Bacterial diversity and abundance near and far from drainage pipes at DuPage County lakes

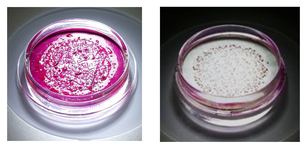
Eileen Colwell[1], Vishal Patel[1] and Helena Puche[2]
[1]DePaul University; [2] Harry S. Truman College

Bacteria and Water Quality

The abundance and diversity of bacteria can be an indicator of water quality[1]. We hypothesized that water drainage pipes would be a vessel of pollutants from the surrounding roads to the water. This lead us to ask the question: **Is there a difference in the abundance and diversity of bacteria near and far from drainage pipes?**

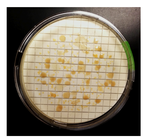
Specifically, we wanted to measure the presence of indicator organisms. **Indicator organisms** are easily measured organisms that signal the presence of contamination and pathogens[2]. The two indicator organisms that we chose to measure were *Escherichia coli*, and *Enterococci***.**

- ***Escherichia coli*** are gram-negative, facultative anaerobes, commonly found in the intestines of warm-blooded animals.
- ***Enterococci*** are gram-positive, facultative anaerobes. They are the leading cause of hospital-acquired secondary infection. They thrive in slightly basic conditions (pH > 7)[3].



m-Endo agar with *E. Coli* (left); m-Enterococcus agar with *Enterococci* (right)

We also wanted to measure **bacterial diversity**. We were curious to see what species of bacteria were living in the water at different locations.

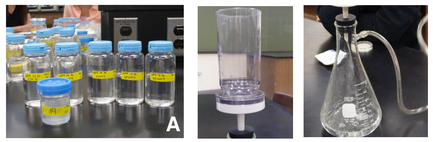


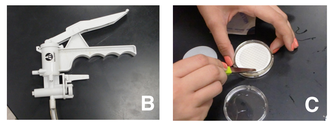
BHI broth, a permissive media, with a diverse array of bacterial species

Materials and methods

1. Collection
   - Two (100 mL) water samples were taken at each location
   - All locations were marked with GPS

2. Dilution & Filtration
   - Water was diluted at ratios of 1:10, 1:100, 1:1,000[A]
   - Diluted samples were filtered through a 45 µm filter using a hand pump[B]
   - Filters were placed on agar plates and incubated[C]





3. Total Aerobic Plate Count
   - BHI broth (permissive media)
   - m-Endo agar (tests for *E. coli)*
   - m-Enterococcus agar (tests for *Enterococci*)

To further identify bacteria species:

4. EMB agar plate
   - SD media
   - Gram-negative bacteria
5. Slide mounts
6. Oxidase test
   - Identifies aerobic bacteria
7. *Enterococcus* test
   - bile-esculin agar slant
   - indole test
8. BBL Enterotube II
   - Self-contained test with 12 media and determines 15 biochemical reactions

Flooding slides with crystal violet dye

To test for bacterial diversity:

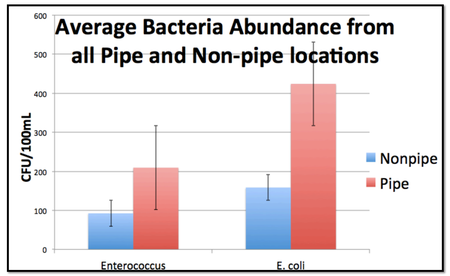
- We used the Shannon-Wiener diversity index

$$H = -\sum_{i=1}^{S} p_i \ln p_i$$

- H = Index
- P_i = proportion of species
- Ln = natural log (base e)

Results

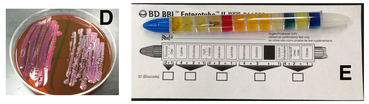
From our total aerobic plate count, we found that the numbers of ***E. coli* and *Enterococci*** were not statistically different between pipe and non-pipe locations (Kruskal Wallis ANOVA, H = 5.3; df = 4; P > 0.05).



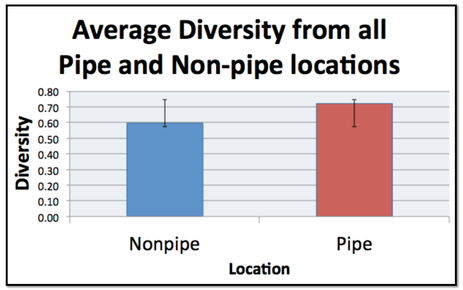
Average count of colony forming units (CFU) per 100 mL of *Enterococci* and *E. coli* at pipe and non-pipe locations.

From the results of our identification experiments (i.e. EMB agar plate[D], slide mounts, oxidase test, *Enterococcus* test, and Enterotube[E]), We were able to confirm the presence of the following bacteria:

- ***Citrobacter freundii***
- ***Enterococcus faecalis***
- ***Enterobacter clocae***



From our diversity calculations, the **diversity index between pipe and nonpipe locations was not statistically different**.



Average diversity of pipe and non-pipe locations, based on the Shannon-Wiener diversity index. A higher diversity index value indicates a more diverse location (Mann Whitney U test: U = 49.5 ; df = 1; P > 0.05).

Conclusions

The abundance of *E. Coli* and *Enterococci* were not different between pipe and non-pipe locations. This lead us to ask the question: **Are there other environmental variables that are shadowing the effects of drainage pipes?**

- Several congregates of ducks and geese were observed at our test sites.
- We found *Enterobacter clocae,* a bacteria found in the intestines and fecal matter of birds.
- The presence or absence of bird species may have overshadowed contamination from water drainage pipes.

Literature cited

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Acknowledgments

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Contact Information

Eileen Colwell eileenmcolwell@gmail.com
Vishal Patel vpatel56@mail.depaul.edu
Helena Puche hpuche@ccc.edu