Executive Summary

eDNA works. Get going.

Introduction and setting

Scientists as well as children and fishers of the sea have long dreamed of knowing exactly what species swim the oceans without the need to capture them or even observe them directly. About 20 years ago researchers glimpsed the realization of that dream when they realized that animals shed DNA in aquatic environments, and that studies of these fragments, known as environmental DNA or eDNA, might provide the evidence for accurate, timely identification of marine life. That dream has matured enough to commit to comprehensive marine eDNA collection and analysis. Experts convened in New York recently to share ideas and test their resolve to move this technique forward.

Approximately 100 American ocean scientists and associated stakeholders with experience, skill and/or interest in marine eDNA assembled at The Rockefeller University in New York City for a conference sponsored by the Monmouth University-Rockefeller University (MURU) Marine Science and Policy Initiative on 29-30 November 2018.

The Conference included participants from academe, federal, state, and local governments, non-governmental organizations concerned with environment, and the private sector. Seven leaders in the field made formal plenary presentations to share the state-of-the-art. The conference website offered a Marine eDNA Primer, a sponsored paper The Ocean as a Living Sensor: Environmental DNA and Acoustics for Detecting Marine Life by Drs. Alison Watts and Jennifer Miksis-Olds (University of New Hampshire), a Press Release by Terry Collins summarizing recent developments, and the Conference Program laid out the agenda and speaker biographies. Jesse Ausubel, Director of the Program for the Human Environment (PHE) at The Rockefeller University opened the conference with a short statement of purpose. Mark Stoeckle, leader of PHE’s NYC/NJ Aquatic Vertebrate Environmental DNA (eDNA) Project, moderated the plenary sessions. The participants broke twice into seven groups for two hours to address four designated topics (basic biological frontiers of eDNA, bioinformatics needs and opportunities, technology development, and priorities for US applications). Vice Admiral Paul Gaffney, USN (Ret.), Fellow at Monmouth’s Urban Coast Institute (UCI), closed the conference by summarizing key points made by the speakers and break-out groups.

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To cap the conference, Monmouth University presented its “Champion of the Ocean Award” to Dr. Marcia McNutt, president of the National Academy of Sciences. Dr. McNutt, an early advocate for marine instrumentation development during her tenure as director of the Monterey Bay Aquarium Research Institute, offered a closing charge to go forward. An article by Steve Leahy for National Geographic, “New DNA tool ‘changes everything in marine science’” offers a good popular summary of the developments.

**eDNA localization in space: bay fish eDNA in bay, ocean fish eDNA in ocean**

- 12-mo shoreline sampling Barnegat Light, NJ

**Top Ten Fish eDNAs**

*Bay*

1. Mummichog
2. Atlantic silverside
3. Striped killifish
4. Four-spined stickleback
5. Sheepshead minnow
6. Killifish species
7. Atlantic menhaden
8. Naked goby
9. Northern pipefish
10. Oyster toadfish

*Ocean*

1. Atlantic menhaden
2. Bluefish
3. Striped bass
4. Bay anchovy
5. Tautog
6. Sand lance
7. Windowpane flounder
8. Drum species
9. Silver perch
10. Northern kingfish

- Top 10 frequently detected fish eDNAs
- 26 ocean/bay samples
- Apr 2017-March 2018

It works in space: Marine eDNA sampled in adjacent ocean and bay habitats shows that the DNA stays close to where the species are. Source: M.Y. Stoeckle, 2018.
Conference findings and recommendations

1. Begin to operationalize marine eDNA observations.
   - Focus first on key basins in the US exclusive economic zone (EEZ). Priority examples include the Gulfs of Mexico and Maine, Monterey Bay, California Current (CalCOFI) area, New York Bight, and the Bering Sea/Arctic.
   - Systematically address unresolved science questions, observation rules and standard operating procedures, reference standards and archival capabilities, but start now to establish eDNA observational baselines at many sites.
   - Start early on Long Time Series (LTS) observations, because they will afford many crucial insights, e.g., about effects of human activities.

Marine eDNA science and its observational and analytical techniques are mature enough to start now with significant federal government investment of tens of millions of dollars per year for several years. Despite needs to determine observational geometry, standardize filter specifications, prioritize species of interest, establish standards, and expand and in some cases certify reference libraries, we believe the techniques are mature enough and the cost low enough.
(large oceanographic ship time is normally an expense that can be avoided) to justify starting now. Our sense of the conference is to get started on long time series eDNA collections in strategic US EEZ basins; such regions offer key data from existing survey techniques that will allow fast calibration of eDNA measurements. In less known and less explored regions, eDNA data itself may become the baseline reference information.

2. **Involve industry and investors.**
   - Shorten the timespan from sample collection to sample identification.
   - Explore decision-making value of marine eDNA, and seize commercial opportunities.

Currently it takes days-to-weeks to identify species from water samples for marine eDNA collections. The process entails multiple processing steps and often remotely located labs and sometimes commercially contracted labs. Technology development should compress the steps and lift speed. Industry has an enviable record of shrinking such technology production processes in space and time to reduce cost and satisfy customer demand for quicker feedback and enable wider use. Recognizing there is a brewing customer base that investors may wish to capture, industry should be encouraged to enter the marine eDNA enterprise and focus on (a) shortening collection-to-identification time and (b) aggregating data to allow discovery of larger patterns than envisioned by those engaged in the original sampling. Fast identification of eDNA could allow adjustment of missions in progress and adaptive sampling, a big potential benefit. The conferees noted a current backlog in eDNA species identification, another opportunity to speed analyses.

Marine eDNA information can be valuable to those who make operational decisions in the ocean (e.g., about vessel routing or seismic testing) and marine regulators (e.g., about closing of areas to commercial or recreational fishing) as well as to scientists. Commercial customers for eDNA information are just over the horizon.

3. **Speed a National Ocean Partnership Program (NOPP) for marine eDNA** with multi-year, multi-investor, multi-participant projects.
   - NOPP leaders should consider streamlining and speeding up the NOPP process to attract and retain industry and philanthropies as NOPP investment partners.

NOPP, a concept introduced into law in 1998 and promoted in the June 2018 [White House Ocean Policy Executive Order](https://www.whitehouse.gov/administration/presidential-actions/white-house-ocean-policy-executive-order/), is the right mechanism to bring together those interested in advancing eDNA collections and science. NOPP investors to consider: Navy, NOAA (NMFS, NOS, OAR, NMSP), NSF, NASA, DOI (BOEM, USGS, FWS, NPS), USACE, EPA, NIEHS/NIH, commercial investors, and large philanthropies. A coordinated national initiative can bring early benefits, both environmental and economic, to the US and position this nation for sustained leadership in what will become a global technology.

Rarely has NOPP investment partnership included commercial investors. Yet in marine eDNA there are new commercial opportunities. Commercial investors and industry should be incentivized to join NOPP generally, but NOPP projects on marine eDNA specifically. That will
mean speeding up the process of bringing together government, philanthropic, and commercial sponsors, and then quickly choosing performers to carry out NOPP eDNA projects.

4. Begin to think about the impact on existing statutes, regulations, and permitting/licensing processes as marine eDNA is introduced as a credible ecosystem census indicator.

Laws have been carefully written, debated, and rewritten to cover many ocean activities. Examples include NEPA, Magnuson-Stevens Fishery Conservation and Management Act, Marine Mammal Protection Act, Endangered Species Act, and others. In carrying out the marine policy laws and their related regulatory processes, affected parties gather evidence to make their cases: species sightings, marine census trawls, models, etc. Because marine eDNA collection and analyses will become increasingly available to regulators and courts, we advise starting now to learn how marine and ocean policy laws may need to be updated to account for marine eDNA information. Corollaries are establishment of marine DNA standards and certification of reference assets.

The way forward

Our “sense of the conference” is that marine eDNA is already a surprisingly reliable, feasible, and affordable ocean observation technology ready for rapid adoption and poised for giant steps forward. In short, it works. Starting to create baselines for eDNA time series commitment should not wait for perfection. Marine eDNA collection and identification procedures can and should be continually refined as baseline observations proceed. Standard protocols for collection (e.g., depth options, horizontal distance between observations, frequency of repeat observations, filter selection) will develop into a set of best practices. In all such work it is vital to ensure recording of metadata. This is especially important as the marine eDNA community refines its collection standards. A corollary is to compare other data collected at about the same time in the same area (trawl results, acoustic measurements, and visual sightings) with marine eDNA results and to explore how these compliment and supplement one another.

We believe that comprehensive marine eDNA collection commitments in a few key areas will provide insights into critical science questions, including eDNA degradation rates, transport speed for various particle sizes and shapes, thresholds of significance for detection, and stable or undisruptive handling of cellular and extracellular eDNA.

Laboratory and field replication studies—e.g., repeating PCRs on a single sample, and analyzing replicate samples collected over fine-scale in time and space will improve sampling strategies. Reference libraries or centers are obviously important in species identification. These are also targets for marine eDNA investment because they will need to grow as new organisms are discovered. Relevant sequences from newly collected specimens should be readily accessible to the marine eDNA community in a timely manner. Along with reference library robustness, one must consider the task of archiving physical samples; how and where. The potential impact of marine eDNA on regulation and law makes reference and archival collections increasingly important.
Notably, initial collection is relatively simple and often does not require a dedicated oceanographic research, exploration, or survey vessel. This reality reduces the cost of collecting, and can offer opportunities for citizen scientists, affordable repeat collections, and a broader field or cube of collection. Further, cruises and sampling planned for other purposes (e.g., bathymetric survey, geology and geophysics research, physical oceanography measurements) can include an eDNA dimension at negligible cost in time and money; collecting and filtering a liter of water will often suffice.

Initial collection and filtration of an eDNA-laden water sample may be relatively simple, but processing the collected sample into useful information to inform the observational plan, soothe customers, and affect decision making requires that current “eDNA sample-to-species identification” processes become speedier and cheaper. Conference discussions highlighted the goal: “Prioritize a push for automation aboard an autonomous platform.” Industries with potential commercial customers will logically drive to increase speed and decrease analytic costs while maintaining credibility through better documentation on chain of custody, replicability of experience, transparency and reproducibility of analysis, and other matters. Hence, we suggest active involvement of industry (hardware and information technology, genomic analysis and systems production companies and investors) with government sponsors and academic performers. The community should exploit capabilities for better visualization of data so that this new information can be used by the widest array of government, student, public, and commercial customers. The NOPP seems a ready opportunity if NOPP leaders recognize private investor timelines and modify the NOPP process accordingly.

As the national oceanographic community accelerates use of marine eDNA, stakeholders must determine priorities for collection and analysis, sector by sector and collectively. For ecological studies, scientists may want to study all that they can see in an area and therefore analyze every bit of eDNA collected. An information company, depending on its customer, may want comprehensive species analyses in area and time, or it may have customers interested in identifying a key species in nearly real time. Government agencies that are regulating or otherwise making policy or operational decisions may want to limit their collection and analysis activities to certain depths that include the most important species and, of course, their analysis may narrow the field to identify the presence of only certain species. Those employing marine eDNA as a tool to detect ecological hazards including harmful algal blooms and invasive species may also have narrow priorities.

Processes such as creating a NOPP call for proposals and forums by groups such as the National Academies’ Ocean Studies Board, the Consortium for Ocean Leadership, and professional societies to help the American eDNA community set priorities and foster a leading position for

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4 Our report mentions three ways in which eDNA science may aspire to be robust: reproducibility, replicability, and generalizability, following the National Science Foundation’s 2015 report Social, Behavioral and Economic Sciences Perspectives on Robust and Reliable Science. An experimental finding is reproducible according to this framework if a researcher is able to duplicate the results of a prior study using the same methods, procedures, code, and data as the original author of the research. An experimental finding is replicable if a researcher is able to duplicate the results of a prior study by applying the same procedures and methods of the original experiment to newly collected data. Finally, an experimental finding is generalizable if a researcher is able to duplicate the results of a prior study using an entirely different experimental design and associated data.
the US in the development and use of what we believe is truly a revolutionary technology for ocean observing. Conference discussions made clear that the marine eDNA community is excited about this new tool that is already finding increasing application and calls for scientific groups to meet regularly to share lessons, propose new ideas, and train new participants in a national marine eDNA collection program. The attendees welcomed an offer of the Southern California Coastal Water Research Project to host another national marine eDNA conference in 2020.

Marine eDNA is coming up to full running speed. It excites the marine science community and the public and will attract the attention of operational decision-makers and regulators. For the marine eDNA community and stakeholders the message is clear: Get going.
Acknowledgment
The MURU Initiative, philanthropically enabled by Drs. Robert and Joan Rechnitz, has earlier convened leaders in ocean exploration, ocean acoustics, US international ocean policy coordination, and the Blue Economy in the Mid-Atlantic region. The MURU team gratefully acknowledges the Rechnitz family for making possible influential research, analysis, and networking on these topics. For eDNA, the Schmidt Ocean Institute, Ocean X (an initiative of Dalio Philanthropies), and the Hudson River Foundation joined as most welcome sponsors. This report draws on insights of the 100 Conference attendees. We thank Monmouth University's Tony MacDonald and Karl Vilacoba for contributions throughout the conference process and editor Dale Langford for help in preparation of this report.
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