eDNA Toolbox Overview: Development and Opportunities

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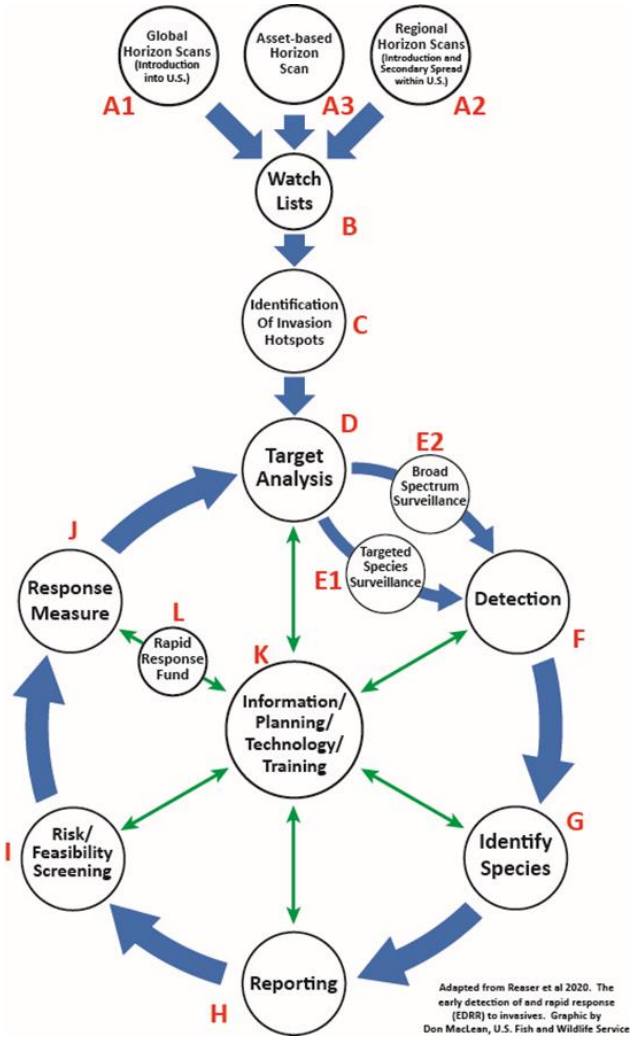
U.S. Department of Interior
U.S. Geological Survey

National Early Detection & Rapid Response Framework



FY22+ EDRR Framework Projects	FY23+ EDRR Framework Projects Planned (*) or Proposed
Horizon Scans & Hot Spot Analysis (A, B, C)	**Targeted Early Detection Surveillance (D, E, F)** Invasion Hotspots Asset Based Protection
Molecular detection @ points of entry (E, F, G)	
Resource Manager's eDNA Toolbox (D-G, K)	**EDRR Molecular Lab Network (G)** *Genetic Marker Development Processing Molecular Samples
Genetic Material Repository & Network (E, F, G)	
READI-Net: eDNA surveillance network (D-K)	**Rapid Response (J, L)** *Rapid Response Fund (Aquatic Invasive Spp) DOI Interjurisdictional Rapid Response Team
INHABIT (terrestrial plant) advancements (K)	
SIREN: National EDRR Information System (K)	

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Needs and concerns

Species of interest or concern

- National horizon scan
- Regional horizon scans
- Something on the state radar

Desire to survey and monitor

- Use of eDNA?

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My definition of eDNA

Detecting DNA in non-biological sources

- Beauty in its simplicity

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The thought process

I am concerned about species X

→ Is eDNA something useful for me?

→ How do I use eDNA to survey for species X?

→ What are the key components a monitoring program?

- Communications
- Best practices; Field → Lab
- Confidence in assay

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What are the key components a monitoring program?

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Are there resources out there to help me navigate this?

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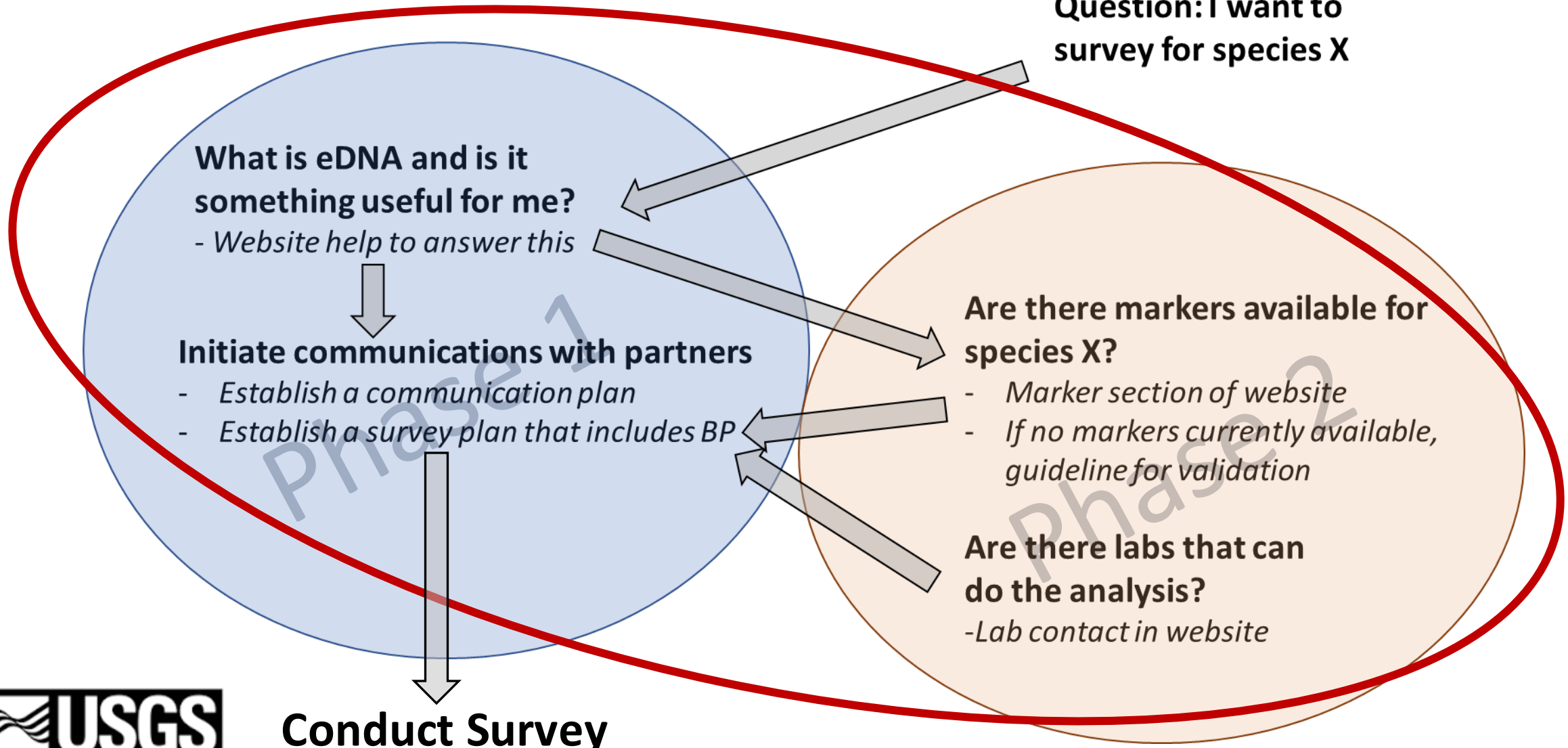
Project Vision



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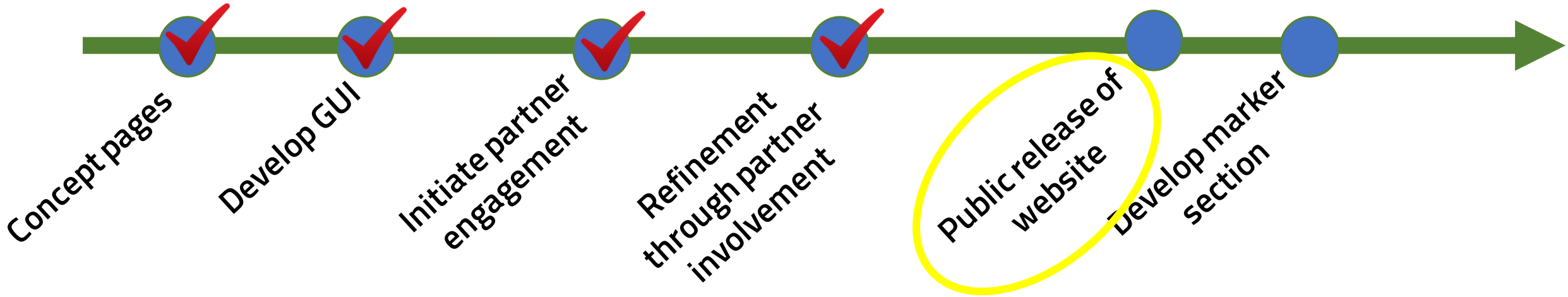
Current status of website



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Landing Page

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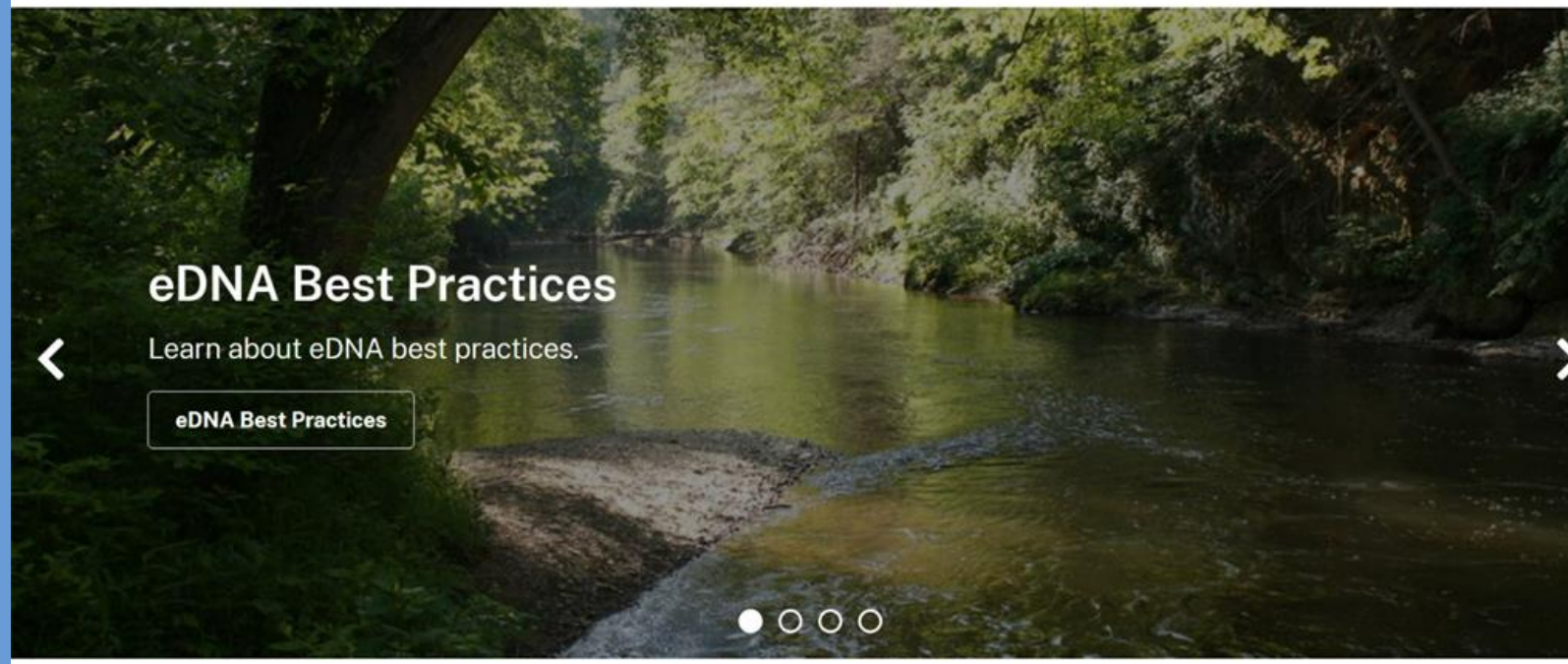
Resource Manager's eDNA Toolbox

ACTIVE

By Upper Midwest Environmental Sciences Center September 7, 2022



Overview | Science | Data | Publications | Software | News

Molecular tools have garnered a lot of interest in natural resource management, particularly in biosurveillance. Unfortunately, there are gaps in monitoring, characterizing, communicating molecular approaches, and easily accessible information. Current resource managers are unaware of the tools that are available within their systems, or are unsure of how best to interpret and communicate results.

Here we propose to create a centralized collection of techniques and resources in an interactive web-based interface that managers can use to assist in decision making and communication. This repository will be a web resource toolbox that allows managers to assess the available approaches, markers, validation

Study Area

Contacts

Upper Midwest

- Why Use Environmental DNA?
- Common Questions When Beginning an eDNA Surveillance Program
- Advantages and Limitations
- The Process of eDNA
- Molecular Methods
- Mitochondrial DNA
- References

Why Use Environmental DNA?

Environmental DNA is a powerful surveillance tool. Instead of relying on the capture of live organisms in the environment, utilizing eDNA methods allows you to monitor species without the need to physically observe them. eDNA also enhances your ability to detect rare species that would be difficult to monitor using traditional surveying methods. This allows scientists and managers to gain a better understanding of what species live in a specific environment. There are many advantages to using eDNA. Environmental DNA techniques can be used for:

- the early detection of invasive species.
- detection of cryptic or endangered species.
- revealing important information about biodiversity.
- monitoring the health of the ecosystem.

What is Environmental DNA?

DNA (deoxyribonucleic acid) is a large, helical shaped biological molecule present in organisms that contains genetic information. The complete set of genetic information is referred to as the genome and it has all the instructions for building and maintaining the organism. While the chemical structure of DNA is the same for every organism, differences in the sequence or order of the DNA building blocks (base pairs) make it possible to identify species, populations, and even individuals. An identifiable sequence at a recognized location in the genome is called a marker.

Environmental DNA (eDNA) is DNA present in various aquatic or terrestrial environments (water, soil, sediment). It's defined as "genetic material obtained directly from environmental samples without any obvious signs of biological source material."[15] eDNA originates from cellular material shed by organisms (via skin, excrement, etc.), and it can be extracted from complex environmental samples and analyzed using molecular techniques.

While eDNA is a powerful tool that resource managers can use for conservation efforts, biodiversity monitoring, and ecosystem management, it should not be used to replace traditional surveying methods. eDNA is used to contribute to the weight of evidence used for decision making and should be used to complement traditional methods.

Landing Page

Best Practices

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eDNA Best Practices - Resource Manager's eDNA Toolbox

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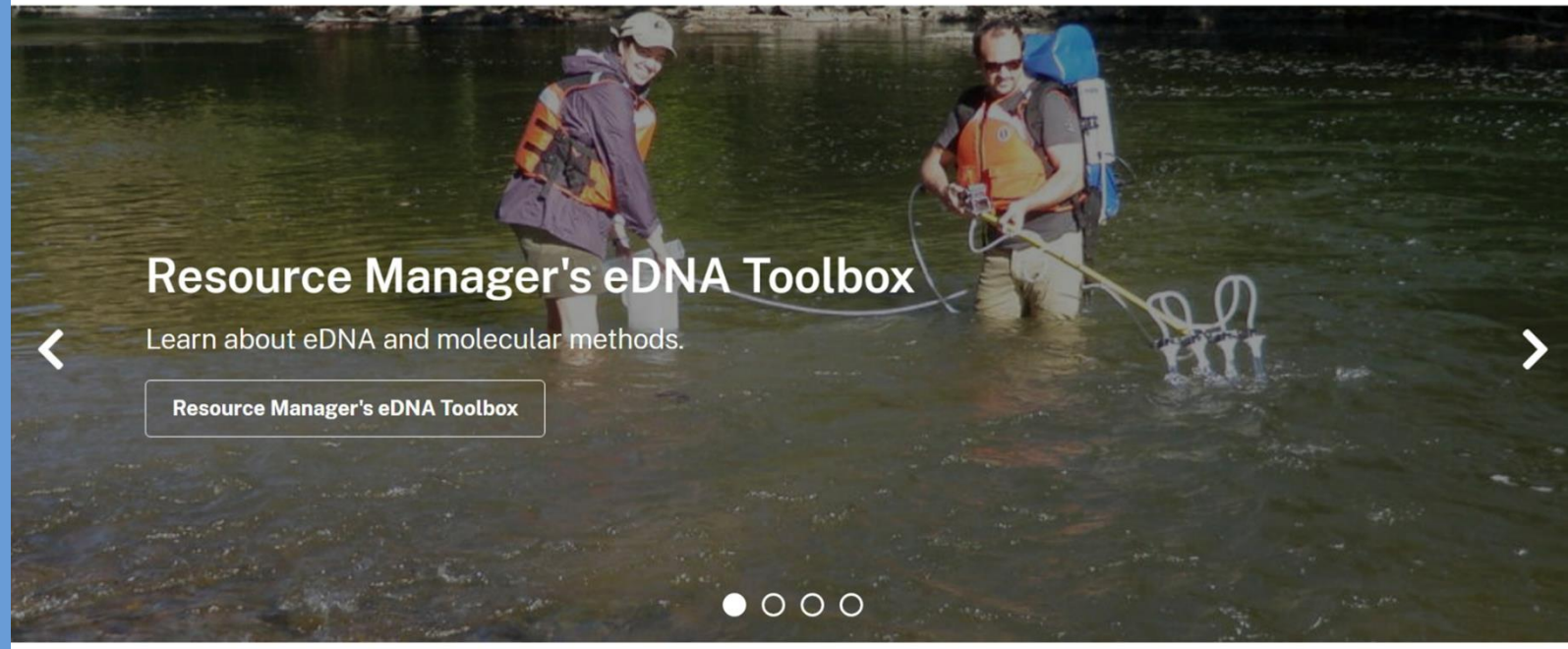
By Upper Midwest Environmental Sciences Center February 22, 2023



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While more than a decade old, the field of eDNA has yet to adopt a set list of standard best practices. The unique attributes of each eDNA study, location, and species have led to challenges for the identification of specific standards (such as protocols on eDNA sample collection, laboratory and data analysis, and concentration methods). We aim to develop broad best practices for eDNA data collection to ensure high-quality and repeatable data and improve manager and stakeholder confidence in data interpretation.

Study Area

Best Practices and Data Standards

The standards have been broken down into sections. Click on the section that you would like to learn more about.

1. **Develop a Communication Plan**
2. **Study Design and Pilot Study**
3. **Assay Validation**
4. **Sample Collection**
5. **Laboratory Analysis**
6. **Data Analysis and Interpretation- Communication**




Study Design and Pilot Study

When starting an eDNA study, the codesign of the study with natural resource managers is important to decide on goals, select proper methodologies, and develop study design. Certain steps are recommended before you begin fieldwork.[6]

- Use probe-based quantitative PCR when targeting a single species or genus.
- When targeting communities or unknown species, use eDNA metabarcoding with high-throughput sequencing.
- You must validate eDNA assays in silico, in vitro, and in situ.
- It's recommended to run a pilot study to assess detection probabilities with given sampling methods.
- This includes data such as pore sizes, sample volume and number, and filter material.
- Test extraction and analysis protocols.
- Engage with partners or other interested parties to decide on a communication plan and the definition of a positive detection.

Explore Search

Biology

eDNA

Best Practices

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Communications

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Communication Strategies- Resource Manager's eDNA Toolbox

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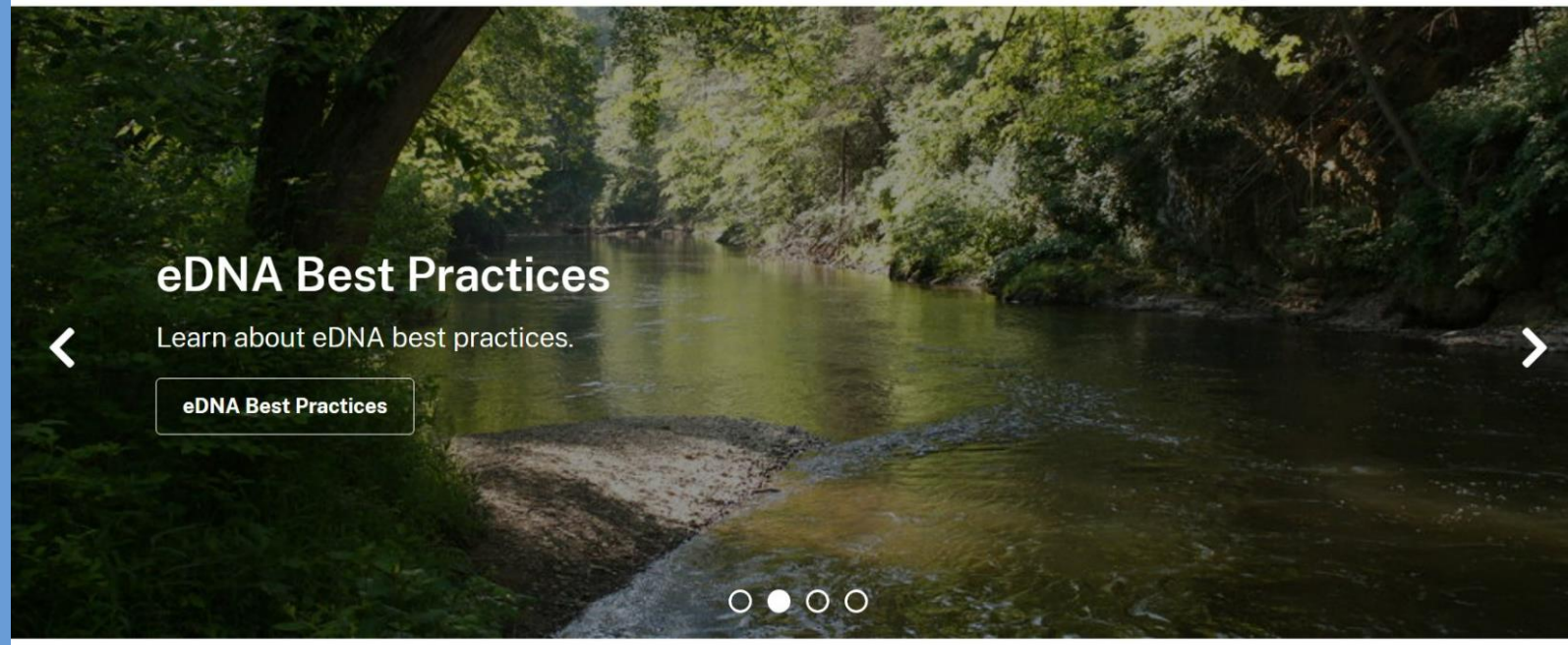
By Upper Midwest Environmental Sciences Center September 7, 2022



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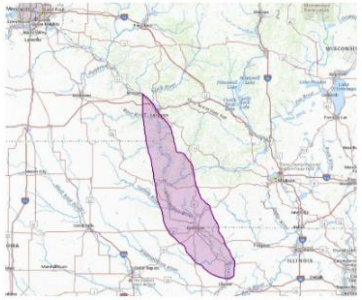
When conducting eDNA studies, communication with both internal and external entities is crucial. Within this section, we highlight templates for communicating results, detection decision points, and detection criteria. You will also find information on false negatives, false positives, and the roles and responsibilities involved in eDNA studies.

Study Area

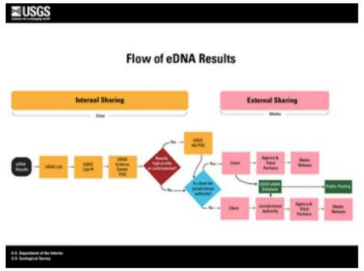


Resource Manager's eDNA

eDNA Best Practices

Flow of eDNA Results

The diagram to the right represents the general flow of eDNA results and how the information is passed internally before being made available to external clients.



Sources/Usage: Public Domain.

Figure 1. This diagram illustrates the general flow of eDNA results and how the information is passed internally before being made available to external clients.

Detection Decision Points

The flowchart to the right highlights important detection decisions involved during eDNA analysis. Please click on the image for further information on the steps involved.

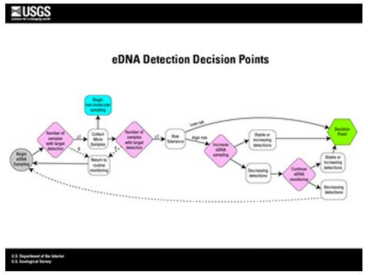


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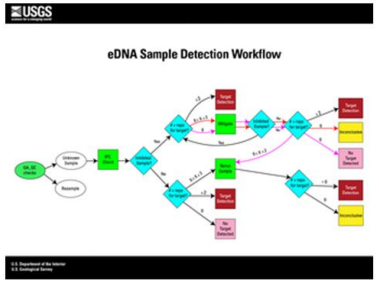
Figure 2. The flowchart above is an example of the steps involved in eDNA detection and the decisions involved during the analysis.

Detection Workflow

The flowchart to the right is an **example** diagram of an eDNA sample detection workflow and the steps involved. The thresholds (percent of PCR reps that amplify for the target) that indicate a detection vs. inconclusive results should be decided upon before you begin sampling. They should also be informed by the risk tolerance of the jurisdictional agency. **The thresholds shown in this diagram are for example purposes only**. Please click on the image for further information.



Sources/Usage: Public Domain.

Figure 3. The flowchart is an **example** diagram of an eDNA sample detection workflow and the steps involved.

Communications

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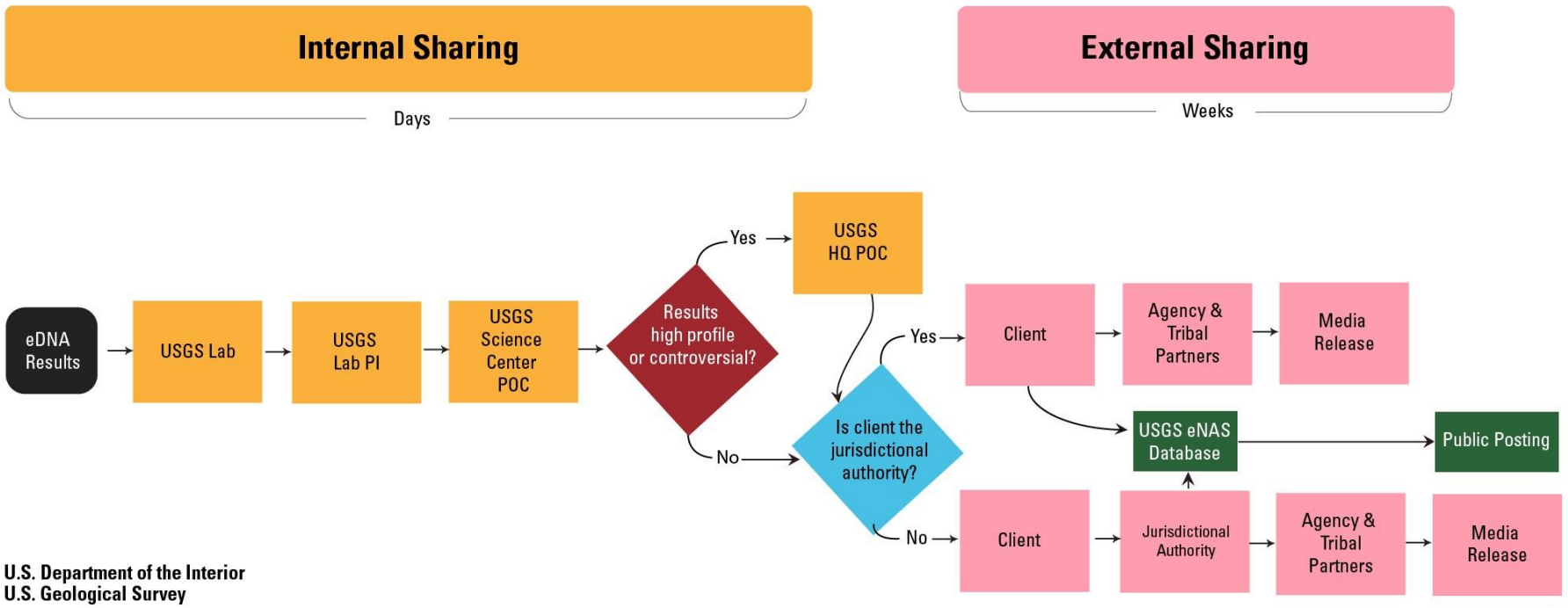
Flow of eDNA Results



U.S. Department of the Interior
U.S. Geological Survey

Things to consider?

How do you initiate the communication?

What are the plans if detected?

- **Agreement on what a positive detection is…**
- **What actions will be taken, if any?**

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Marker Section (Phase II)

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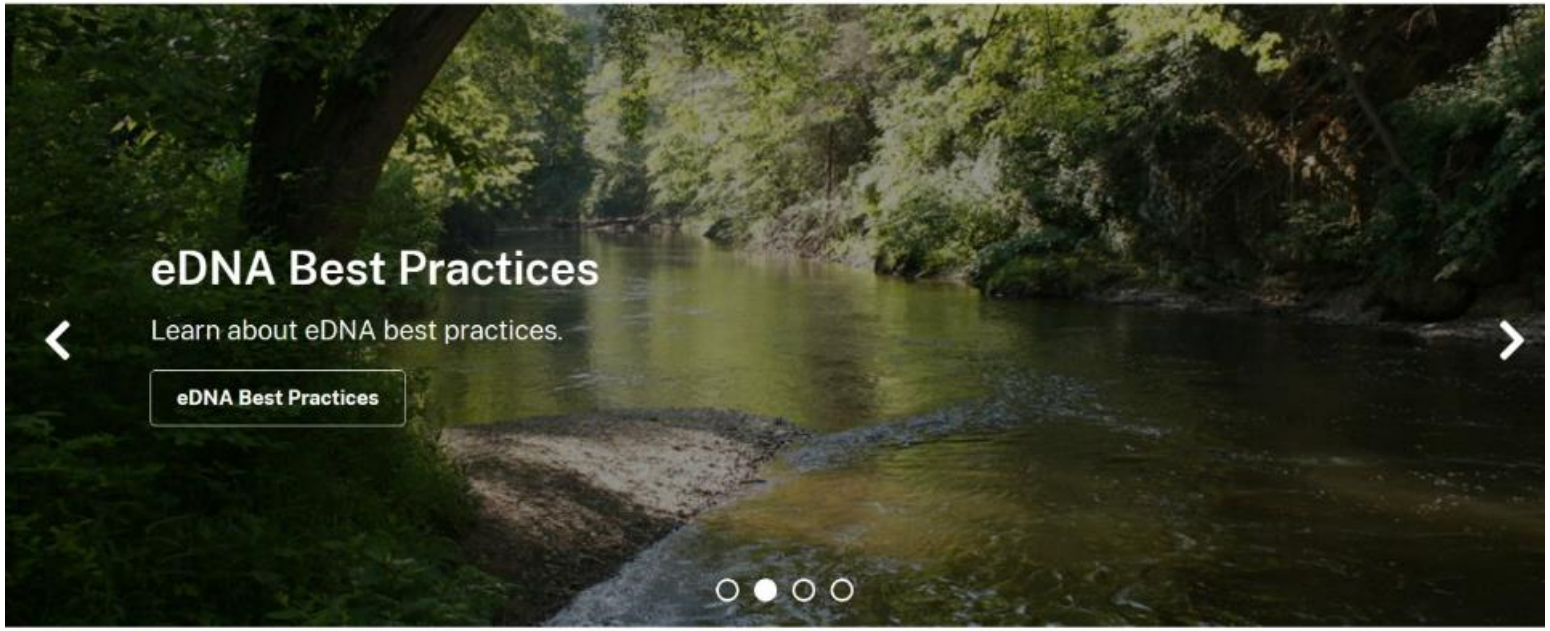
Genetic Markers- Resource Manager's eDNA Toolbox

By Upper Midwest Environmental Sciences Center September 7, 2022



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Study Area

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Explore Search

Biology

eDNA

Silver Carp

Molecular Method	Markers	Species validated against	LOD/LOQ	Point of Contact
qPCR	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	user@usgs.gov
LAMP	• Lorem Ipsum ○ Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	user@usgs.gov
Pan-Asian qPCR	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	user@usgs.gov

Bighead Carp

Molecular Method	Markers	Species validated against	LOD/LOQ	Point of Contact
qPCR	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	user@usgs.gov
LAMP	• Lorem Ipsum ○ Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum	• Lorem Ipsum • Lorem Ipsum • Lorem Ipsum	user@usgs.gov

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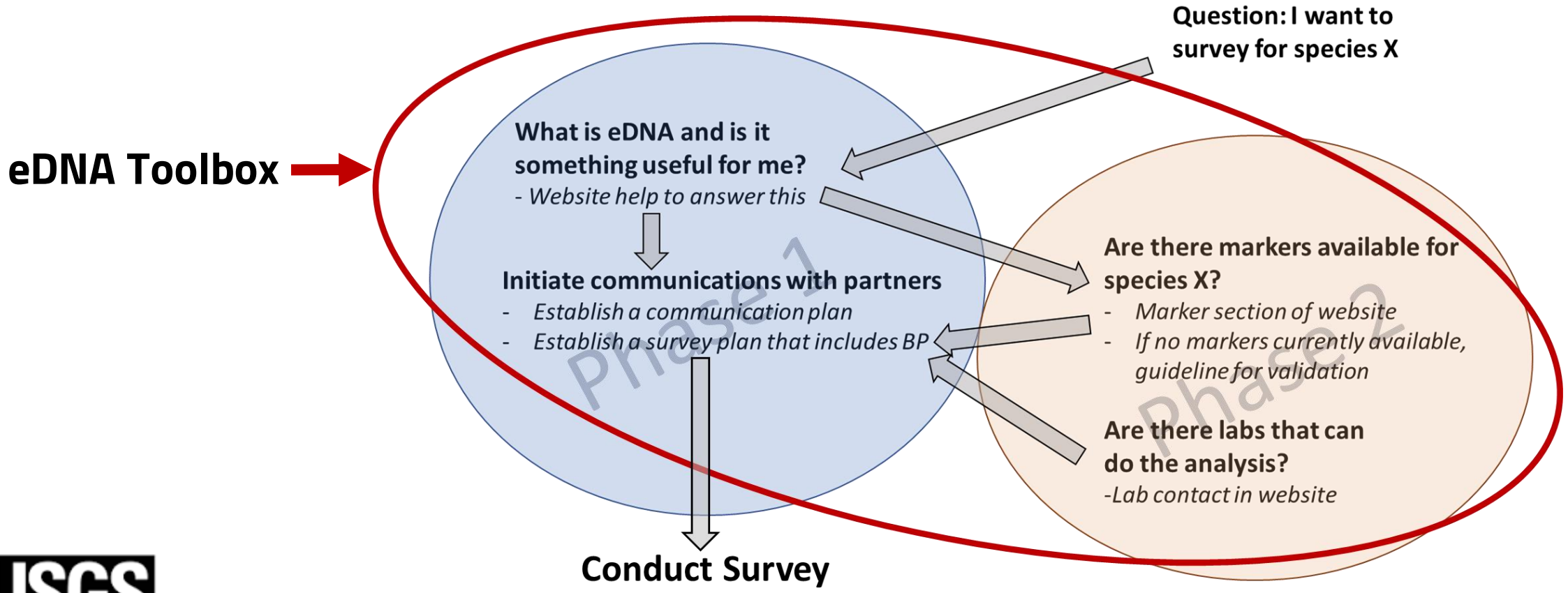
The hope…..

Site to help navigate the use of eDNA for early detection



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Questions

Contact info: Jon Amberg (jamberg@usgs.gov)

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