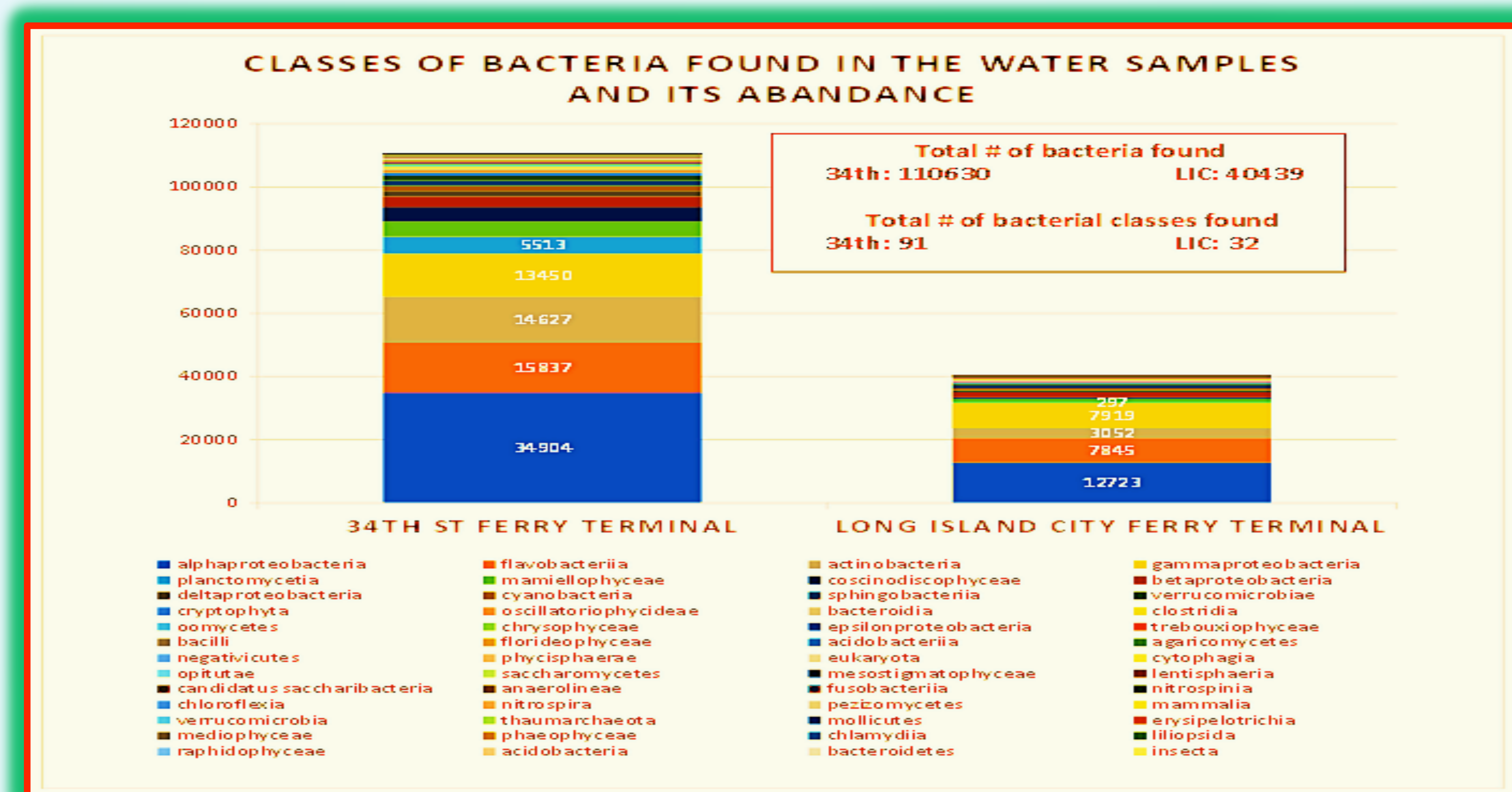

Figure 1. The results of the metagenomic analysis of DNA from each water sample indicates that the water from 34thst ferry terminal, which was collected when the tidal height of East River was lower and its current velocity was faster, has greater population as well as variety of bacteria than the water from Long Island City ferry terminal which was taken when the tidal height was higher and the curent velocity was slower . (Collection Time/Data of Water sample tested: 34th Ferry Terminal 7:30AM 09.24.19 / LIC Ferry Terminal 7:50AM 09.26,19)

Location	Salinity (mg/L)	pH	Temperature (℃)	Dissolved O_2 (ppm)	Dissolved CO_2 (ppm)
34thst Ferry Terminal	29,000	7.8	20	5.8	25
LIC Ferry Terminal	24,000	8.0	21	4.6	N/A

Table 1. Summary of Physical and Chemical Characteristics of the Water Samples. The only significant difference observed between these two samples was the dissolved oxygen level while both samples display similar results in the other test items.

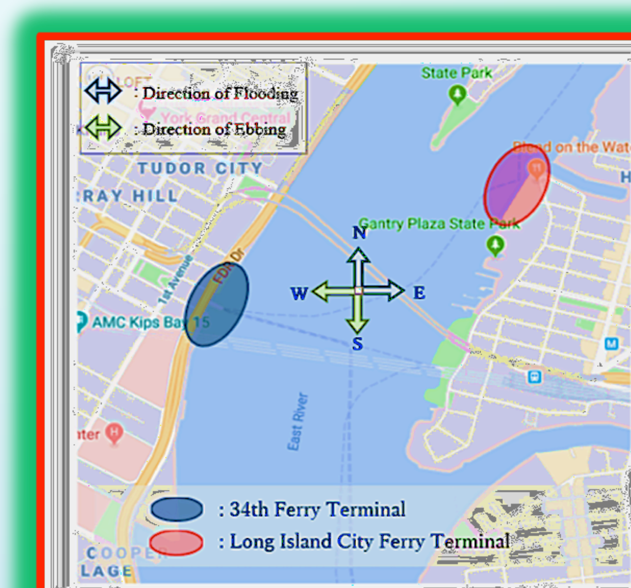

Figure 2. Map with the two locations where water samples were collected. East River flows towards North and East when the tide is flooding, then it flows backwards South and West when the tide is ebbing. Photo was taken from Google.

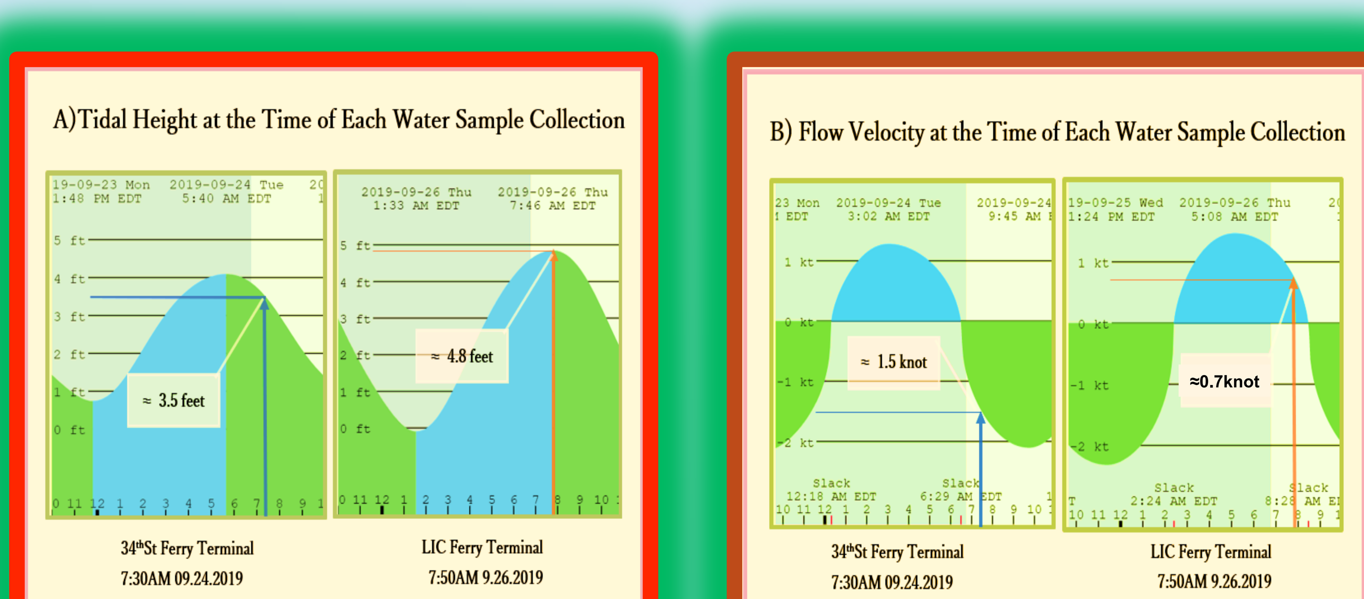

Figure 3, Figure 3-A and B Shows different tidal heights and flow velocity of East River at the time of collections of each sample. When 34thst sample was taken, the tidal height of the site was approximately 3.5 ft and the flow velocity was approximately 1.5kt. On the other hand, when LIC water sample was taken, the tidal height of the site was approximately 4.8ft and the flow velocity was approximately 0.7 kt. Photo taken fromhttps://tides.mobilegeographics.com/

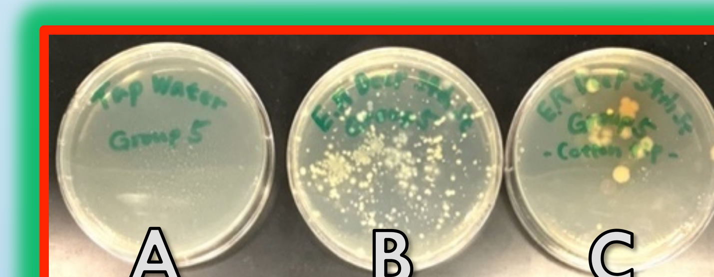

Figure 4. Visible bacterial growth in agar plates with water samples added (B and C in figure) provided evidence for the presence of bacteria in the water samples; the agar plate A with tap water added showed no visible bacterial growth.

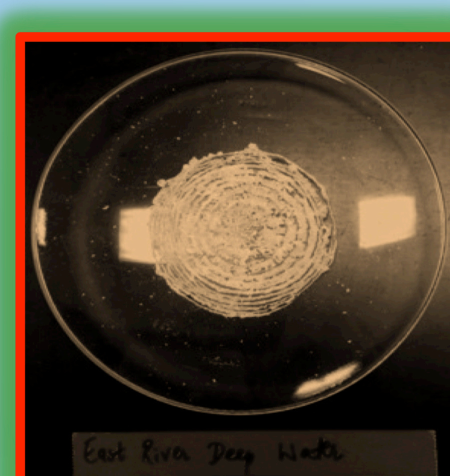

Figure 5 (LEFT). 0.29g of salt was collected after evaporating 10.0mL of 34thst water sample. From the mass, Total Dissolved Salt was directly calculated.

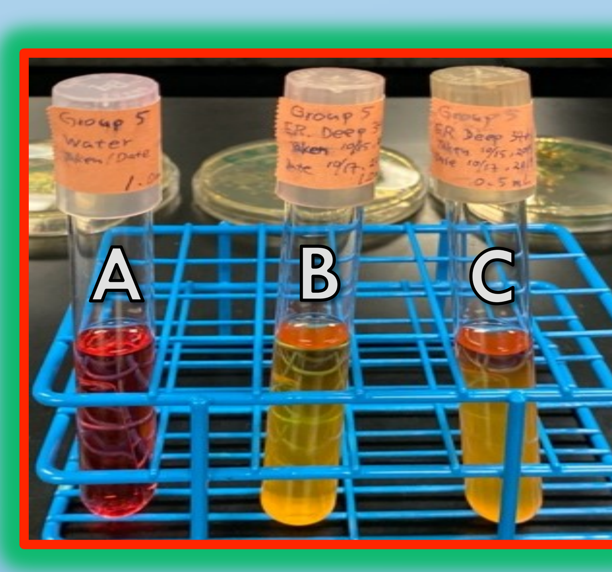

Figure 6 (LEFT). Inoculation of lactose broth with 34thst water sample confirmed the presence of enterobacteria bacteria, which are atypical of brackish water. Turbidity and color change of test tubes B & C, which contained water sample 0.1ml and 0.5ml respectively, showed the existence of Coliform bacteria in water sample.

Discussion and conclusions

The objective of our research was to compare the bacterial abundance in two different sites; East River (deep) and Long Island City (deep) considering different factors that affect the growth. Bacterial growth is affected by the temperature, pH, dissolved oxygen, salinity and tide.

The results we obtained from measuring the salinity of our sample confirm its characterization as brackish water. The bacterial classes that we found in both water samples were mesophiles (surviving in 20-45 degrees)

Through the inoculation of lactose broth with water sample from ER, presence of anaerobic bacteria was confirmed. While observing the turbidity of the B and C test tubes with water sample, no gas formation was observed but the color of broth changed into yellow which is a sign of production of acid by bacteria present in the water sample as opposed to test tube A which was a control sample of distilled water. These results imply the presence of Enterobacteria, a form of bacteria that is responsible for a range of conditions in humans. These conditions can include, meningitis, bacteremia (a blood based bacterial infection), urinary tract infections, eye and skin infections and pneumonia.

Another experiment to see whether there is bacteria in the water sample was by using agar plates to culture microorganisms. After a week of incubation, bacterial growth was seen in the B and the C agar plates which had a sample of water from the east river and the A agar plate was clear with no microorganisms due to the sample being tap water.

While comparing the class percentage of bacteria found in both sites through metagenomic analysis, we found a significant difference in bacterial classes. The New York tide chart shows that the tide has just moved from ebbing to flooding in LIC and ER on the day and the time when water samples were taken. The results of the study done by Kirchman et al. showed relatively high abundance of bacteria attached to surfaces during ebbing tide. (Kirchman et al. ,1984). The only measurable differences between the two samples is the tide and speed of water flow. The sample drawn from LIC was taken during a period of flooding tide at a water speed of 0.7 knots, while the sample drawn from the East River was taken at a period of ebbing tide at a water speed of 1.3 knots. This shows that during periods of lower tide and higher water flow speed bacteria are more likely to congregate and attach to surfaces in areas that match these criteria.

Future work

In order to ascertain any correlation between direction and intensity of water flow with the amount and variety of bacteria build up a more extensive experiment would need to be conducted. We would need to examine sites up and down the river at various times of day at both rising and ebbing tide as well as points of fast- and slow-moving water flow and submit them for metagenomic analysis in order to compare the amount and strains of bacteria present.

It would be beneficial to create artificial surfaces of various materials for the bacteria to cling to at points of fast flowing water in order to ascertain which materials the bacteria are most likely to adhere to. This would allow us to find what materials would be best to create dams that would stop the bacteria from continuing to move through the water.

References

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