

The Biodiversity of Green Fluorescent Coral Zooxanthellae

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Abstract: Our project was to determine the biodiversity of zooxanthellae living in green fluorescent corals. This is a continuation of our previous project in which we identified the fluorescence of many corals. Previously, one of our mentors, Charles Mazel, told us that the proteins had the possibility of being used for medicine, which is what inspired our current project in the first place. We went through the Cold Spring Harbor DNA extraction procedure extracting and purifying the DNA from the coral tissue followed by amplifying a specific region of the chloroplast by adding polymerase chain reaction (PCR). The sequencing, however, wasn't as successful as we hoped. We believe the reason for its failure was due to the method of storage instead of our method of extraction. We will attempt a more effective way of storage for the following year.

Introduction: Our project is to identify the different species of zooxanthellae produced by corals containing green fluorescent protein. We wanted to determine if every green fluorescent coral contains the same species. We were able to identify GFP (green fluorescent protein) in a few of the corals in our school's coral tank, mainly Green Acropora and other SPS corals. Our current objective is to DNA barcode the zooxanthellae and to successfully culture the proteins that would be extracted. Our hypothesis is completely based on the successful extraction of protein from zooxanthellae.

Materials and Methods: Our samples were all corals that produced green fluorescence. We began the collection of our samples in late September and continued until mid April. All the samples were taken from our tank at school, as they are cultured here. Then we would attempt to isolate the zooxanthellae through the use of a centrifuge, which divides substances based on density. We would then put the now centrifuged zooxanthellae under a microscope where we are able to photograph it.

Results: Our 1st graph on our board has a lot of missing base pairs. We believe the reason for this is possibly because it was stored and put away for too long. Our 2nd graph also has a lot of missing pairs, possibly due to the same reason due to its similar circumstances. Our 3rd graph has the most similar base pairs, however, it couldn't be fully sequenced which may be due to the time in the freezer as well.

Discussion: We determined that our extraction methods were capable of extracting the zooxanthellae successfully, however our storage methods were flawed. This is because our imaging displayed successful

zooxanthellae extraction, but the sequencing displayed insufficient results, which were stored in saltwater, determining that saltwater was the improper liquid to store it in. We can attempt to use a different method of storage for better specimens to sequence and/or send the samples in for sequencing sooner. Next year, we will enhance our research in the outside uses for zooxanthellae in the biology and medical field along with increasing our sample sizes. We learned that our extraction methods were successful, but our storage methods weren't

References

Valdivia, R. H., Hromockyj, A. E., Monack, D., Ramakrishnan, L., & Falkow, S. (1996). Applications for green fluorescent protein (GFP) in the study of host-pathogen interactions. *Gene*, 173(1), 47–52.

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Rosental, B., Kozhekbaeva, Z., Fernhoff, N., Tsai, J. M., & Traylor-Knowles, N. (2017). Coral cell separation and isolation by fluorescence-activated cell sorting (FACS). *BMC Cell Biology*, 18(1). <https://doi.org/10.1186/s12860-017-0146-8>

Roth, M. S., Latz, M. I., Goericke, R., & Deheyn, D. D. (2010). Green fluorescent protein regulation in the coral *Acropora yongei* during photoacclimation. *Journal of Experimental Biology*, 213(21), 3644–3655. <https://doi.org/10.1242/jeb.040881>

Dove, S. G., Hoegh-Guldberg, O., & Ranganathan, S. (2001). Major colour patterns of reef-building corals are due to a family of GFP-like proteins. *Coral Reefs*, 19(3), 197–204. <https://doi.org/10.1007/pl00006956>

Mazel, C. H., Lesser, M. P., Gorbunov, M. Y., Barry, T. M., Farrell, J. H., Wyman, K. D., & Falkowski, P. G. (2003). Green-fluorescent proteins in Caribbean corals. *Limnology and Oceanography*, 48(1part2), 402–411. https://doi.org/10.4319/lo.2003.48.1_part_2.0402