Battle Of the Shrimps

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Funded by the Thompson Family Foundation

Introduction:

Figure 1. *Hyalella puna*

- Amphipods are crucial to the environment— they recycle nutrients, scavenge, and are a major food source for animals (New World Encyclopedia, n.d.)
- Pollution from humans is affecting ecosystems:
  - Van Cortlandt Lake—one of the cleanest lakes in NYC—is considered eutrophic (Department of Environmental Conservation, 2009)
- Greater ammonia, nitrate and phosphates contributes to more algae
- More algae means less oxygen because bacteria use the oxygen to decompose dead algae
- These factors, phosphates, nitrates, and ammonia could impact the biodiversity of amphipod species

Materials and Methods:

- The amphipods samples were collected in Van Cortlandt Park in 3 different locations. Amphipods of similar phenotypic features (color and size) were placed in the same vial.
- The amphipods were crushed up and had their DNA extracted. They were put in 300 mL lysis solution and then went through a process of centrifuging, removing supernatant, heating up, and vortexing to isolate the DNA.
- PCR was performed using COI primer set and ready-to-go PCR beads with dehydrated Taq polymerase, nucleotides, and buffers. Then samples were put through the thermal cycling possess.
- Finally, electrophoresis was performed to analyze the PCRs. A 2% agarose solution with 0.25% of Gelgreen® Nucleic Acid Gel Stain was used to create the wells. The surface was covered with TBE buffer. A 100-bp marker was used as a control and 5 microliters of the samples were put in the remaining wells. The gel was run in the electrophoresis chamber at 130V for 30 minutes. The PCRs were then viewed with UV transillumination.

Results:

#	Accession #	Details	Aln. Length	Bit Score	e	Mis-matches
1(1).	MG318747.1	**Hyalellidae sp. BIOUG23330-A05 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23330-A05 cytochrome oxidase subunit 1 (COI) gene, partial cds	526	922	0.0	6
2(2).	MG317537.1	**Hyalellidae sp. BIOUG23329-F12 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23329-F12 cytochrome oxidase subunit 1 (COI) gene, partial cds	526	922	0.0	6
3(3).	MG316840.1	**Hyalellidae sp. BIOUG21719-D04 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG21719-D04 cytochrome oxidase subunit 1 (COI) gene, partial cds	526	922	0.0	6
4(4).	MG312019.1	**Hyalellidae sp. BIOUG23329-B04 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23329-B04 cytochrome oxidase subunit 1 (COI) gene, partial cds	526	922	0.0	6

Table 1. Blastn Results of Sample 4 (forward and reverse). The four most probable sequences produced by Blastn (Basic Local Alignment Search Tool) for sample 4 are displayed with the corresponding accession #, a unique identifier for each sequences; the organism and sequence description; the alignment length; the bit score, a measure how aligned the sequences are; the e-value, the certainty of the results (lower is more probable); and the number of mismatches.

#	Accession #	Details	Aln. Length	Bit Score	e	Mis-matches
1(1).	☐ MG318747.1	**Hyalellidae sp. BIOUG23330-A05 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23330-A05 cytochrome oxidase subunit 1 (COI) gene, partial cds	589	1031	0.0	7
2(2).	☐ MG316840.1	**Hyalellidae sp. BIOUG21719-D04 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG21719-D04 cytochrome oxidase subunit 1 (COI) gene, partial cds	589	1031	0.0	7
3(3).	☐ MG312019.1	**Hyalellidae sp. BIOUG23329-B04 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23329-B04 cytochrome oxidase subunit 1 (COI) gene, partial cds	572	1001	0.0	7
4(4).	☐ MG317537.1	**Hyalellidae sp. BIOUG23329-F12 cytochrome oxidase subunit 1 (COI) gene, partial cds** - Hyalellidae sp. BIOUG23329-F12 cytochrome oxidase subunit 1 (COI) gene, partial cds	559	982	0.0	6

Table 2. Blastn Results of Sample 16 (forward and reverse). The four most probable sequences produced by Blastn (Basic Local Alignment Search Tool) for sample 16 are displayed.

#	Accession #	Details	Aln. Length	Bit Score	e	Mis-matches
1(1).	☐ KX396277.1	**Phytophthora boehmeriae isolate ALT-18 cytochrome oxidase subunit 1 (cox1) gene, partial cds** - Phytophthora boehmeriae isolate ALT-18 cytochrome oxidase subunit 1 (cox1) gene, partial cds	643	395	1e-106	159
2(2).	☐ MH136922.1	**Phytophthora macilentosa strain ex-type CPHST BL 125 cytochrome c oxidase subunit 1 (COI) gene, partial cds** - Phytophthora macilentosa strain ex-type CPHST BL 125 cytochrome c oxidase subunit 1 (COI) gene, partial cds	635	379	3e-102	160
3(3).	☐ MN866111.1	**Phytophthora macrochlamydospora isolate VN1006 cytochrome oxidase subunit I (cox1) gene, partial cds** - Phytophthora macrochlamydospora isolate VN1006 cytochrome oxidase subunit I (cox1) gene, partial cds	643	377	4e-101	163
4(4).	☐ GU594810.1	**Phytophthora captiosa strain P10719 cytochrome c oxidase subunit I (COX1) gene, partial cds** - Phytophthora captiosa strain P10719 cytochrome c oxidase subunit I (COX1) gene, partial cds	643	377	4e-101	163

Table 3. Blastn Results of Sample 5 (forward). The four most probable sequences produced by Blastn (Basic Local Alignment Search Tool) for sample 5.are displayed.

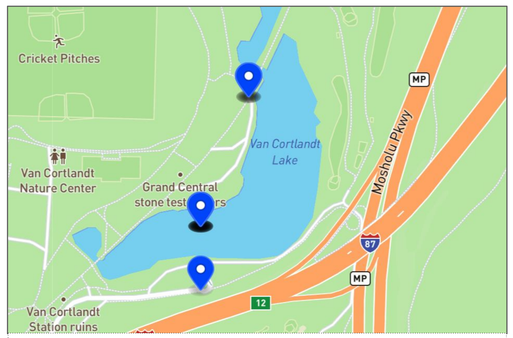

Figure 2. Map of Locations in Van Cortlandt Lake Where Amphipods Were Collected. Site 1 (bottom) refers to the edge of a freshwater pond located near a oak tree. Site 2 (top) refers to a freshwater lake in a marshy region under a bridge. Site 3 (middle) refers to a freshwater pond by a forest edge located near a bald cypress tree.

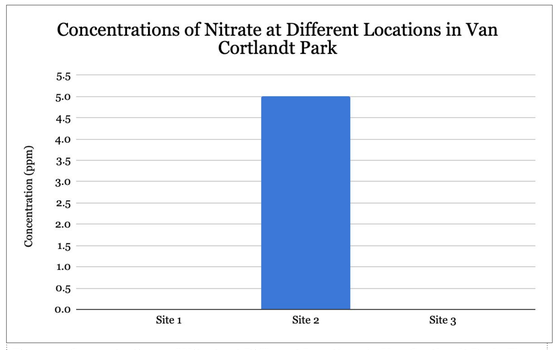

Figure 3. Concentrations of Nitrate at Different Locations in Van Cortlandt Park. There was 0 ppm concentration of nitrates in sites 1 and 3, and 5 ppm in site 2.

Discussion:

- 2 viable samples
- Likely from Hyalellidae family
- High bit scores and low e-values with few mismatches
- 3 samples were 'blasted', but one was classified a plant pathogen with high mismatches and low bit score (sample 5)
- Inconclusive species
- Phosphate tests were inconclusive
- Concentration of nitrates or phosphates cannot be correlated to the biodiversity of amphipod species
- Not enough samples

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Acknowledgements:

1) Mr. Howard M Waldman
→ Our science research mentor and coordinator
2) Van Cortlandt Park Alliance
→ Guided us to collect our amphipod samples in Van Cortlandt Park