



Project Location

Gulf of Mexico, Northeast, Northwest, Pacific

Project Duration

November 2017 to April 2020

Project Leads

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Project Type

Collaborative research – Generating science that informs decisions

Products

- [eDNA Protocols](#)
- [Recommended eDNA Sampling Plan](#)
- [Estuaries DNA website](#) that includes data visualization tools
- [Newsletter series](#) presenting preliminary results
- eDNA sequence data and archived DNA extractions from water samples
- [Fish species data summary tables](#)
- Journal articles summarizing project findings

Project Partners

- Apalachicola National Estuarine Research Reserve
- Great Bay National Estuarine Research Reserve
- He'eia National Estuarine Research Reserve
- Hubbard Center for Genome Studies, University of New Hampshire
- Hudson River National Estuarine Research Reserve
- New Hampshire Fish and Game Department
- Oregon Department of Fish and Wildlife
- South Slough National Estuarine Research Reserve
- Wells National Estuarine Research Reserve

Project Webpage

nerrsciencecollaborative.org/project/Watts17

New Technology for Old Problems: Developing eDNA Methods to Monitor Invasive Species and Biodiversity in Estuaries

Overview

Biological monitoring programs are essential foundations for effective management of estuaries and coasts, but they can be expensive to conduct and methods such as the capture of living organisms may be traumatic for some target species. Advancements in DNA methods now make it possible to identify the organisms in an area by the DNA they leave behind. Environmental DNA (eDNA) comes from feces, gametes, scales, and cells that an organism sheds, and is easily collected from water and sediment samples. Rapid reductions in analytical costs now allow scientists to analyze eDNA in water samples and identify dozens of species without having to capture live animals or plants; however, when this project began there were few proven methods suitable for highly variable estuarine conditions.

This project established a Community of Practice around eDNA sampling within the National Estuarine Research Reserve system and explored the potential for eDNA to support estuarine management. Scientists and staff from six reserves worked collaboratively with researchers, resource agencies, and a technical advisory team to design and implement eDNA-based monitoring protocols for specific issues and species of interest in estuarine ecosystems. Researchers tested several methods to collect, filter, and extract DNA and developed protocols and how-to guides for practical, time efficient approaches that work in unique estuarine conditions. While the new methods were not suitable for all monitoring objectives, several reserve sites are now integrating eDNA into their monitoring programs to track fish species that can be more efficiently monitored with eDNA.

Project Approach

The project established a Collaborative Learning Community of Practice for the project team, the broader National Estuarine Research Reserve community, and resource agencies associated with each region. Using an iterative approach and input from end users, the project team identified research focus areas, collected samples, interpreted results, and formed recommendations for the use of eDNA at reserve sites. Regular communication through a project website and newsletter shared information and results with a wider community of interest.

In the project's first year, researchers conducted initial pilot sampling at Wells, Great Bay, and South Slough reserves, testing several field sampling and lab analysis methods. The project deployed two methods to analyze DNA in samples: metabarcoding that identifies a wide range of species, and digital droplet Polymerase Chain Reaction (ddPCR) that identifies a single target organism (see Figure 1). Based on the initial pilot, the team created a revised sampling design and protocols that reflect the capacity and needs of reserves. In the second year, fish species were tracked in freshwater streams, tidal streams, and in estuaries at six reserves, and baseline biodiversity assessments were conducted at multiple sites in five reserves. The team developed a series of lab and field protocols that were explained in written summaries and video, and created a set of example datasets illustrating what's possible with eDNA and how the data can be analyzed.

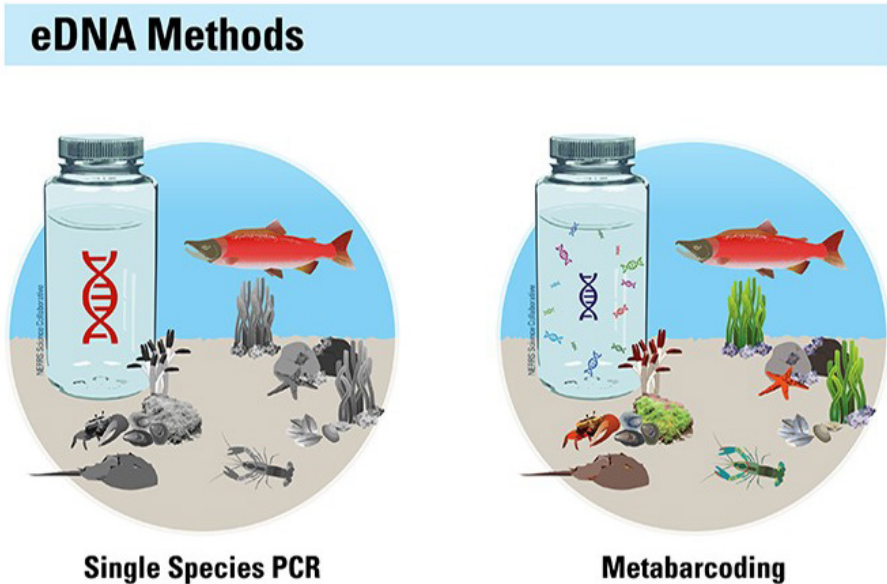


Figure 1. The project deployed two methods depending on the question of interest – (1) single species PCR and (2) metabarcoding for biodiversity assessments and fish species assemblage.

Results

The project found eDNA monitoring to be both exciting and challenging to implement. Challenges identified included vulnerability to contamination, limited DNA databases, challenges with sample analysis, and misinterpretation of results, some of which will be overcome with careful application of consistent method and ongoing expansion of databases. For some applications the technology is appropriate and ready to deploy, yet in other areas its effectiveness is currently limited. For example, pilot studies found that environmental DNA methods are not well suited to the early detection of invasive crabs or the taxonomic identification of copepods. However, monitoring of fish communities and biodiversity are two promising areas for the incorporation of eDNA methods. A few highlights of results related to these focus areas are discussed here.

Optimizing methods for estuaries

The turbidity, complex flow paths, and varying temperatures and salinity make it challenging to design eDNA sampling in estuaries. This project developed a suite of [lab and field protocols](#) that have been tested in estuaries across the country, including recommendations for how much water should be collected and how it should be filtered and how to design an appropriate sampling plan.

Comparing eDNA with seining to identify fish species

Great Bay and South Slough reserves conducted paired sampling using both eDNA and traditional seining methods to identify fish species. Results from the comparison showed that eDNA identified some but not all of the species found using seine nets when collecting 6 1-liter samples at each location. A larger sampling network increased the number of fish species found in eDNA so that most expected fish species were identified.

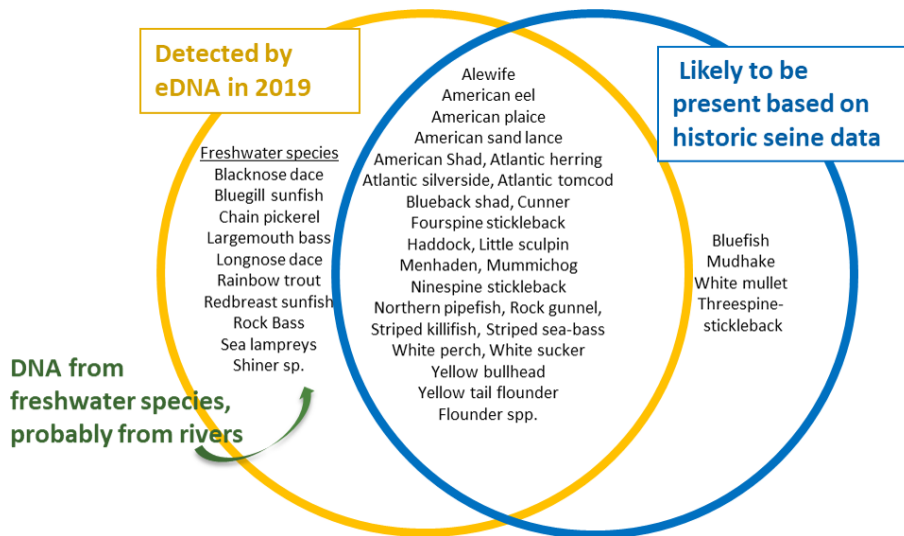


Figure 2. Fish species detected by eDNA compared to species expected to be present based on seine surveys in Great Bay, NH. Water samples were collected at three locations over the course of 2019. Most of the species expected to be present were detected in eDNA samples, but multiple freshwater species were also detected, indicating transport of DNA into the estuary from tributary streams.

Comparing metabarcoding to microscope identification for plankton

Traditional methods to identify larval fish and plankton involves using a microscope to examine specimens in a tow sample, which can be time consuming and error prone. The project compared these methods with eDNA metabarcoding at Wells and Apalachicola. For larval fish, eDNA methods detected more species than microscope identification methods, but some of this DNA is likely from adult fish. Detecting copepods using eDNA showed potential but DNA databases will need to expand to allow a wider range of plankton species to be identified.

Mapping anadromous fish in streams

The project explored metabarcoding and single species eDNA methods to locate specific fish species in streams, including river herring, American eel, and lamprey. In general, both eDNA methods were found to be successful, although the single species method is easier to interpret and less subject to error.

Biodiversity

Environmental DNA monitoring can be used to develop baseline biodiversity metrics by analyzing existing reserve monitoring samples. The project used eDNA methods to analyze biodiversity at five reserves, identifying differences between estuaries and habitats and tracking seasonal and long-term changes.

Detecting invasive green crab

One original goal of the project was to develop an eDNA method for the early detection of invasive green crab. However, monitoring results showed that eDNA is not well suited to this goal because hardshell and softshell crabs shed very little DNA (with the exception of crabs carrying eggs). Other invasive species may be more readily identified by eDNA, but a large number of samples would likely be required to develop a reliable detection program.

Benefits

- Reserves have the tools and information they need to begin to incorporate eDNA into their monitoring programs. The project's research findings and its associated products answer key questions for reserves about potential applications of eDNA for estuarine monitoring, appropriate sampling approaches, and the reliability of results. The project's protocols have been adopted by several state agencies, including New Hampshire Department of Environmental Services, and the Maine Department of Environmental Protection.
- There is limited eDNA monitoring data in estuarine systems and project results are of significant interest to other agencies considering the use of eDNA. For example, team members have been invited to serve on advisory committees and present to a workshop about managing endangered species in San Francisco Bay.
- Some reserves, including Great Bay, South Slough, and He'eia, have adopted eDNA monitoring methods for targeted needs, enhancing the reserve system's ability to characterize biodiversity and providing an additional tool to explore locally relevant management questions.
- This project strengthened collaborations among academic researchers, reserve scientists, and natural resource managers. The project brought together a large team from six states and shared data, lessons, and experiences with a wide range of coastal management stakeholders, and the group continues to pursue opportunities and offer each other technical assistance.

What's Next

There is significant interest in incorporating eDNA into monitoring programs within the National Estuarine Research Reserve system. Several reserves that participated in this project have elected to continue to use eDNA monitoring for targeted needs. For example, South Slough has launched eDNA monitoring for native lamprey using citizen volunteers, Great Bay will continue to collect samples at long term monitoring sites, and He'eia is continuing to collect and store filtered water samples hoping to identify funding for analysis later.

About the Science Collaborative

The National Estuarine Research Reserve System's Science Collaborative supports collaborative research that addresses coastal management problems important to the reserves. The Science Collaborative is managed by the University of Michigan's Water Center through a cooperative agreement with the National Oceanic and Atmospheric Administration (NOAA). Funding for the research reserves and this program comes from NOAA. Learn more at nerrsciencecollaborative.org or coast.noaa.gov/nerrs.