

# Using CO1 DNA Barcoding to Assess Butterfly Biodiversity from the Long Island Aquarium

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## Abstract

This project analyzes butterfly species biodiversity at the Riverhead Aquarium Butterfly exhibit, providing crucial data for future conservation. A DNA database for unknown species is being generated using DNA barcoding to determine species identification. Long Island Aquarium staff collected 20 butterfly molts (upon eclosion) for DNA analysis. All specimens were legally imported from tropical farms in Costa Rica, Malaysia, and/or Kenya under USDA-APHIS guidelines. DNA was collected from chrysalis samples by swabbing, followed by silica DNA extraction. We anticipate identifying multiple species, potentially discovering new ones, and confirming or correcting traditional taxonomy identifications using the DNA barcoding system.

## Introduction

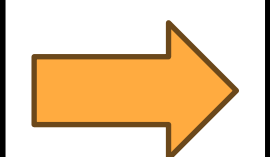
The project is a study of the biodiversity of butterflies from the long island aquarium. Butterflies go through 4 stages in their life cycle. After the 4 stages, the butterflies will reach their final form. The motivation for this is to better understand butterfly diversity. There is limited knowledge of species diversity, and many butterfly species look physically similar due to convergent evolution (live in similar but isolated habitats and have similar adaptations) By determining the butterflies' DNA barcoding scientists can use the information to measure biodiversity. The butterflies were provided to by the Long Island Aquarium.

Butterfly DNA from the CO1 gene in the mitochondria is being sequenced to create a DNA fingerprint to identify organisms. DNA barcoding is a method of specimen identification using short, standardized segments of DNA from the CO1 gene in the mitochondria. Every species has its own barcode, just as every person has their own fingerprint. These DNA barcodes can be compared to a reference library to provide an ID. It's important to study the biodiversity of butterflies because they are pollinators which have a crucial role in the ecosystem. Butterflies biodiversity needs to remain stable in order for ecosystems to work. Conserving butterflies will improve our whole environment for wildlife and enrich the lives of people now and in the future. Butterflies give intrinsic values, aesthetic values, educational values and many more. Biodiversity in an environment is important because it provides for stability in the environment and prevents the loss of organisms in the environment and prevents the loss of organisms in the environment. Biodiversity is what makes an environment unique and stable. Biodiversity contributes to a stable ecosystem where plants and animals can thrive and survive.

## Materials and Methods

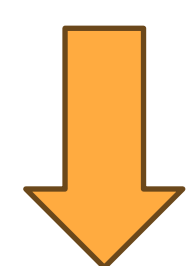
### Sample Collection

Butterfly samples were provided by the Long Island Aquarium



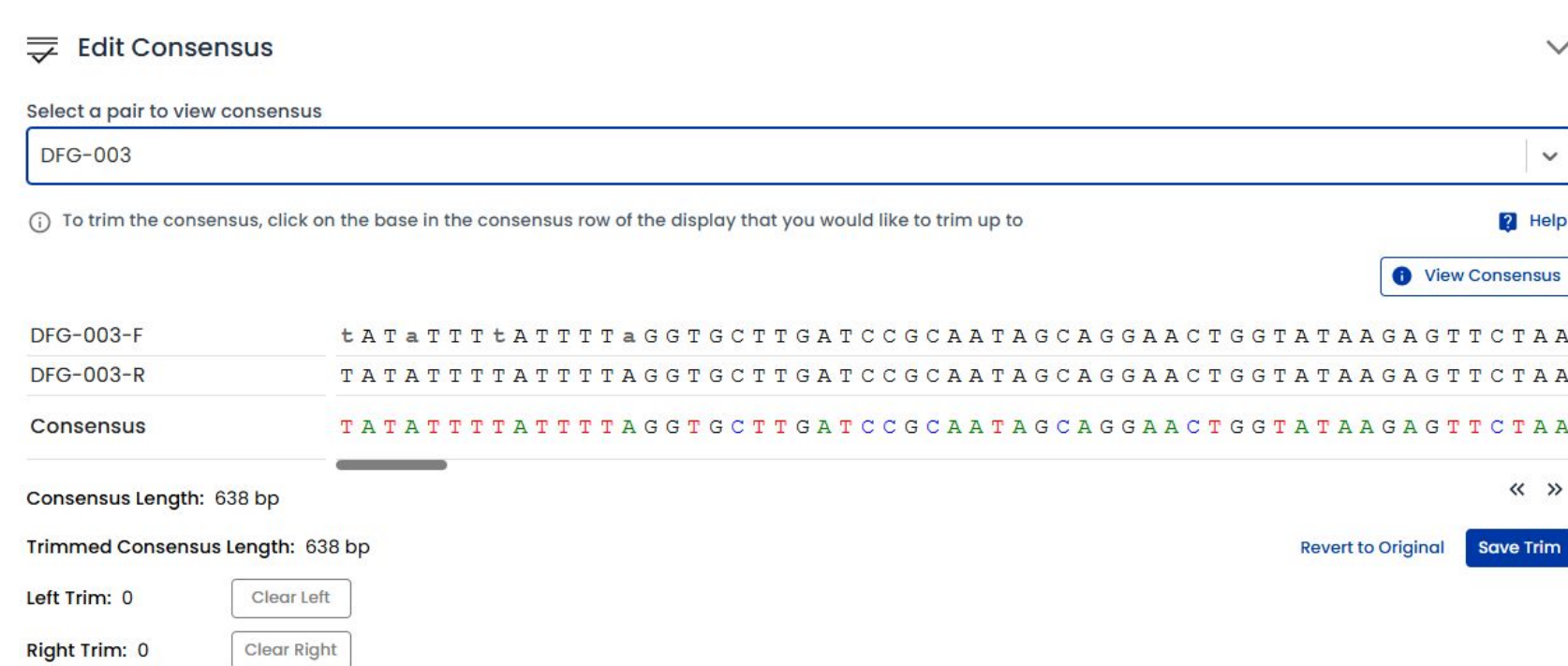
### Experimentation

A small pieces of the butterfly chrysalis was removed and crushed.  
The DNA was extracted  
PCR was used to make copies of the CO1 gene  
Gel electrophoresis identified genes that were copied



### Data Analysis

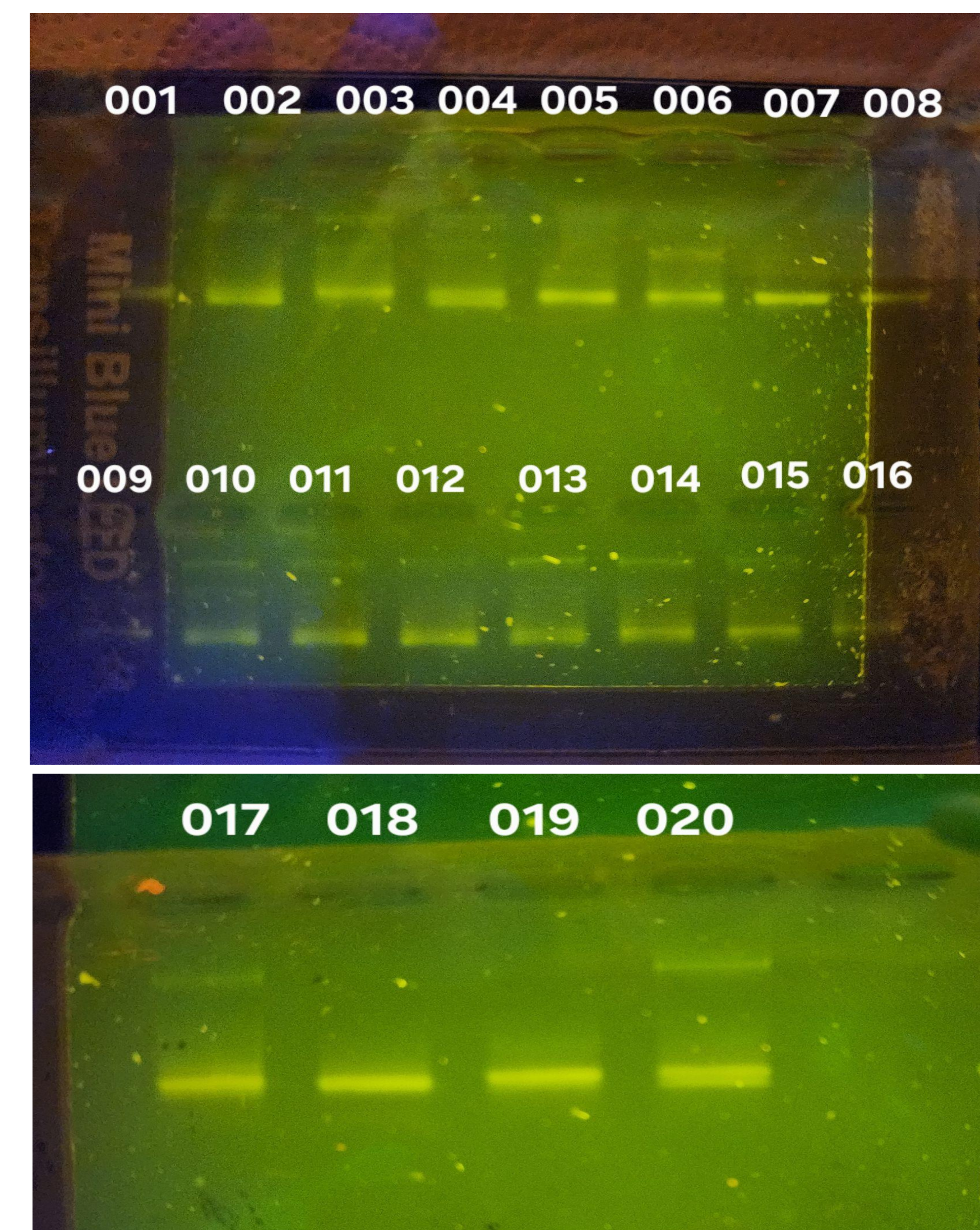
The DNA Subway will be used to determine the sample by telling us its DNA sequence and the BLAST tool will tell us what genes the DNA sequences had.



DNA Subway image created by student researcher

## Results

Gel electrophoresis results from PCR amplification of the butterfly CO1 genes



- The PCR indicates that the genes were successfully amplified
- The DNA samples are currently being sequenced with nanopore sequencing
- Complete sequences will be available within the next week

## Discussion

- PCR samples indicate good amplification of the CO1 gene
- The next steps will be to determine the identification of samples
- These organisms are classified based upon physical characteristics (color, wing shape, hairs, mouth parts, etc.) and DNA analysis will confirm their identify
- Some species may look identical due to convergent evolution and similar adaptations for similar environments but genetically they will have different CO1 sequences.
- If any samples are inconclusive additional DNA extraction and PCR amplification will be needed to positively identify these samples

## Future Directions

Additional sequencing and PCR for any samples that did not amplify. Compare the identities of the samples to the ones provided and determine if there are any major differences.

## References



## Acknowledgements

Thank you to the DNA Learning Center Staff for support with proposals, providing laboratory equipment, and supporting sequence analysis. Special thanks to Jeff Petracca for technical support with sequencing.