

Cyanobacteria of the Klamath Basin

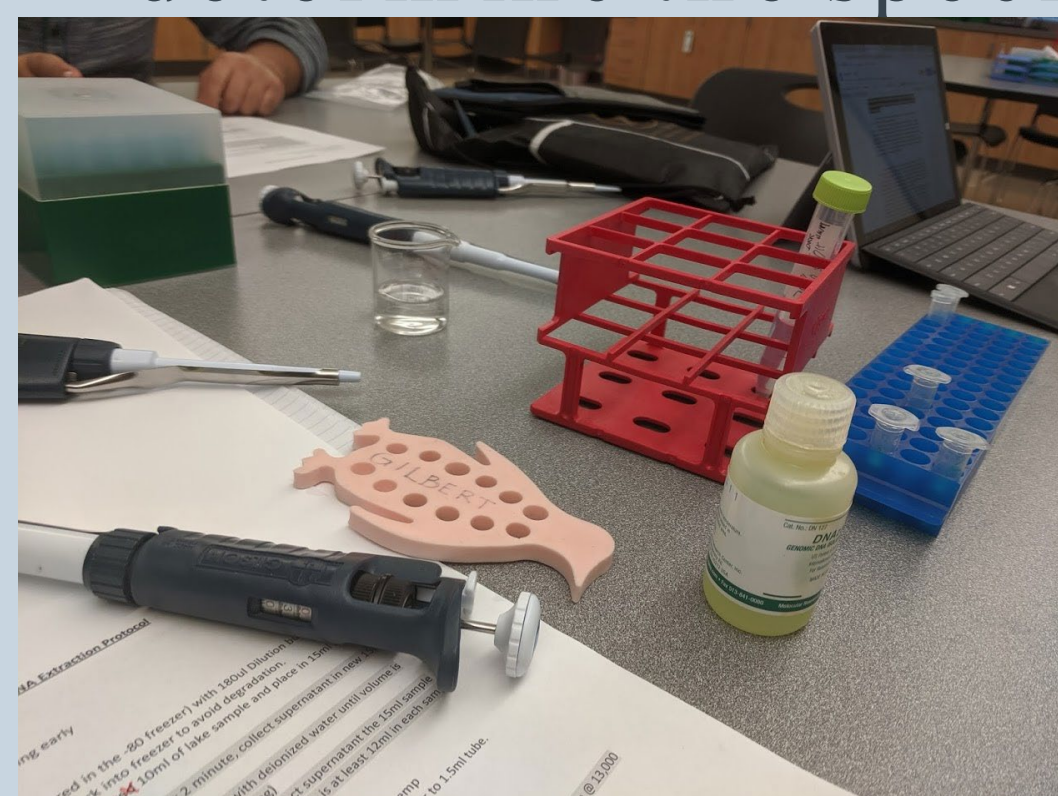
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1. Introduction

There are many species of cyanobacteria present in Upper Klamath Lake. Some species are used as nutritional supplements while others produce a toxin implicated in several neurodegenerative diseases and commonly kills livestock, pets, and wildlife. Because of these effects, it is important to understand the species, growth, and fluctuation of the cyanobacteria.

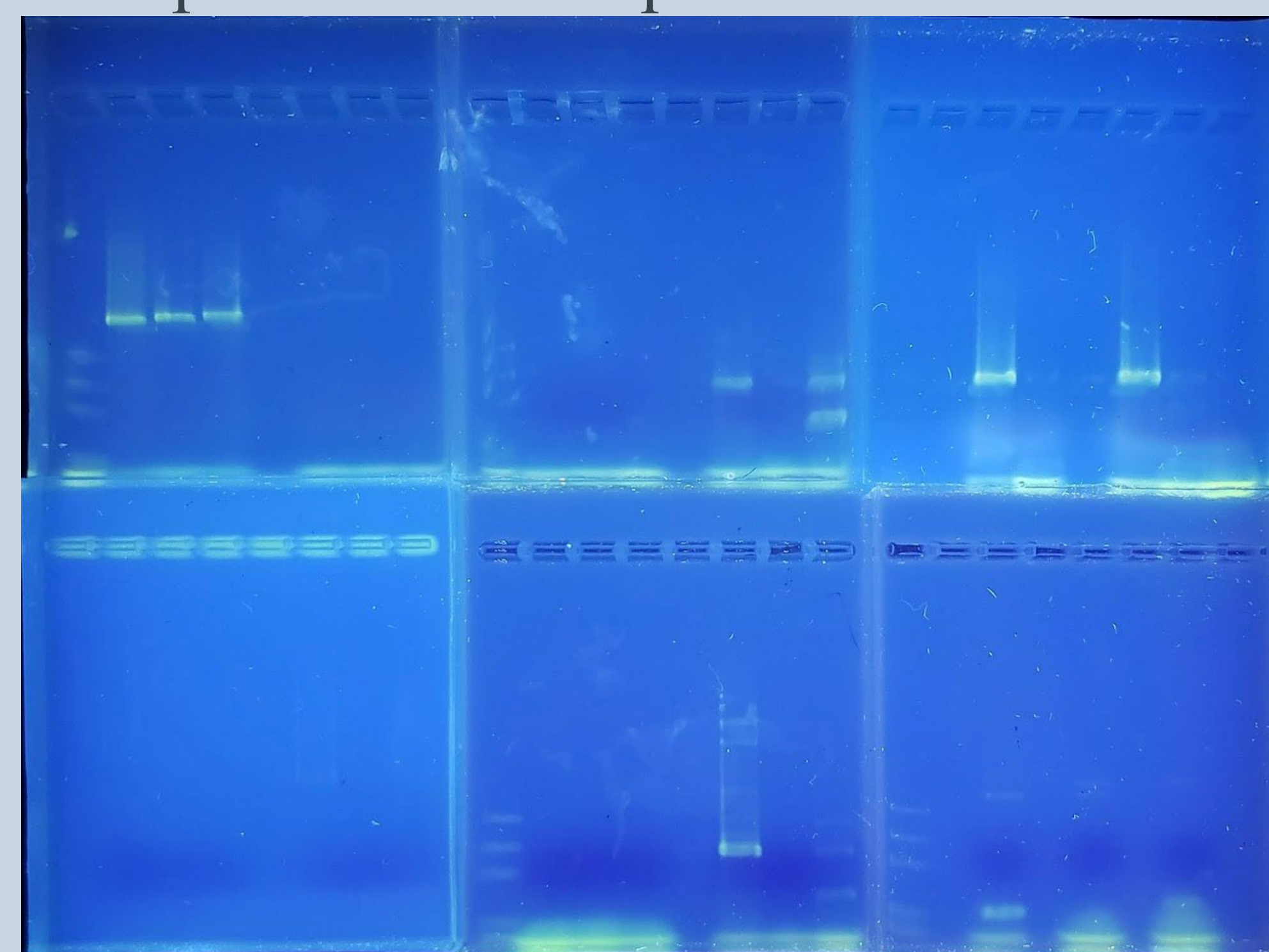
2. Methods

1. Collect samples from Upper Klamath Lake
2. Remove debris and sediment and extract DNA from the cells present
3. Use PCR to amplify specific genes known to be present in cyanobacteria
4. Use gel electrophoresis to identify results of PCR *note: this year only done on known controls.
5. Sequence the DNA from the gel to determine the species present



3. Results

In Gels 1-4, the lanes 2-4 and 6-8 represented *Anabaena*, *E. Coli*, and *Aphanizomenon flos-aquae* (AFA) amplified fragments respectively. Gels 1-3, tested for presence of *Eubacteria*, *Microcystis aeruginosa*, *Anabaena*, cyanobacteria and the *Nostoceae* family specific fragment amplification with positive and negative results as expected. In Gel 4 an error occurred. Gel 5 tested for the presence of *Arthrospira* and *Anabaena*, and in lane 8 an AFA sample was amplified for a chosen "*Anabaena* only sequence". Gel 6 tested for amplification of an "*Arthrospira* only sequence" in lanes 3, 5, and 7 but all samples were amplified.



Top_Left: Gel 1 Positive for *Eubacteria* L2-4, Negative for *Microcystis* L6-8; Top_Middle: Gel 2 Positive for *Anabaena* L3 and cyanobacteria L6 and 8; Top_Right: Gel 3 Positive for *Anabaena* L3 and AFA L6
Bottom_Left: Gel 4 Error; Bottom_Middle: Gel 5 Negative for *Arthrospira* L2-4 positive for *Anabaena* L6 and AFA L8; Bottom_Right: Gel 6 Positive for *Arthrospira* for all samples

4. Conclusion

After sequencing the positive bands from last year's gel, it was determined that there were multiple species of cyanobacteria present in the sample due to a scrambled sequence. This led to the reconstruction of the methods to more accurately target specific genes.

Using known samples of *E. Coli*, AFA, and *Anabaena* as controls, the chosen target sequences were evaluated. This was helpful in identifying some bacteria, but there were also unexpected findings. For example, in Gel 5 Lane 8 an AFA sample was amplified when the gene should not have been present, and in Gel 6 what should have been a *Arthrospira* specific sequence was identified in *Anabaena*, *E. coli*, and AFA.

From these findings it is clear that further reference is required to properly identify sequences to be utilized as species specific markers. These new species specific markers can then be used to identify specific bacteria within the samples from Upper Klamath Lake.

5. Acknowledgements

We would like to thank the Resource Budget Commission for continuing to fund this project. We would also like to thank Dr. Usher and Dr. Gandhi for their endless patience and support while guiding this group.