DNA Barcoding from NYC to Belize

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Traditional means of studying biodiversity depend on expert knowledge from individuals with years of education and training. Recent techniques like DNA barcoding, the process of identifying species based on short fragments of DNA, can be used to quickly identify species and to provide easy access to taxonomic information, a particular benefit to both students and developing nations (1). We use DNA barcoding as the foundation for introducing students to modern biological research. Initially, we set out to develop a research course that serves as an alternative to more traditional laboratories, which often have known outcomes and lack student-generated investigations. Our goal was to provide an experience and skill set to students that would drive interest in the sciences and prepare them for the rigors of studying Science, Technology, Engineering, and Mathematics (STEM). Although we are in the age of genomics, too often the practical knowledge and necessary skills to succeed in science are not taught to students at the high school or even undergraduate level.

The Student DNA Barcoding Project curriculum allows students to pursue research projects by using field ecology and advanced molecular biology methods. Students are exposed to possible careers in STEM fields and the laboratory methods serve as the foundation for future research in academic labs. The course materials outline the infrastructure needed for a teacher to run his or her own DNA barcoding lab at a high school or undergraduate institution, arguably the biggest obstacle in pursuing molecular biology research. Plans for low-budget portable lab space and larger, more advanced labs are included, and all protocols use nontoxic reagents. The course has been field-tested in New York City (NYC) and Belize, and the resulting DNA sequences, made publicly available on The Barcode of Life Data Systems, are a resource for the international scientific community.

The Student DNA Barcoding Project began in 2010 at a public high school in NYC through the National Science Foundation’s (NSF’s) Graduate STEM Fellows in K-12 (kindergarten through high school) Education (GK–12) program conducted by the Center for Advanced Study in Education at the Graduate Center. Within the science research class, we realized that there was an opportunity to integrate laboratory training, mentorship, and student-generated research (see the photos). We decided to seek funding to build a molecular lab space in our school, a place where students and scientists could meet and learn from one another. A companion curriculum was developed to introduce ecological and molecular knowledge and skills with a focus on local and authentic research questions.

The yearlong curriculum has five major units designed to culminate with student-developed research questions; however, it is flexible enough that educators can work with individual units to fit time restraints and specific student populations. Reading scientific literature is embedded throughout the curriculum and students are introduced to the C.R.E.A.T.E. protocol (2), a method for unpacking complex scientific text and generating research questions.

In unit 1, Sampling Local Biodiversity, students practice observation, questioning, and ecological research methods by collecting local samples as a first step in generating a DNA barcode. Emphasis is placed on systematically documenting information about samples. Sampling can occur in local parks as a way to inventory biological diversity or at local markets to investigate potential mislabeling.

Unit 2, Molecular Biology: Theory and Practice, introduces students to the protocols and laboratory skills needed to produce and analyze DNA barcodes. Students learn to efficiently manage small volumes with micropipettes and are introduced to DNA extraction, amplification via the polymerase chain reaction, and gel electrophoresis.

Units 3 and 4, The Science of DNA Barcoding and Analyzing DNA Barcodes, respectively, both build on processing samples from Unit 1. In the lab, students sequence the cytochrome oxidase 1 gene to generate a DNA barcode for their sample and move to the classroom where they use bioinformatics to turn a string of nucleotides into information that can be used to answer biological questions. They are trained to process DNA by checking the overall quality, manually editing and trimming low-quality bases, understanding and using tools like BLAST (3) to identify the species of unknown samples, and aligning sequences across multiple species in order to investigate evolutionary relationships. We take advantage of the user-friendly bioinformatics pipeline implemented in DNA Subway (4) to teach analysis skills and later use the Barcode of Life Data System Student Data Portal (BOLD-SDP) (5) to
create a permanent resource for student-generated barcodes.

In unit 5, Generating DNA Barcoding Research Questions, students develop proposals and produce results that can be presented at local science competitions, or uploaded online to the GenBank Sequence Database or the Barcode of Life Data System. Example research projects include finding the genetic diversity of bed bugs in NYC identifying bioindicator species in polluted parks, and investigating the mislabeling of fish fillets from local fish markets in Belize.

We tested the versatility of the curriculum by implementing different units outside of NYC. In 2012, we partnered with The Petters Research Institute in Dangriga, Belize (www.pribelize.org), where we adapted unit 1, Sampling Local Biodiversity, into a weekend workshop for 24 local students, ages 12 to 16. We introduced concepts like biodiversity, sampling for scientific study, and the value of conserving local ecosystems. We took students into a nearby empty lot and had them collect insects, taught them how to mount and identify the samples to the order level, and showed students how to build a Web site to share information with the public.

Similarly, we adapted units 3 and 4 for an intensive 3-day DNA barcoding workshop at Galen University in San Ignacio, Belize. We worked with undergraduate and master’s students to investigate fish sold in local markets because of a previous report of fish mislabeling in Belize (6) (see the images). Students successfully sequenced 9 out of 12 samples and found that 66% of fish fillets were mislabeled. Pictures, global positioning system (GPS) coordinates, and DNA sequences are publicly available on the BioBelize Web site, and students presented the project and results during a nationwide evening news broadcast. Given our success over the last 2 years, we established The Biodiversity Center of Belize to continue our work (BioBelize, www.biobelize.org).

BioBelize uses a locally relevant curriculum in order to engage Belizean students in STEM fields.

To date, more than 150 students from NYC and Belize have participated in parts of our DNA Barcoding course. U.S. students engaged in the curriculum have competed in multiple science competitions with one student receiving a scholarship from Cornell University to continue her research from unit 5. In Belize, student projects are publicized online, and DNA results are stored in the Barcode of Life Data System Student Data Portal. Our DNA Barcoding curriculum is suitable for both high school science classes and university courses. The course is modular and transportable, and it guides educational facilities in the creation of a functional lab space where student researchers can study local biodiversity.

References and Notes
6. C. E. Cox et al., Conserv. Lett. 6, 132 (2013)

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Supplementary Materials
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Belizean investigation of mislabeled local fish. (A) Size and physical characteristics of purchased fish fillets. (B) Results from gel electrophoresis confirming amplification of COI gene. (C) Trace file of DNA sequence from local fish fillet sample. (D) Red Snapper, confirmed by DNA barcode but mislabeled by vendors.

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