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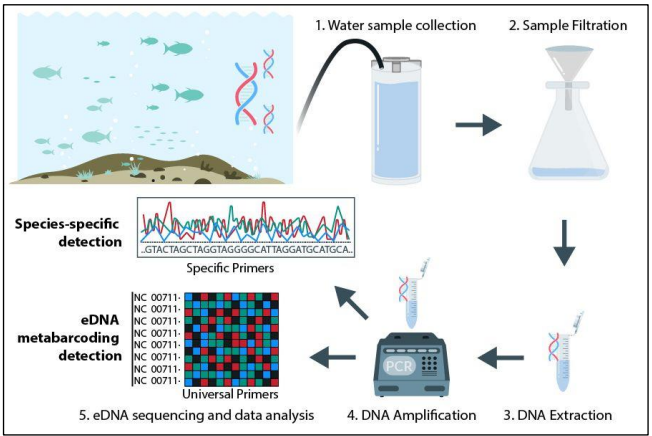
eDNA/eRNA: Scientific Value in What's Left Behind

Environmental deoxyribonucleic acid (eDNA) and environmental ribonucleic acid (eRNA) are trace amounts of genetic material collected from an environmental sample, such as from soil, terrestrial and aquatic sediments, freshwater, seawater, wastewater, or air. This genetic material can come from shed skin, hair, and other cells; secreted waste or mucus; and carcasses, among other biological sources. It can provide information on the recent presence of organisms where a sample was collected without directly observing the organisms. Multiple federal agencies have begun using eDNA/eRNA for various research, monitoring, and decisionmaking purposes. Decreased costs and increased access to eDNA/eRNA analysis technologies have also allowed other entities to develop monitoring projects and programs.

How It Works

To analyze eDNA/eRNA, researchers extract partial DNA/RNA from an environmental sample, amplify it (make copies), and compare it against a reference sequence from a previously sequenced organism in order to make a positive match. Depending on research needs, different types of eDNA/eRNA analyses can be performed on an environmental sample (see **Figure 1**). For example, a targeted analysis aims to detect a particular species with high accuracy. Alternatively, a non-targeted approach, a method referred to as "metabarcoding," can more broadly characterize the various different species whose DNA/RNA is found in the sample.

Figure 1. eDNA Sampling in Aquatic Environments



Source: Songqian Huang, Kazutoshi Yoshitake, and Shugo Watabe, et al., "Environmental DNA Study on Aquatic Ecosystem Monitoring and Management: Recent Advances and Prospects," *Journal of Environmental Management*, vol. 323, no. 116310 (2022).

eDNA/eRNA Reference Databases

The availability and quality of a reference sequence can affect the analysis of and ability for an eDNA/eRNA sample to be used for various research and decisionmaking purposes. For example, accurate identification of a particular species requires reference sequences of sufficient quality to exist and be available. Individual databases have varying standards that govern access as well as the type and quality of information they contain. Therefore, the design, management, quality standards, and data protocols for reference databases can affect eDNA/eRNA's utility. Databases containing certain genetic sequence information, such as for viruses, can also have implications for biosafety and biosecurity, depending on who has access to sequences and if their use is regulated.

Some federal agencies are developing, or already have, databases for eDNA/eRNA analysis. For example, the U.S. Geological Survey (USGS) has, and plans to develop, databases for eDNA that include assays and genetic markers. The Aquatic eDNAtlas Project, managed by the U.S. Forest Service National Genomics Center for Fish and Wildlife Conservation, is a database that provides information on eDNA sampling detection/nondetection results for freshwater species in the United States. The database, updated annually, contains over 20,000 data points. Natural resource agencies and nongovernmental organizations use a standardized field sampling protocol to gather and enter environmental samples for the database.

Nonfederal entities—such as universities, private companies, and international organizations—also compile their own databases of species sequences. For example, the International Barcode of Life (iBOL) is an international research consortium developing DNA reference libraries, sequencing facilities, informatics platforms, and analytical protocols to help inventory and assess biodiversity. iBOL has entered sequences from 500,000 species into reference libraries and intends to add another 2.5 million species by 2026.

eDNA for Environmental Monitoring

Federal agencies use eDNA for various monitoring purposes. eDNA data can provide information on the presence or absence, geographic distribution and range, relative abundance, and status of organisms. Applications include tracking species of interest, such as particular native species, tracking protected species for species assessment and conservation purposes; identifying the presence of small or rare (lesser known) species that are difficult to observe directly; and detecting the presence of non-native species that may adversely affect ecosystems.

eDNA analysis can be limited in its ability to determine how many of a particular species may be in the study area. eDNA data also cannot determine with certainty whether a particular species was located in the sampling area at the

time the sample was collected or whether the eDNA collected was transported to that area via other mechanisms (e.g., a species' DNA present in vessel ballast water). Additionally, eDNA degrades at varying rates depending, in part, on microbial activity and environmental conditions, which can affect the utility and accuracy of eDNA.

eRNA for Disease Surveillance

Public Health authorities and researchers increasingly use wastewater surveillance—in the form of eRNA—to identify and track disease prevalence. People infected with a virus shed viral RNA in their feces. Wastewater samples can be collected and analyzed for the presence of eRNA from a virus of interest, if that particular virus has been sequenced previously. Similar methods are used to identify non-viral disease (e.g., stool screening for certain types of cancer).

In response to the COVID-19 pandemic, the Centers for Disease Control and Prevention launched the National Wastewater Surveillance System in September 2020 to coordinate and build U.S. capacity to track the presence of SARS-CoV-2, the virus that causes COVID-19. In September 2021, the Environmental Protection Agency released *A Compendium of U.S. Wastewater Surveillance to Support COVID-19 Public Health Response*. The report describes different COVID-19 wastewater surveillance programs, as well as case studies to highlight best practices and lessons learned from programs across the United States. The report also discusses federal and nonfederal funding opportunities that aid in developing and implementing wastewater surveillance programs for SARS-CoV-2.

Selected U.S. Federal eDNA Initiatives

Federal agencies are engaged in multiple projects and programs that develop and use eDNA. Many of these initiatives involve collaborations with international, state, tribal, and local authorities.

One of the longest-standing, best-known applications of eDNA has been for operational surveillance of invasive fish species in the Great Lakes region. The Great Lakes Restoration Initiative, made up of nine federal departments and agencies, seeks to continually test and refine the collection, processing, analysis, and interpretation of eDNA samples and results.

The USGS, in collaboration with the Monterey Bay Aquarium Research Institute, is developing the Rapid eDNA Assessment and Deployment Initiative and Network (READI-Net). Part of the program's goal is designing and constructing autonomous robotic eDNA samplers that can be deployed as a network in aquatic environments across the United States. USGS intends for READI-Net to enhance early detection and rapid-response methods to aid resource managers in containing and controlling aquatic biological threats.

The U.S. Forest Service's National Genomics Center for Wildlife and Fish Conservation provides eDNA services to researchers within and outside the Service, including sample analysis and assay development, along with development of protocols and training in eDNA research.

The U.S. Government eDNA Working Group is an informal interagency working group comprised of federal employees working with eDNA. The group focuses on the exchange of technical information, including sharing best practices, lessons learned, and new advances.

The Marine Biodiversity Observation Network (MBON) is a global initiative composed of regional networks of scientists, resource managers, and end-users working to integrate data from existing long-term programs to understand changes and connections between marine biodiversity and ecosystem functions. The U.S. node of MBON is a partnership among the National Oceanic and Atmospheric Administration, the National Aeronautics and Space Administration, the Bureau of Ocean Energy Management, and the Office of Naval Research. The U.S. Integrated Ocean Observing System Program Office provides coordination. MBON has developed best practices and advanced means to collect samples for eDNA analysis using autonomous underwater vehicles.

Considerations for Congress

Congress may consider several eDNA/eRNA policy questions related to protocols, databases, and different uses for eDNA/eRNA data.

Multiple federal agencies have developed protocols for developing and using eDNA/eRNA. These protocols differ according to scientific purpose, study location, species, and sampling media. Congress may consider the potential benefits and challenges of tasking a federal agency to develop national standard protocols for sampling, collecting, and analyzing eDNA/eRNA. Standardizing protocols could aid in research collaboration, interoperability of reference databases, and quality control, as well as affect how data is analyzed, shared, and used. Standardizing protocols—for better or worse—could affect certain research, and potential innovative solutions, where a unique or specific protocol is needed. Congress may also consider whether current and proposed agency investments in developing and maintaining eDNA/eRNA databases are sufficient to meet federal objectives, as well as the need of the broader scientific community and public.

Policymakers may also consider how current federal efforts related to the collection, use, retention, and access to digital DNA/RNA sequence data address data accessibility, reliability, and biosafety and biosecurity concerns. Congress may question whether the federal government should facilitate or regulate access to certain data or regulate certain uses. Congress may also consider how local, state, and federal agencies currently use eDNA/eRNA for decisionmaking purposes and whether current policies enable or hamper its use.

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