

The *COX1* gene reveals that species relatedness corresponds to habitat similarity

Ovalle, M.¹, Lee, A.², Traylor, L.³, Vazquez, A.^{4,5}

¹CVLCC High School, ²Canyon Hills High School, ³San Diego High School, ⁴Boz Life Science Research and Teaching Institute, ⁵UC San Diego, Division of Extended Studies

Summary

- The *COX1* gene was used to investigate genetic similarities between organisms
- Land organisms were placed closer on the phylogenetic tree as opposed to marine organisms

Abstract

The *COX1* gene is a barcoding gene often used to identify and distinguish organisms. To determine if similar environment conditions correlate to genetic similarities, DNA samples of red abalone, medaka, human, zebrafish, fruit fly, and roundworm were isolated, duplicated and sequenced. DNA sequences from the NCBI database were obtained for organisms from various living environments, such as the elephant, dolphin, whale, zebra, polar bear, red panda, and frog. A phylogenetic tree was created using the sequences. We found that organisms in similar living conditions were more closely related on the phylogenetic tree.

Introduction

Hypothesis:

- Organisms that inhabit similar conditions tend to have similar sequences of the *COX1* gene

Species relatedness and their environments:

- Organisms that live closer to each other tend to share similar sequences in the *COX1* gene (Rodrigues, Morelli, Jansen, 2017)
- A species environment plays a factor in the way the organism develops and functions, as it influences which genes are turned on and off (Lee et al., 2011)

COX1 Gene:

- The cytochrome c oxidase subunit 1 mitochondrial (*COX1*) gene encodes a key protein subunit on the electron transport chain
- Highly efficient for discriminating vertebrate and invertebrate species (Rodrigues, Morelli, Jansen, 2017)
- Using the *COX1* gene as an identifier among closely related species has been a success (Rodrigues, Morelli, Jansen, 2017)

Methodology

DNA Extraction:

- Medaka, zebrafish, and red abalone samples were cut to approximately 25 mg tissue chunks
- Roundworm, fruit fly, and human samples were also collected
- Samples were lysed using buffer and heat
- DNeasy Blood & Tissue Kit, a column-based DNA extraction kit was used

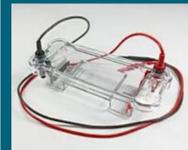
DNA Amplification:

- Used Nanodrop to examine the purity and quantified the yield of extracted DNA
- Performed PCR to amplify isolated sample DNA (Taq Polymerase was used)



Gel Electrophoresis:

- Dissolved agarose in buffer to create 1.5% gel
- Loaded ladder and sample DNA into their respective wells
- Compared DNA samples to ladder under UV light



Sanger Sequencing:

- Allows for the determination of a nucleotide sequence for DNA
- 1st step: PCR with fluorescent, chain-terminating ddNTPs
- 2nd step: size separation by capillary gel electrophoresis
- 3rd step: laser excitation & detection to sequence DNA

Phylogenetic Tree:

- Tissue sequences were combined with black abalone and white abalone sequences from the Boz lab database, and elephant, zebra, red panda, polar bear, frog, whale, and dolphin sequences from the NCBI database
- Generated phylogenetic tree using Clustal Omega



Results

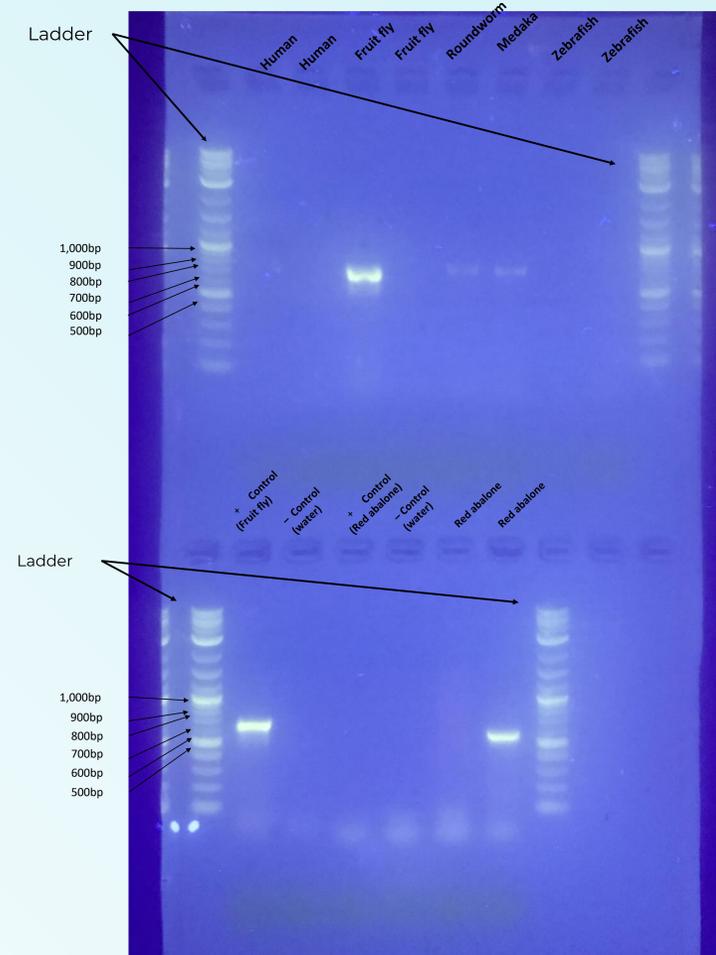


Figure 1. Agarose gel of in-lab samples. Labels on the side indicate ladder sizes. Labels on the top of the wells indicate organism/sample.

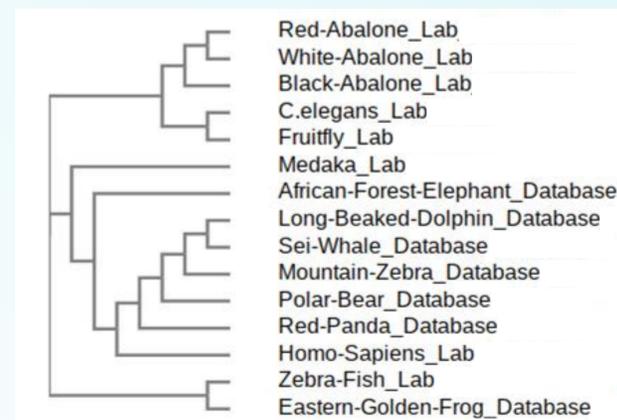


Figure 2. Phylogenetic Tree created using *COX1* sequences of in-lab samples and database samples. Phylogenetic tree including lab and database sequences (indicated at the end of the sequence name). Created in Clustal Omega using neighbor-joining method.

Discussions & Conclusions

Our hypothesis was supported by our data indicating that organisms living in similar environments tended to be similar in their *COX1* gene sequence.

- Both sei whale and long-beaked dolphin had similar sequences (Figure 2), and they both reside in similar parts of the ocean.
- The polar bears, placed close to sei whale and long-beaked dolphin on the phylogenetic tree (Figure 2), live primarily on land but also predate in water.
- High levels of genetic parallelism observed among populations in similar environments (Turner *et al.*, 2018).
- However, being genetically similar in a barcode does not mean that the organism will have a high percentage of genomic DNA similarity (Jorder *et al.*, 2003).
- Our data contributes to the understanding of genetics in relation to their environment.
- Implications for conservation efforts: help determine if organisms could be affected by certain environmental stressors similarly.

Study Limitations:

- Organisms were only categorized into land and water habitat groups, which have variation within them but are not narrowed down to specific regions.
- Some sequences were from databases, which could create noises in the data caused by potential methodology inconsistencies.

Future Directions:

- Habitat environment types should be narrowed further down to a specific region of an ocean or body of water (type of water, temperature, etc.), or land (desert, forest, etc.)
- Most if not all sequences used in the tree should be taken from tissue samples and processed the same way to have more consistent quality control.

Bibliography:

