

DNA Barcoding of Aquatic Invertebrates from Cold Spring Harbor, Long Island

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Abstract

Genetic diversity of aquatic invertebrates has been poorly studied. Despite their ecological importance, many species still remain to be discovered. They play important roles in their ecosystem; for example, as prey for bigger animals, water cleaning, containing the spread of microorganisms, among many ecological functions. The goal of our project was to test the efficiency of four genetic markers for generating DNA Barcodes of aquatic invertebrate collected from Cold Spring Harbor, Long Island, NY. DNA barcoding is a genetic method based on comparisons of DNA sequences to a reference database, in order to identify the species of origin of our samples. The four-targeted genetic markers were: Cytochrome Oxidase I (COI), 12s, 18s, and 28s (ribosomal genes). We evaluated 32 organisms from 10 distinct groups, such as amphipods, crabs, and isopods, among others. We found that 18s yielded 24 DNA sequences out of the 32 specimens, 15 of which were of high quality, followed by 28s, that produced 23 sequences, 10 of which were high quality. 18S produced the largest number of DNA sequences that matched similar sequences in GenBank (26 out of 32, 81%), followed by COI (11 out of 32, 34%). Twenty-two of the 18S sequences were useful for species identification. In addition, we obtained 30 novel sequences not yet reported in GenBank, of which 16 were obtained using the 18s primer set. Overall, our findings showed that the primer set targeting 18s was the most useful for species identification, as well as discovering novel sequences. Our study of aquatic invertebrates helped us to gain a better understanding of the diversity and relevance of these organisms.

Methods

Process of DNA Barcoding

Preparatory Work

-Research Question, Field Work, Sample Collection, and Organism Description

Lab Work

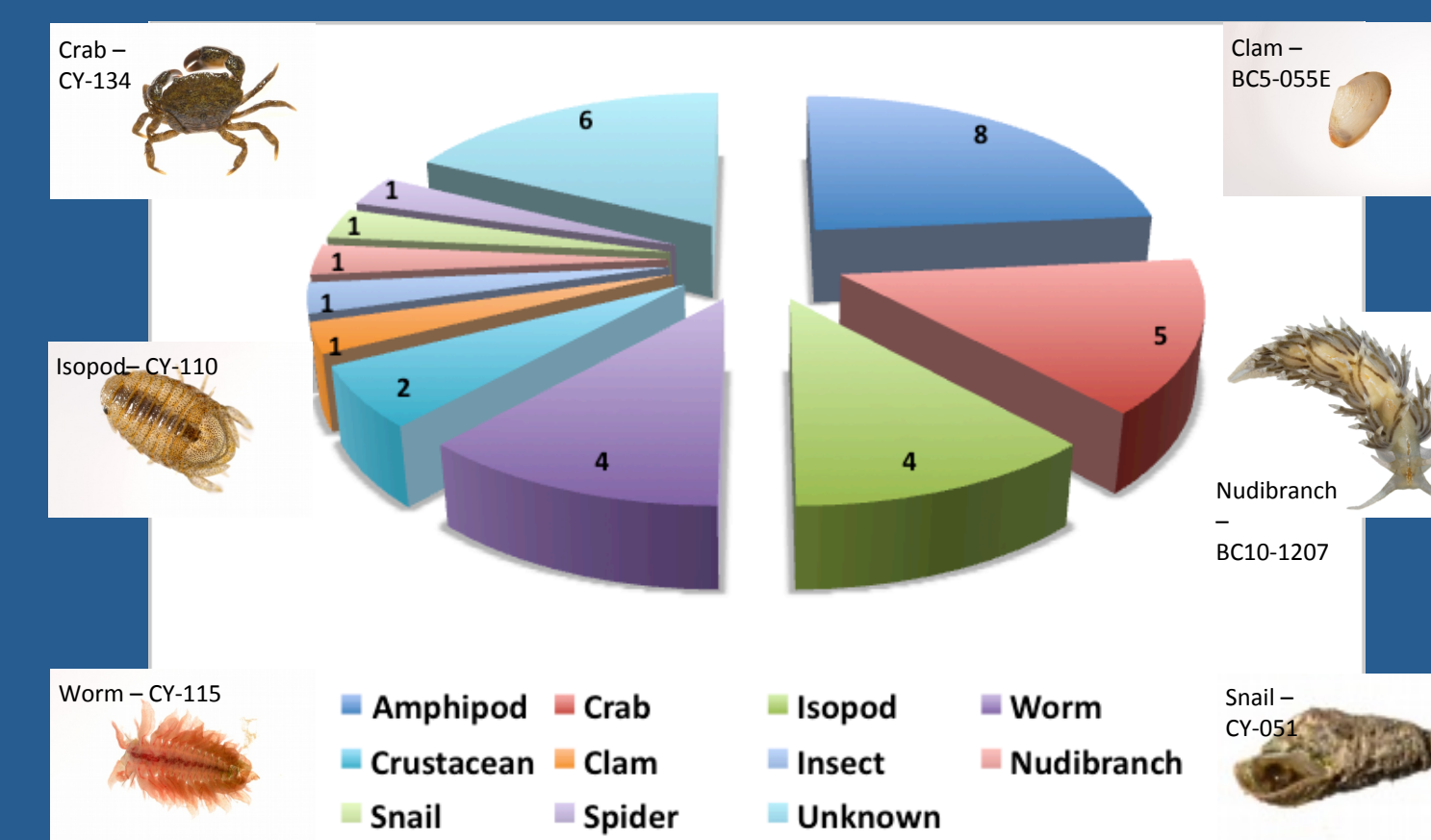
-DNA Extraction, PCR Amplification, Gel Electrophoresis, DNA Sequencing

Phylogenetic Analysis

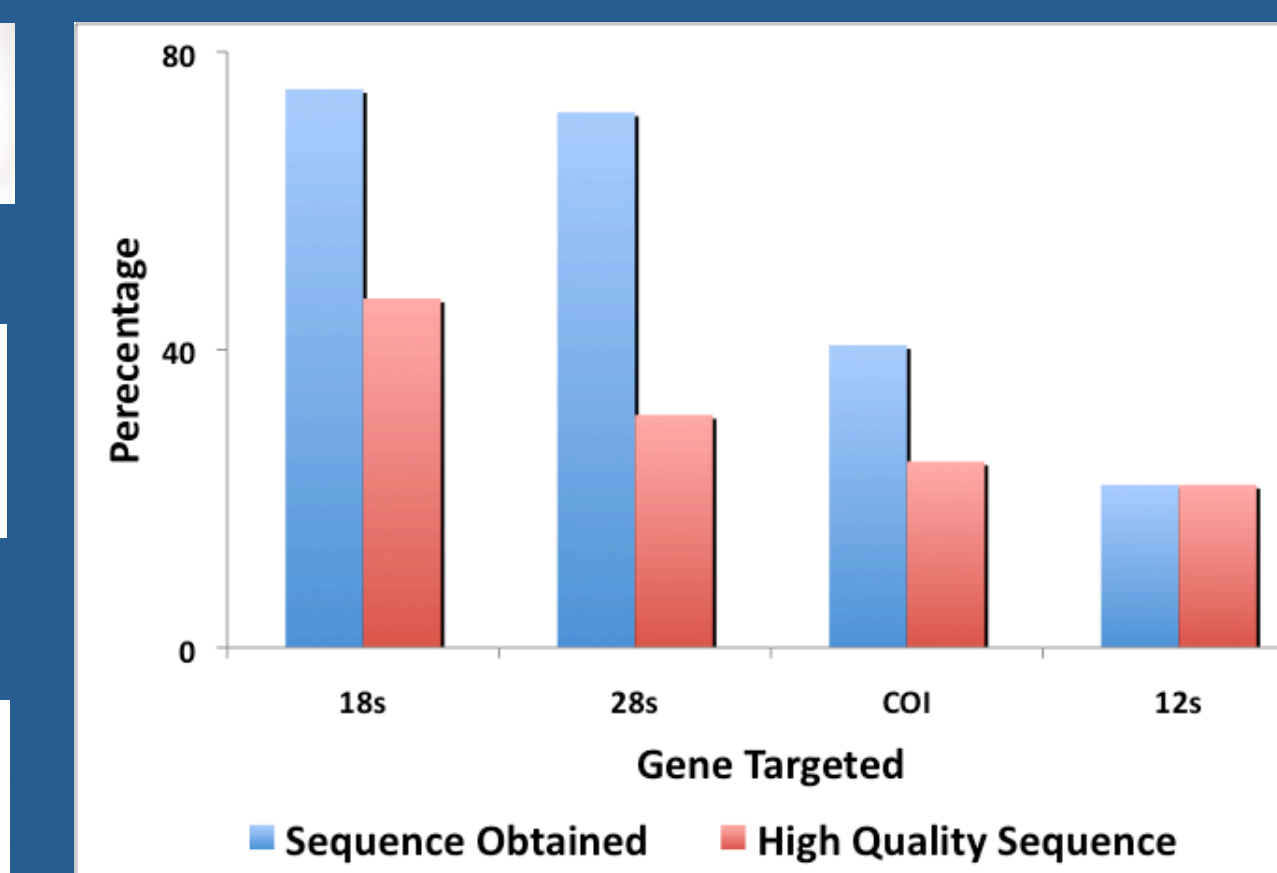
-Sequence Editing, Finding Similar GenBank Sequences (BLAST), Alignment of Sequences(MUSCLE), Evolutionary Tree Building

Results

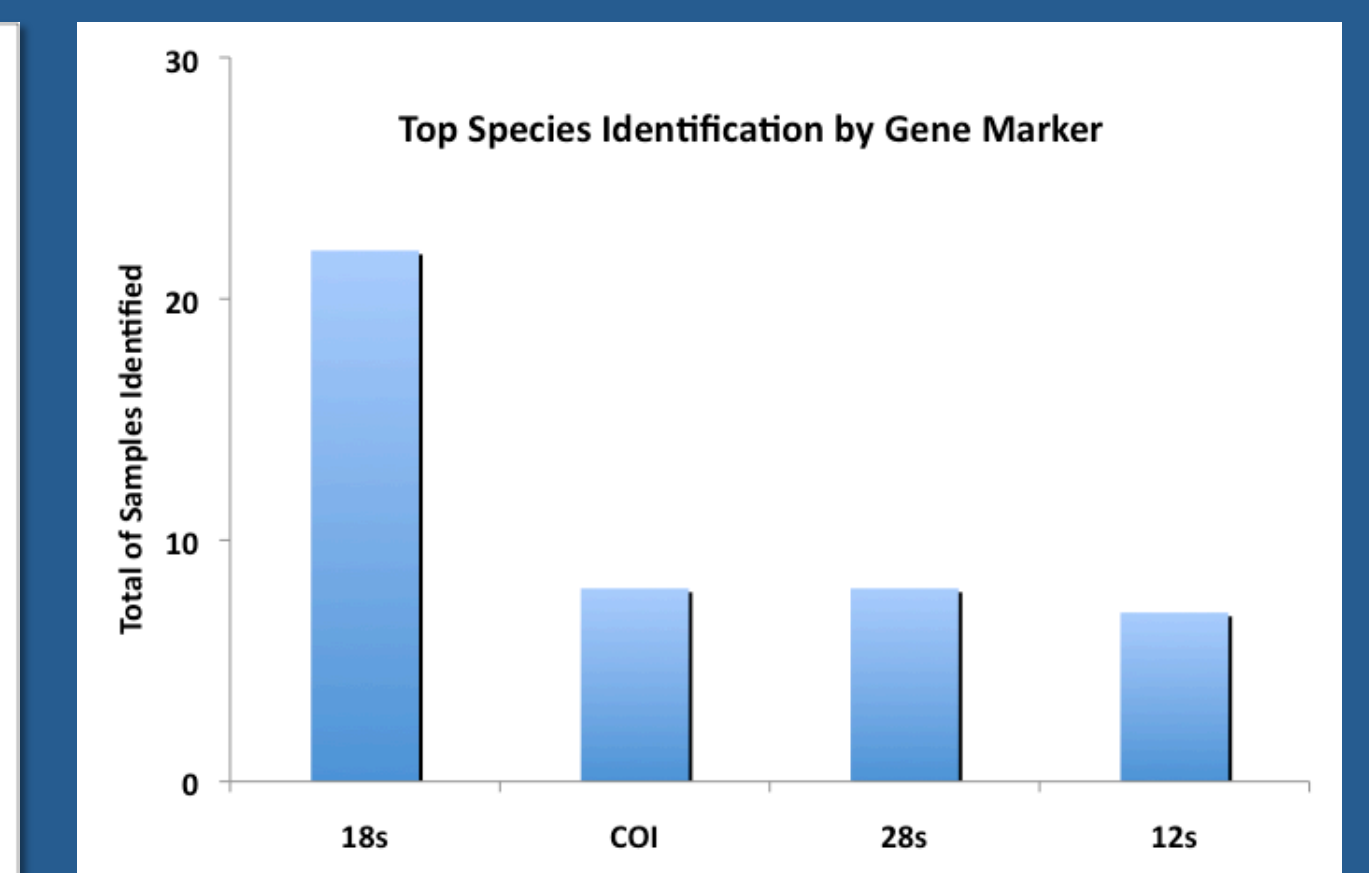
Distribution of Organisms by Type



Comparing Primer Effectiveness

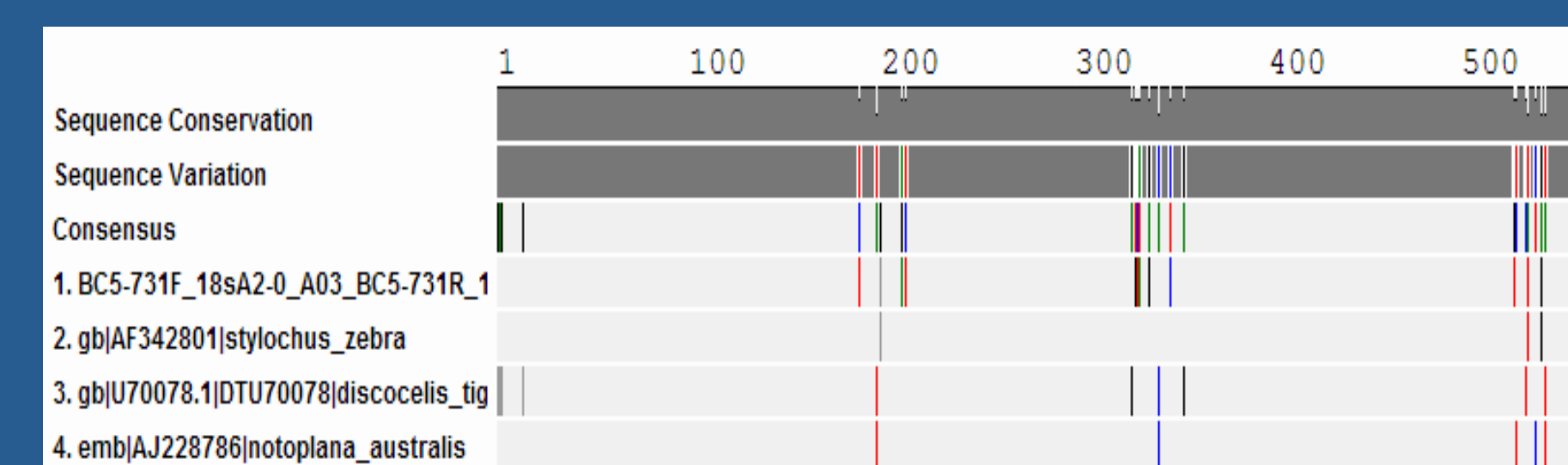


Top Species Identification by Gene Marker

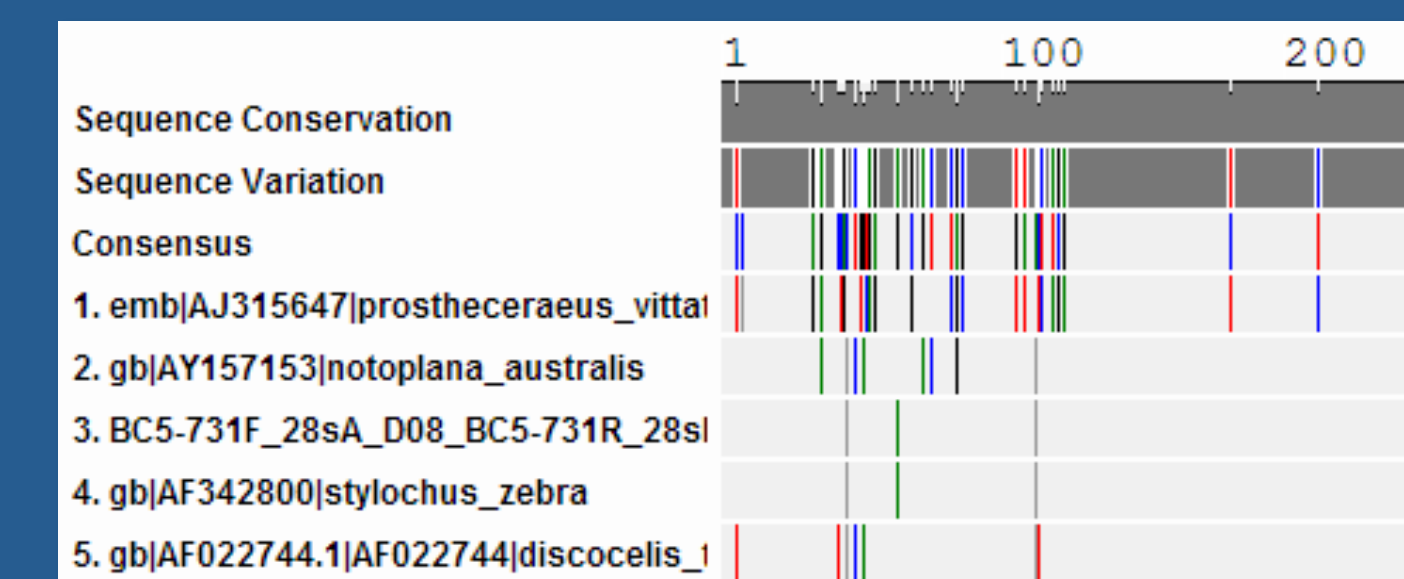


Alignment of Nudibranch DNA Sequences:

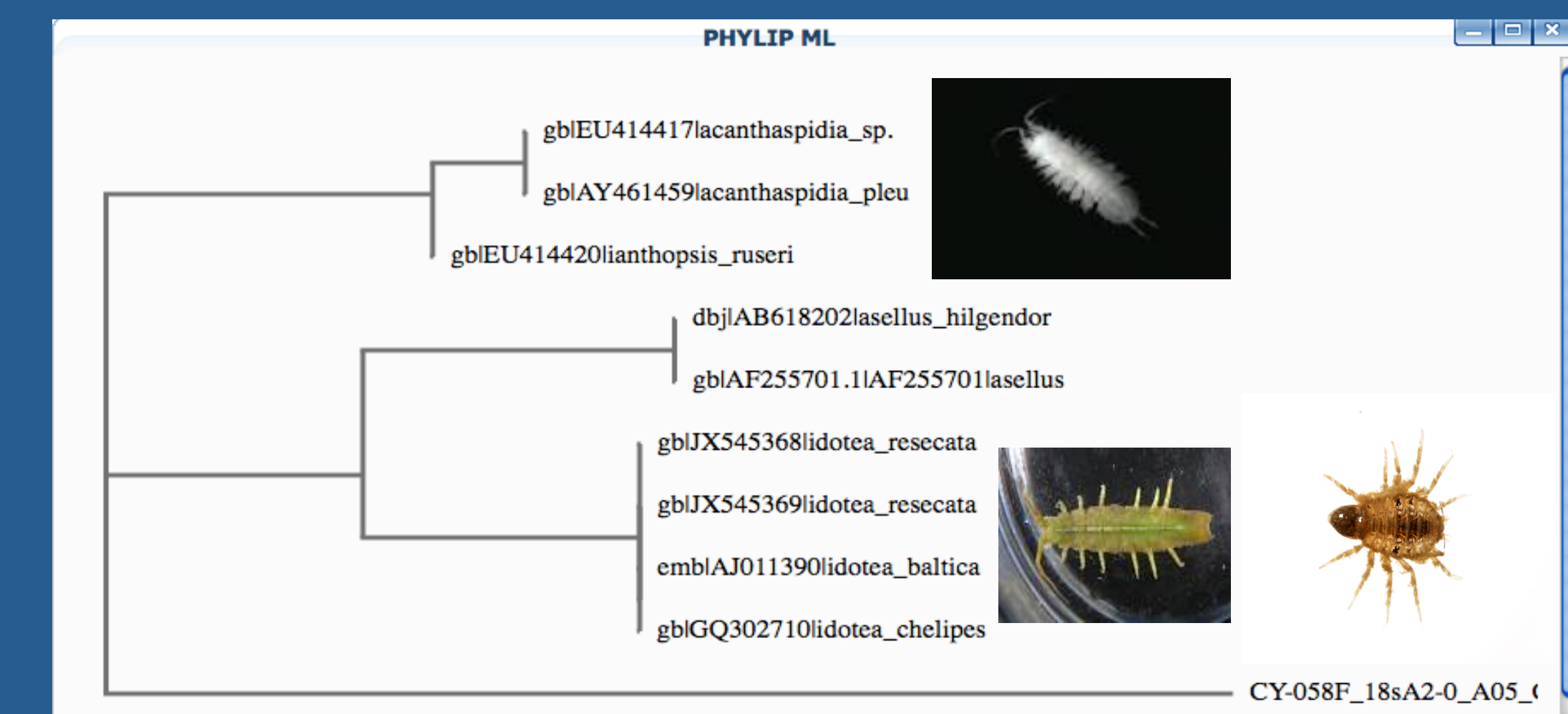
18S



28S



Analysis of a sample distantly related to any other similar sequences.



Conclusions

- 18s is the most efficient primer set; it yielded the most high quality sequences from amplification, while also providing more successful matches with reference sequences from GenBank, for species identification.
- 30 of our sequences weren't found in GenBank. This shows that lots of the genetic biodiversity of aquatic invertebrates is yet to be studied or discovered.
- This genetic diversity shows that some of the organisms that we collected (the 30 novel sequences) could be of undiscovered or undescribed species.
- After studying the genetic biodiversity of aquatic invertebrates, we can gain a better understanding of the roles of aquatic invertebrates in their habitats, as well as their ecology as a whole.

Acknowledgements

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