ENVIRONMENTAL DNA SURVEYS AND MANAGEMENT OF WILDLIFE ON MILITARY LANDS

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Camp Ripley
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Funding
- NDCEE
- ESTCP
- Installations: Fort Johnson, Fort Cavazos, Fort McCoy, USAG-HI

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Traditional Wildlife Surveys

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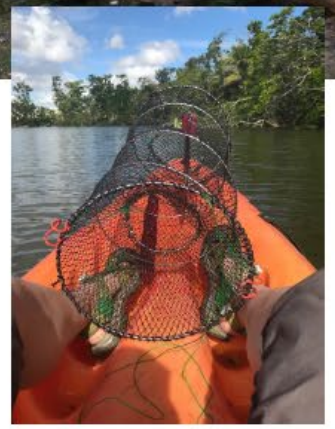
Traditional Wildlife Surveys



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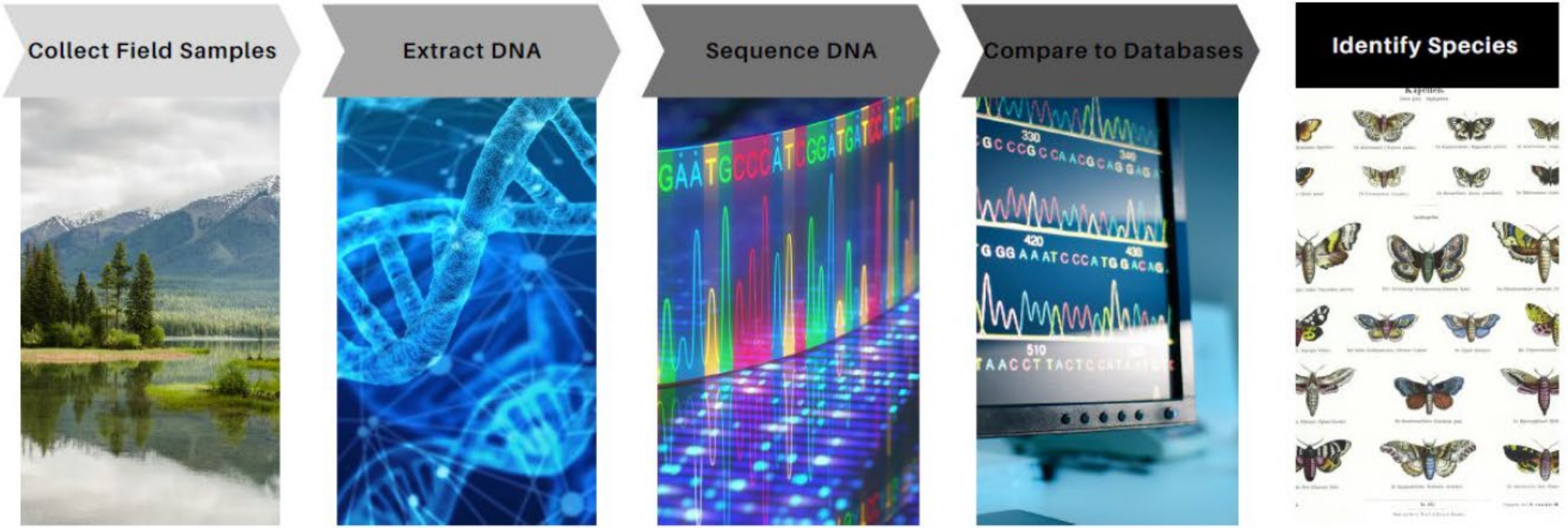
Environmental DNA

DNA shed by organisms into their environment (e.g., soil, water, air), which can be sampled and analyzed to detect species.

The eDNA Process



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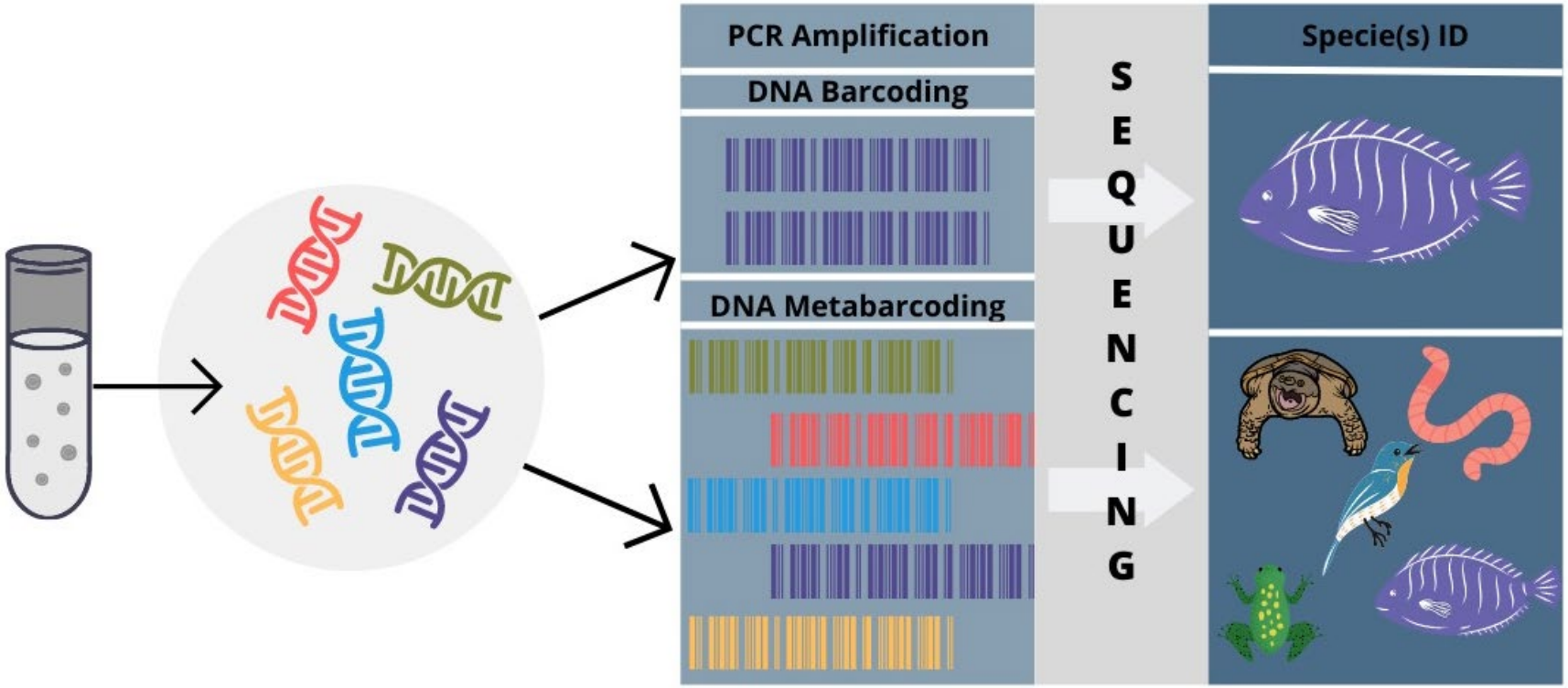
Single Species qPCR versus Community Metabarcoding



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Emerging eDNA Research

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Emerging eDNA Research

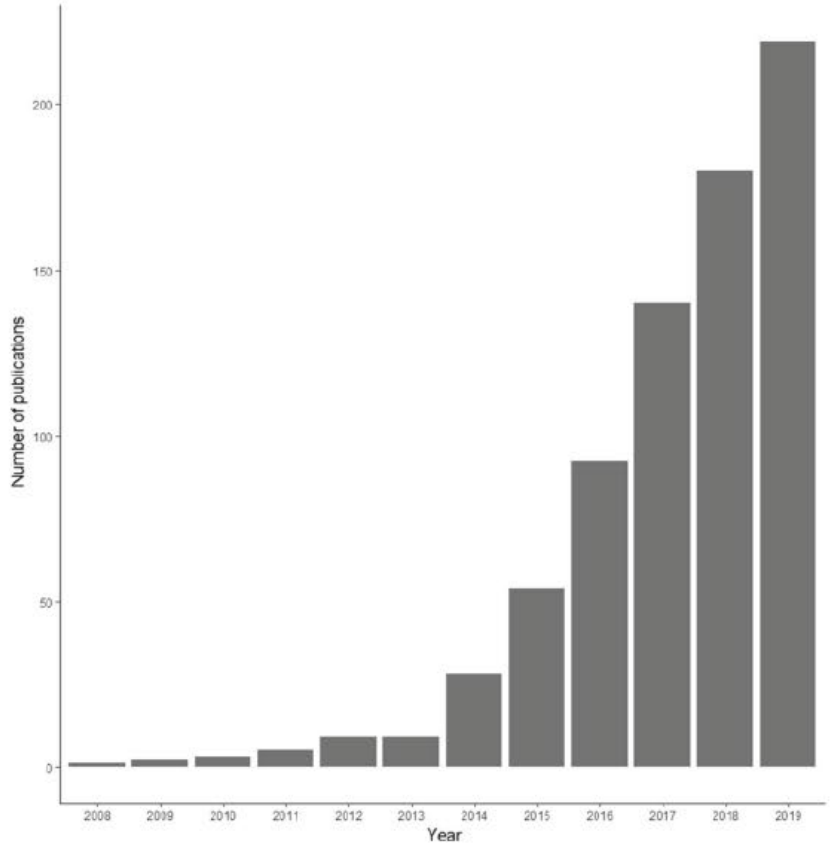
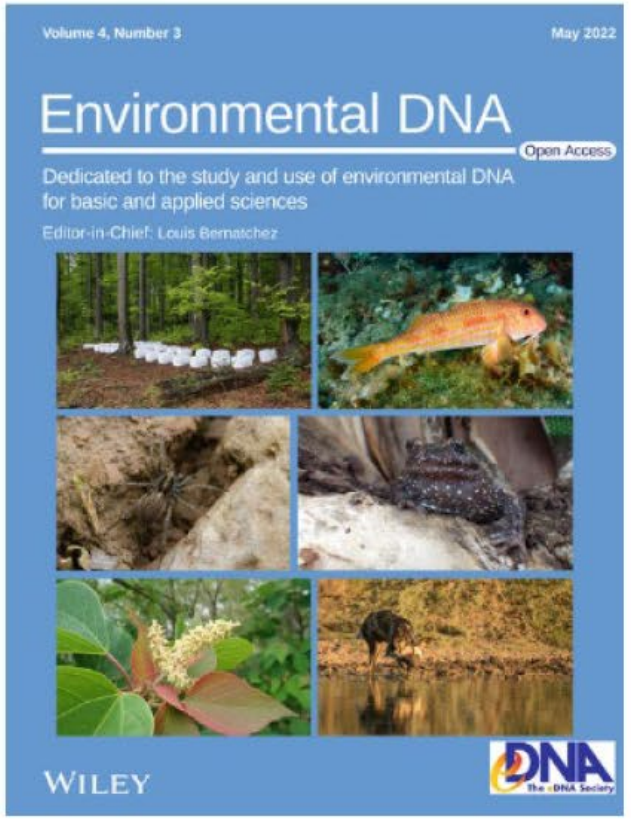


Fig. 1 Number of studies using environmental DNA (eDNA) recovered from a literature search with the words 'environmental DNA' OR 'eDNA' for the period between 1 January 2008 and 31 December 2019

Beng and Corlett. 2020. Biol. Cons. and Div.



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Examples of Management Questions for eDNA

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Where does this species occur on my installation?

Are conventional surveys effective?

What survey sites should be prioritized?

Does this species have parasitic or pathogenic infections?

What is the diet of this species?

Has this invasive species reached my installation?

What species pollinate or disperse this at-risk plant?

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eDNA on military lands for at-risk species

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Single-species qPCR
- Alligator snapping turtle (Fort Johnson)
- Blandings Turtle (Camp Ripley)
- Louisiana pine snake (Fort Johnson)
- Cave plethodon salamander (Fort Cavazos)

Multi-species Metabarcoding
- Freshwater mussels (Fort Johnson/Cavazos)
- Roosting bats (Fort Johnson)
- Aquatic communities (Fort McCoy)

Species Interactions
- Trout and parasitic gill lice (Fort McCoy)
- Fecal metabarcoding (Fort Johnson/Cavazos)
- Plants and pollinators (Fort McCoy and Oahu)

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Single Species qPCR

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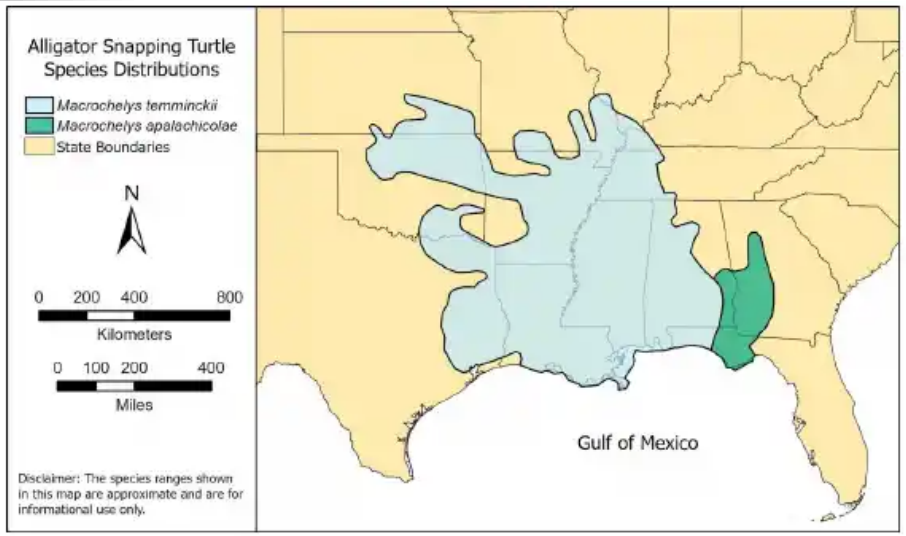
Alligator Snapping Turtle on Fort Johnson, LA



Distribution on Military Sites: The Alligator Snapping Turtle is confirmed present on the following 17 military sites:

- Air Force: Eglin AFB (Florida); Little Rock Air Force Base (AFB; Arkansas); Tyndall AFB (Florida); Moody AFB (Georgia); Barksdale AFB (Louisiana)
- Army: Fort Benning (Alabama/Georgia); Fort Polk (Louisiana); Camp Shelby (Mississippi); Red River Army Depot (Texas)
- Army National Guard: Robinson Maneuver Training Center (Arkansas); Fort Chaffee (Arkansas)
- Navy: Naval Air Station (NAS) Pensacola - Main Base (Florida); NAS Pensacola - NOLF Bronson Field (Florida); NAS Pensacola - Saufley Field NETPDTC (Florida); NAS Whiting Field (Florida); NAS JRB NOLA (Louisiana); NCBC Gulfport (Mississippi)

Department of Defense
Legacy Resource Management Program

Recommended Best Management Practices for the Alligator Snapping Turtle on Department of Defense Installations

Department of Defense Partners in Amphibian and Reptile Conservation

November 2021

Single Species qPCR

Alligator Snapping Turtle on Fort Johnson, LA

Conventional trapping

88.5 trap-nights/detection
29.2 work-hours/detection
$1152.81/detection

eDNA (qPCR)

16.3 samples/detection
17.5 work-hours/detection
$560.52/detection

eDNA compared to trapping:

- 5.55 times higher detection rates
- 51% cost-reduction per detection
- 40% fewer labor hours per detection
- 84% more streams surveyed per year.

Sternhagen et al. Environmental DNA, *in press*

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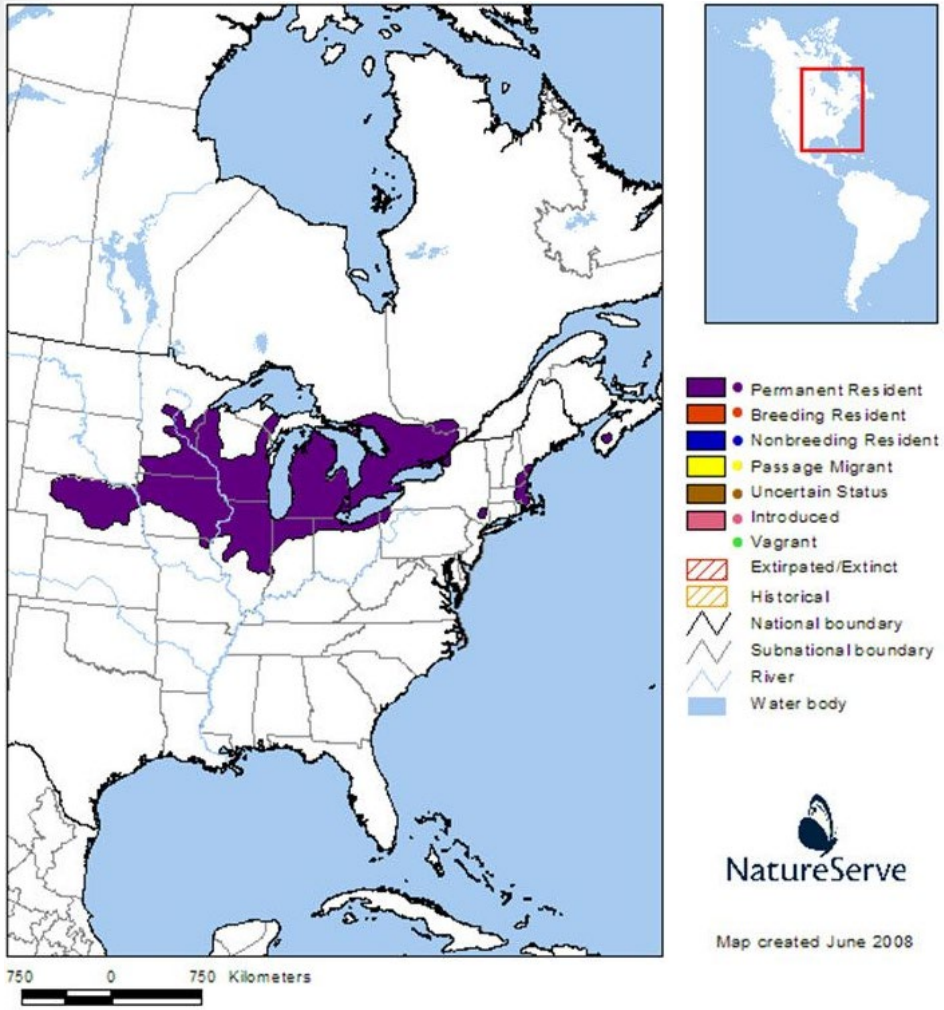
Single Species qPCR

Blanding's Turtle on Camp Ripley, MN



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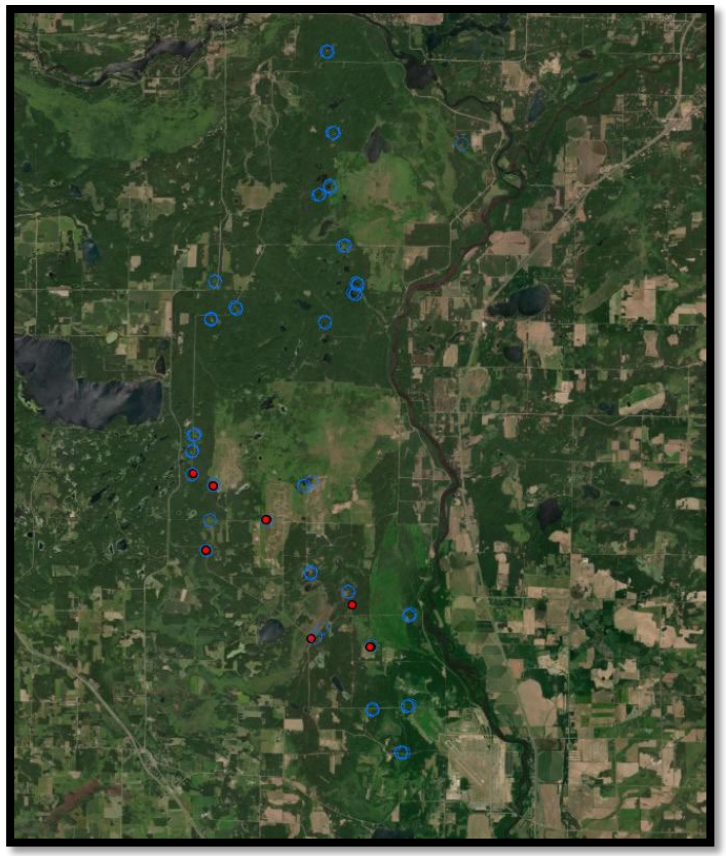
Single Species qPCR

Blanding's Turtle on Camp Ripley, MN



Sampling across 10 days (July – Sept)

Blanding's Turtle DNA detected at 7 Sites

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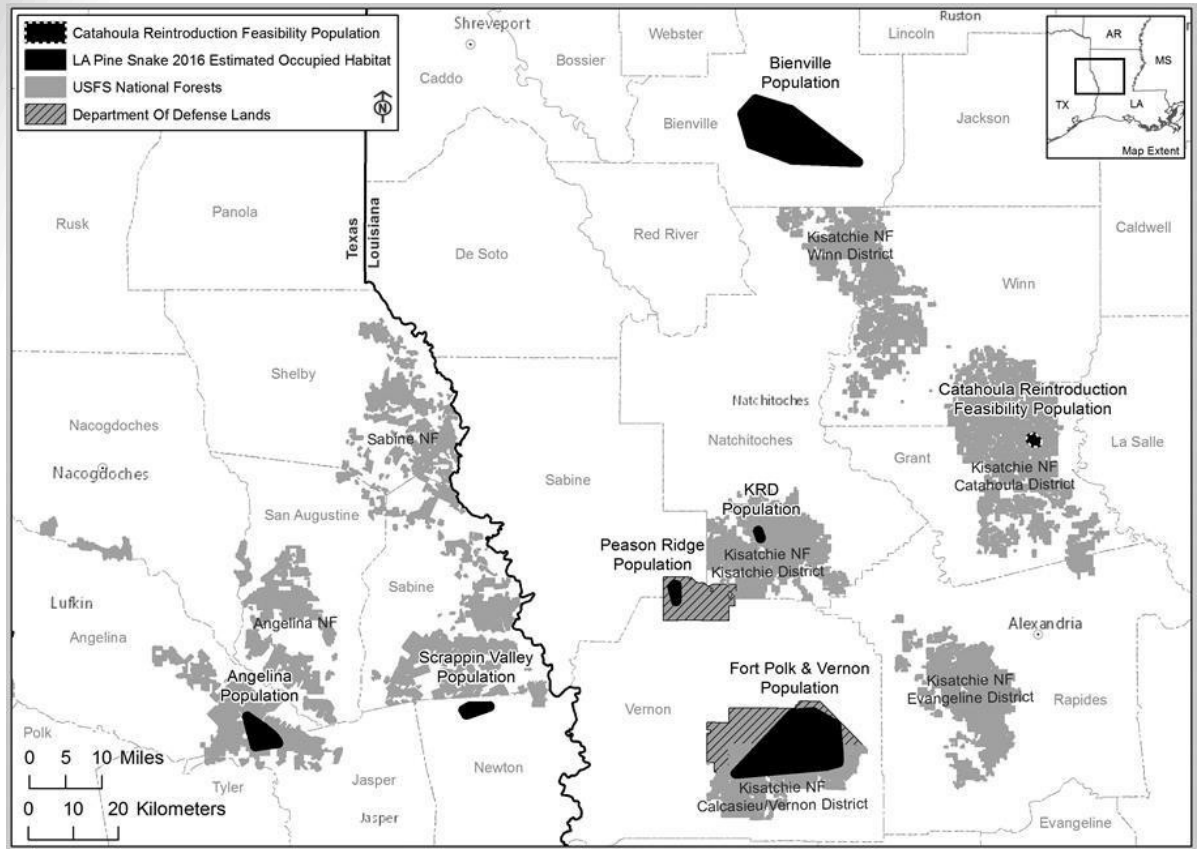
Single Species qPCR

Louisiana Pine Snake on Fort Johnson, LA



State	Estimated Occupied Habitat Area	Trap success (2008-2017)
LA	Bienville	1:1,259
	Kisatchie	0:20,293
	Peason Ridge	1:4,666
	Fort Johnson/Vernon	1:3,330
	Catahoula Reintroduction	1:770
TX	Scrappin' Valley	1:13,260
	Angelina	0:36,773

USFWS, SSA, 2018

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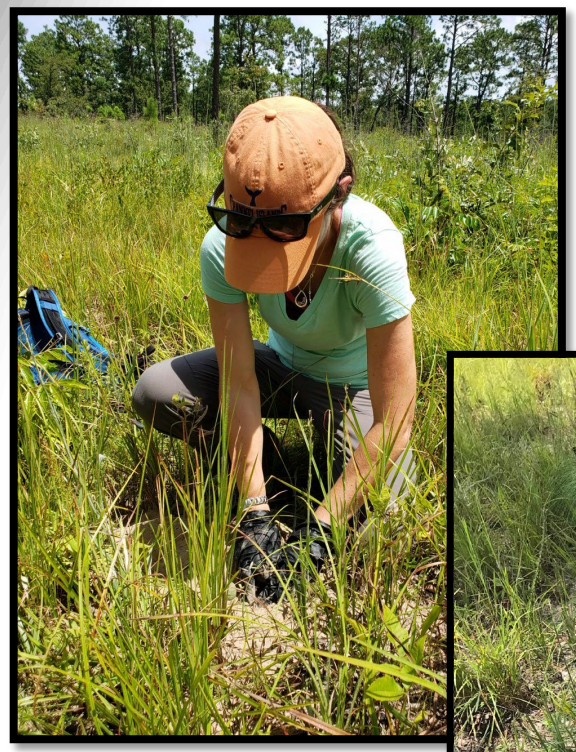
Single Species qPCR

Louisiana Pine Snake on Fort Johnson, LA



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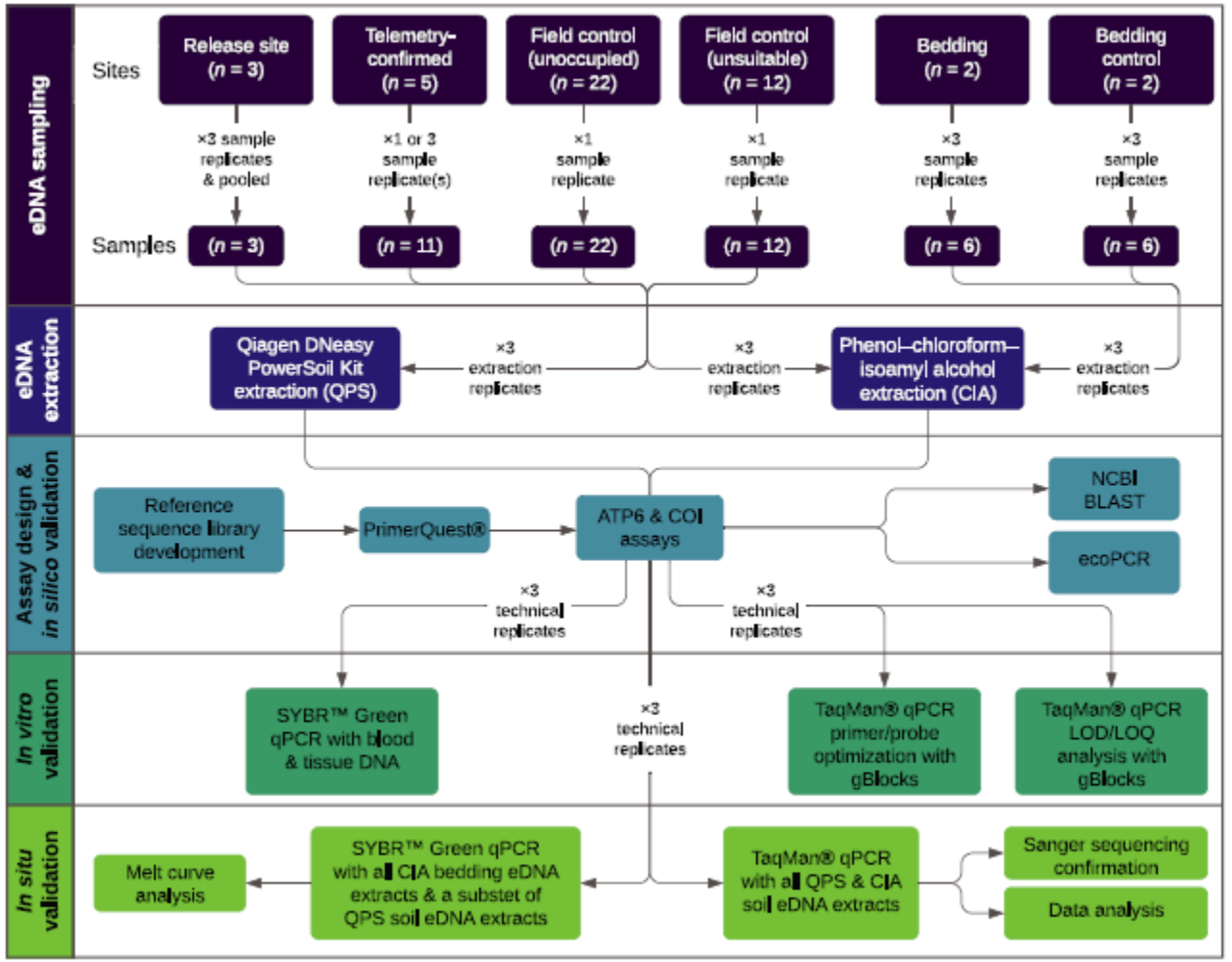
Single Species qPCR

Louisiana Pine Snake on Fort Johnson, LA



Single Species qPCR

Louisiana Pine Snake on Fort Johnson, LA

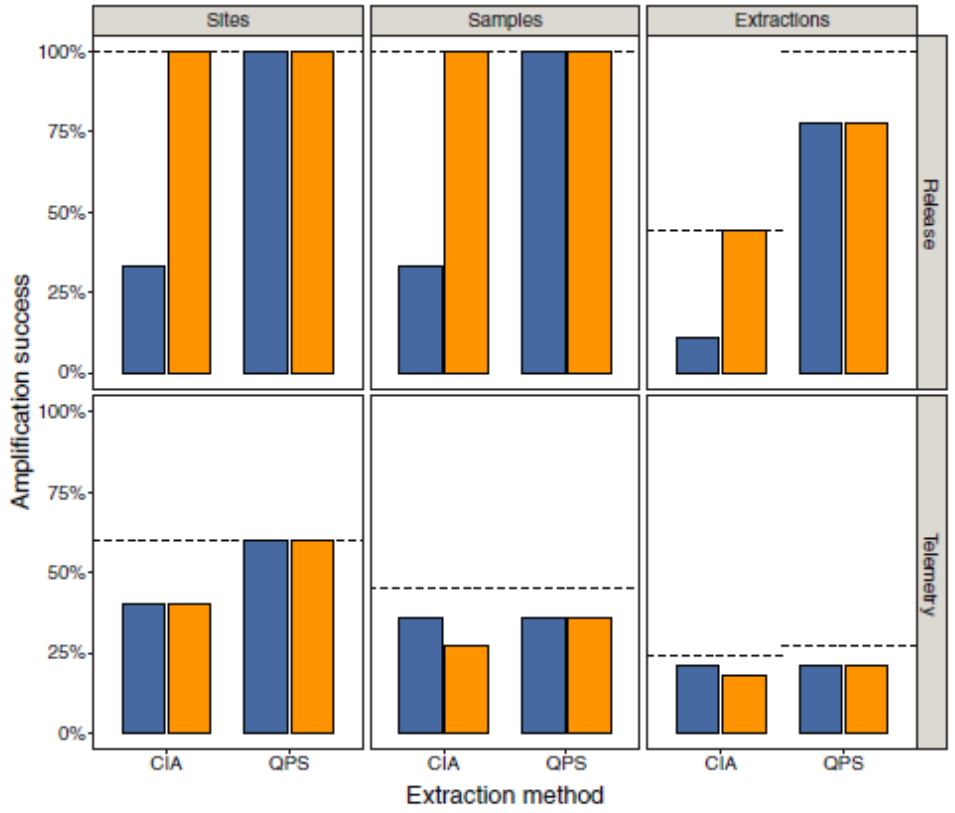
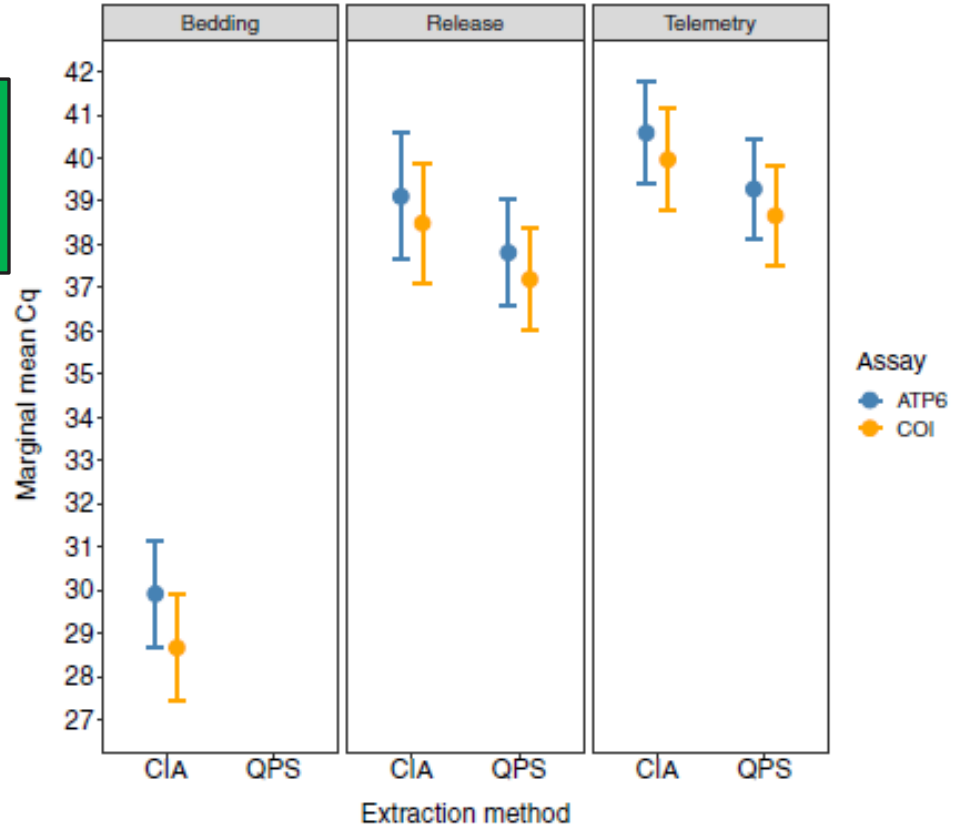


FIGURE 2 Percentage of sites, soil samples, and extractions with Louisiana Pinesnake eDNA detections by site category (release and telemetry-confirmed sites), extraction method (CIA and QPS), and assay (COI and ATP6). Overall amplification success, regardless of extraction method (for sites and samples) or assay treatment, is indicated by dashed lines

LPS DNA detected in bedding, release and telemetry sites!



Katz et al. 2020, eDNA

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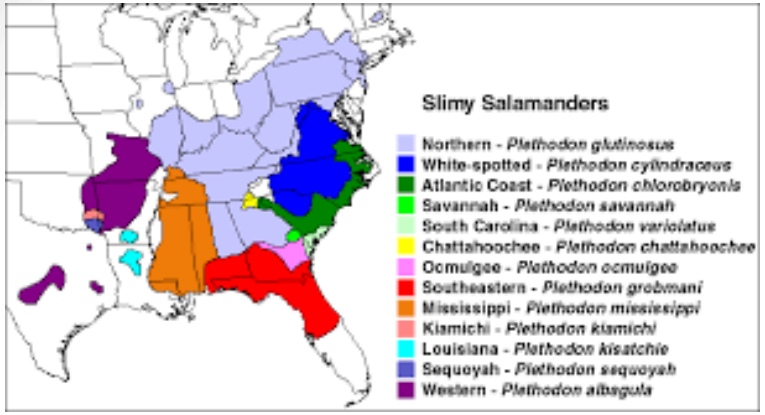
Single Species qPCR

Plethodon albagula in caves at Fort Cavazos, TX



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Single Species qPCR

Plethodon albagula in caves at Fort Cavazos, TX

Conventional Surveys:
- Detections at 18/37 (49%) of karst sites

eDNA:
- Detections at 18/37 (49%) of karst sites
- 89% (16/18) of known occupied sites
- Two sites where salamanders not detected via conventional

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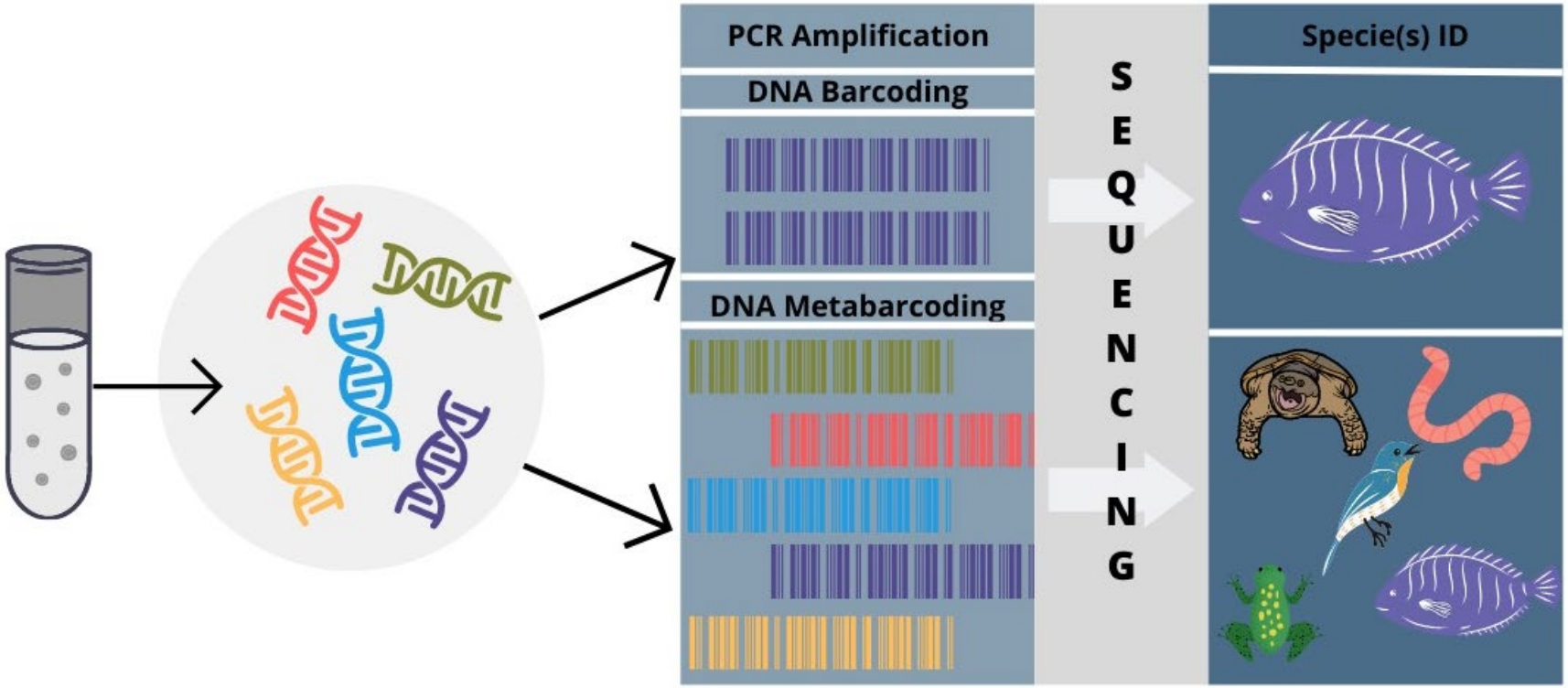
Single Species qPCR versus Community Metabarcoding



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eDNA Metabarcoding

Freshwater mussels at Fort Johnson and Cavazos

Balcones Spike
(*Fusconaia iheringi*)

Louisiana Pigtoe
(*Pleurobema riddellii*)

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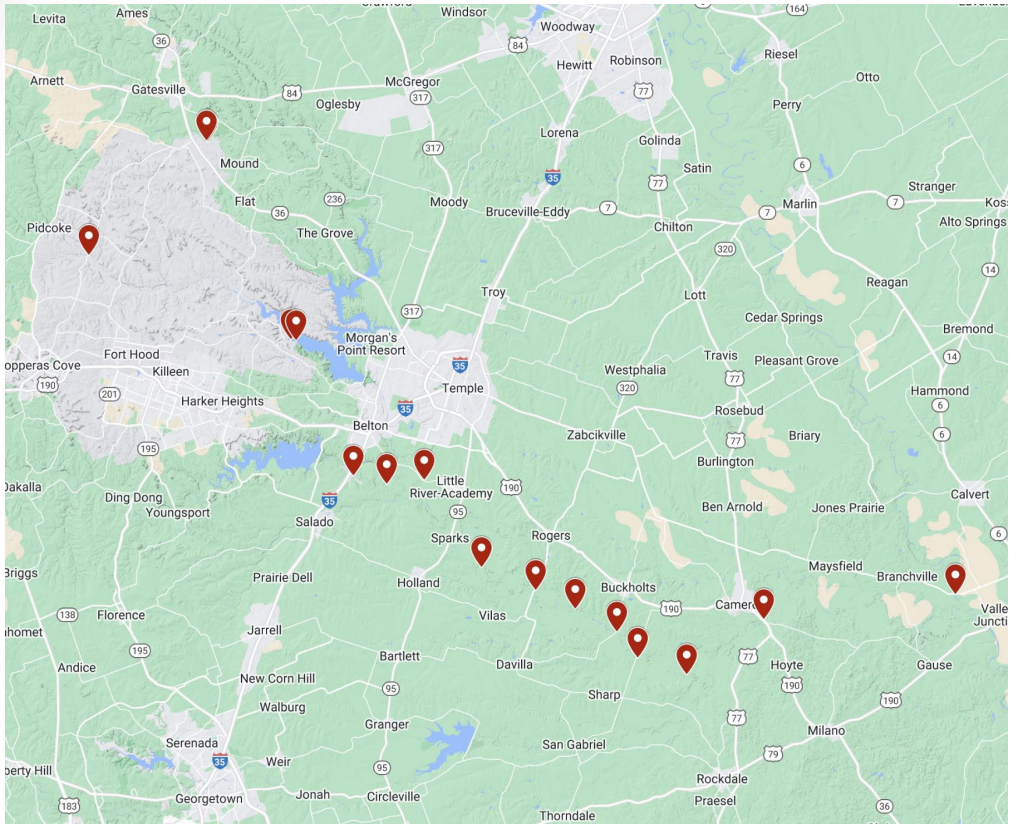
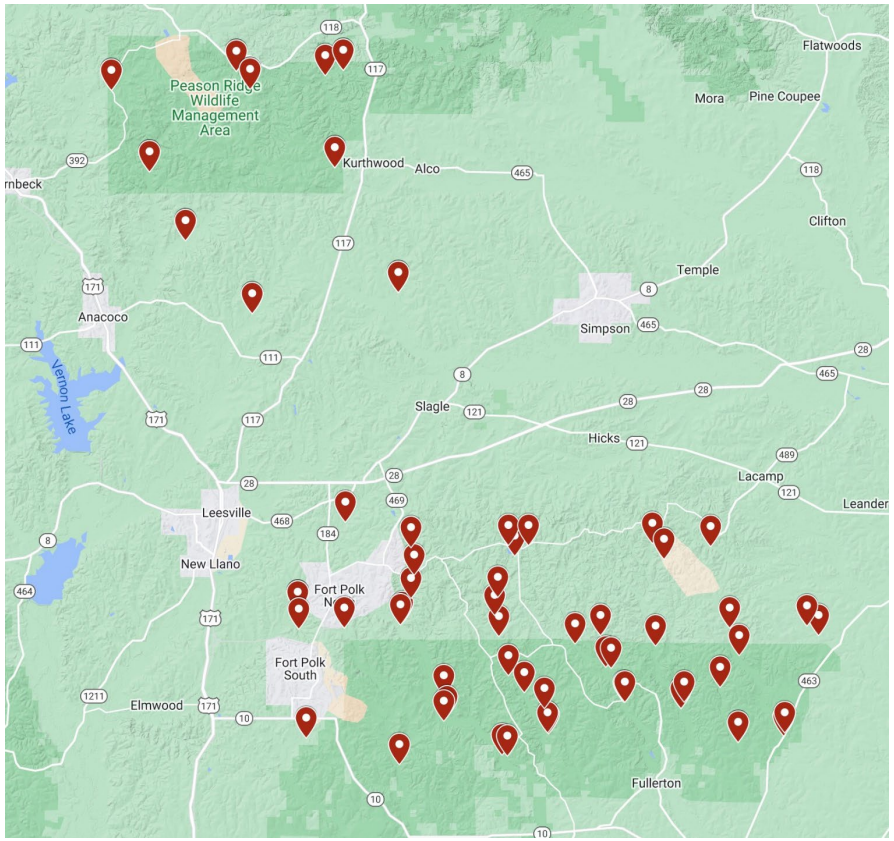
eDNA Metabarcoding

Freshwater mussels at Fort Johnson and Cavazos

16 sites in 4 days



70 sites in 9 days



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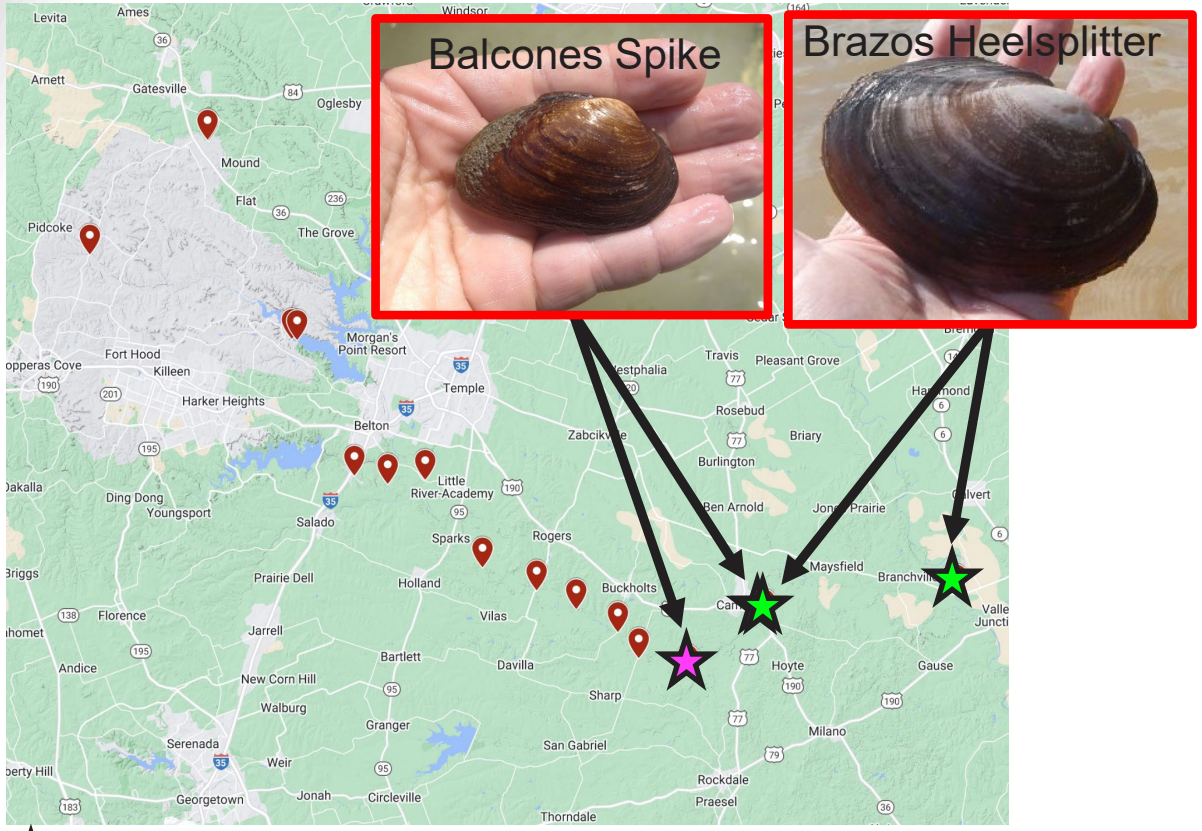
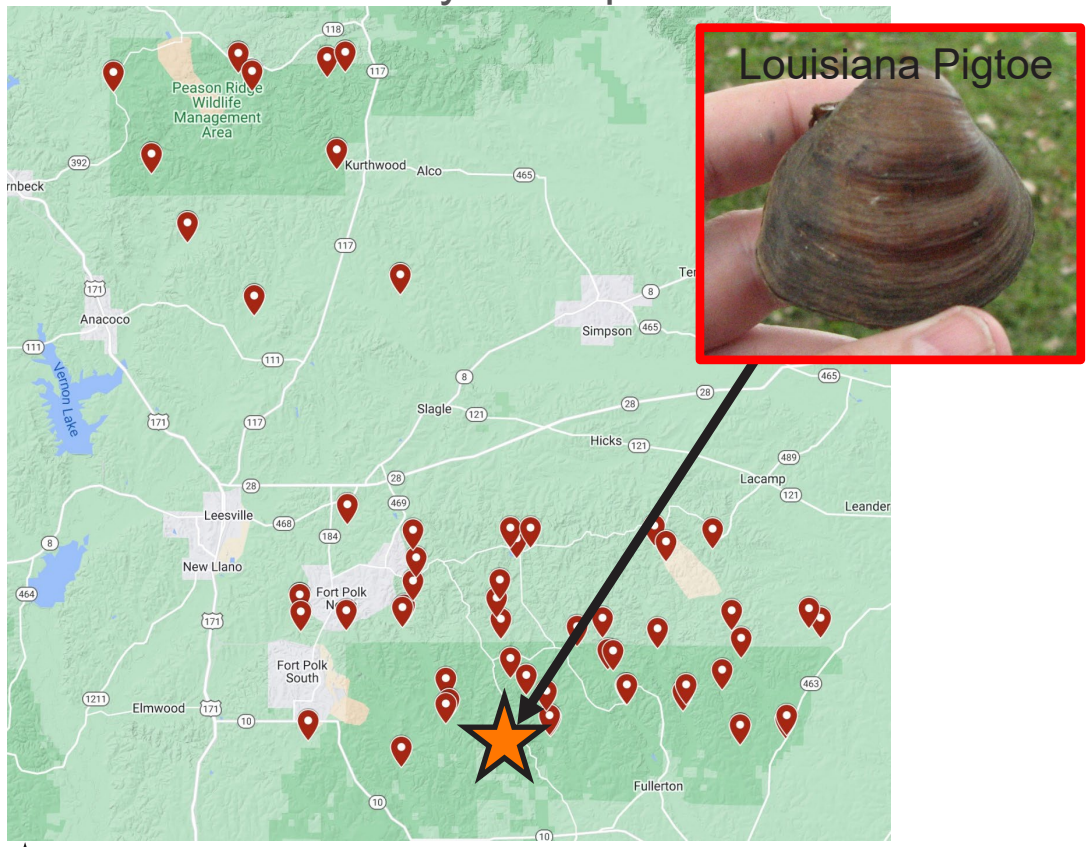
eDNA Metabarcoding

Freshwater mussels at Fort Johnson and Cavazos

16 sites in 4 days: 11 species



70 sites in 9 days: 10 species



Fusconaia iheringi, State Threatened

Potamilus streckersoni, State Threatened

Pleurobema riddellii, Proposed ESA Threatened

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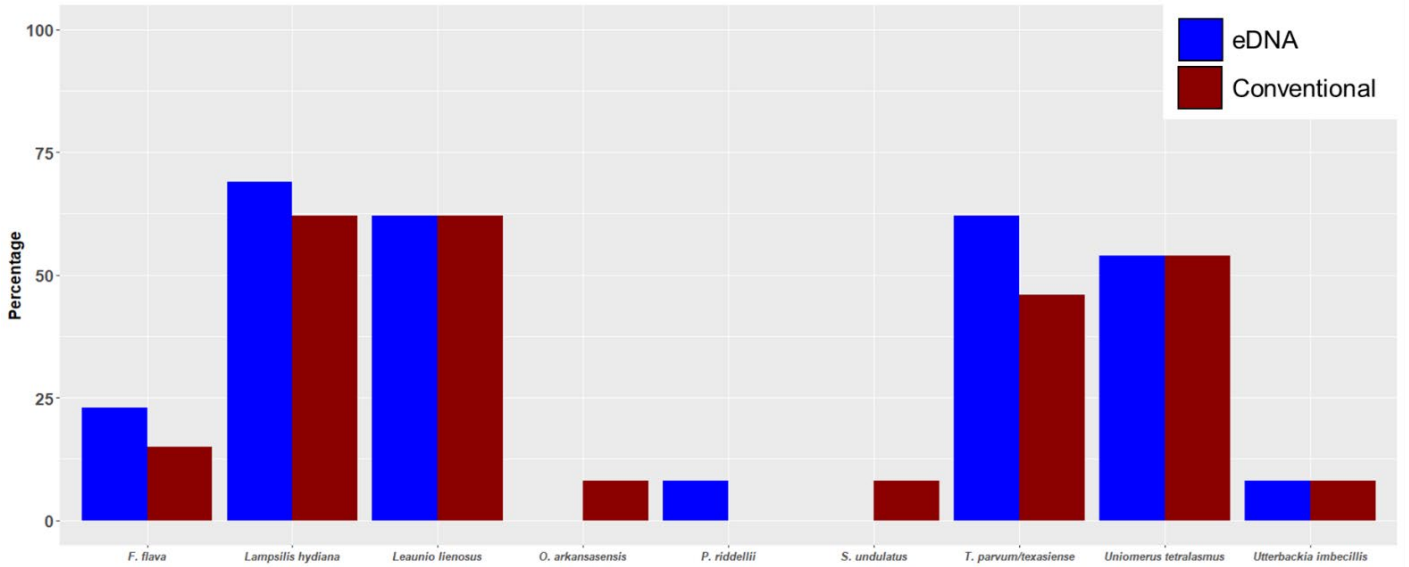
eDNA Metabarcoding

Freshwater mussels at Fort Johnson and Cavazos



eDNA:

- 84% concurrence with conventional surveys (2017, 2019, and 2023)

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eDNA Metabarcoding

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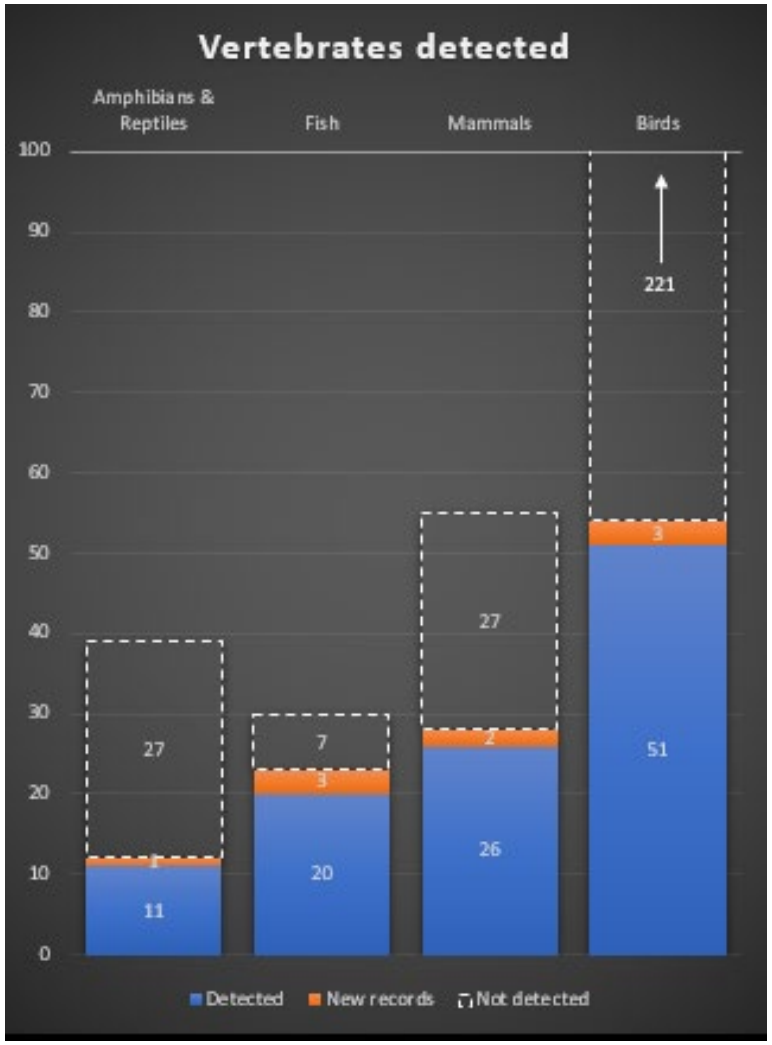
Freshwater aquatic communities at Fort McCoy, WI



- Water was sampled at 38 sites in 5 days
- 3 metabarcoding libraries (verts, inverts, plants)
- Sequenced on NovaSeq 6000 SP flowcell

Vertebrate groups	Species	% of known species detected
Amphibians & Reptiles	12	31%
Fish	23	77%
Mammals	28	51%
Birds	54	20%
Total	117	27%

117 total vertebrates
27% of known records
9 new records

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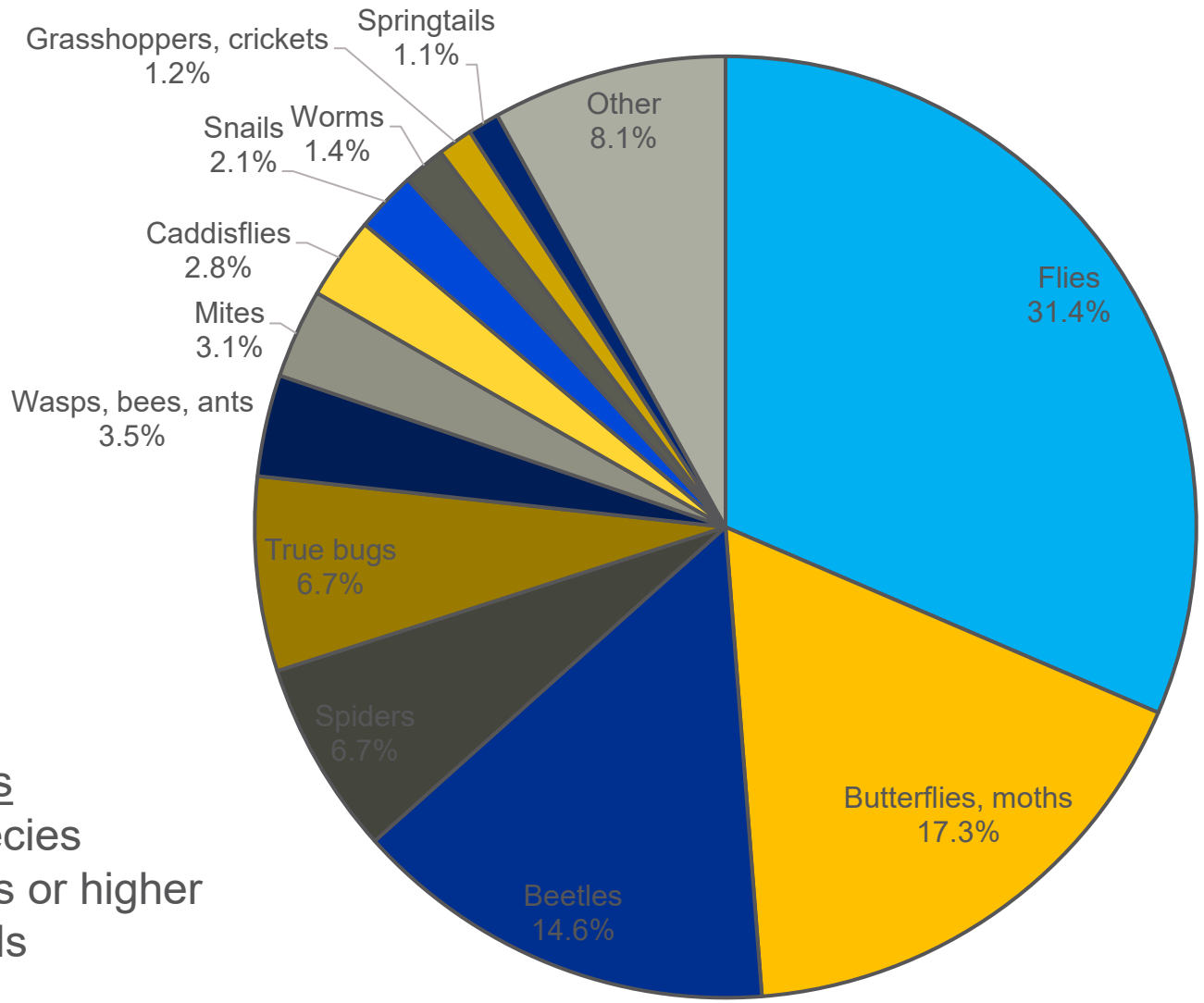
eDNA Metabarcoding

Freshwater aquatic communities at Fort McCoy, WI



1,663 total inverts
1,415 taxa to species
248 taxa to genus or higher
1,476 new records

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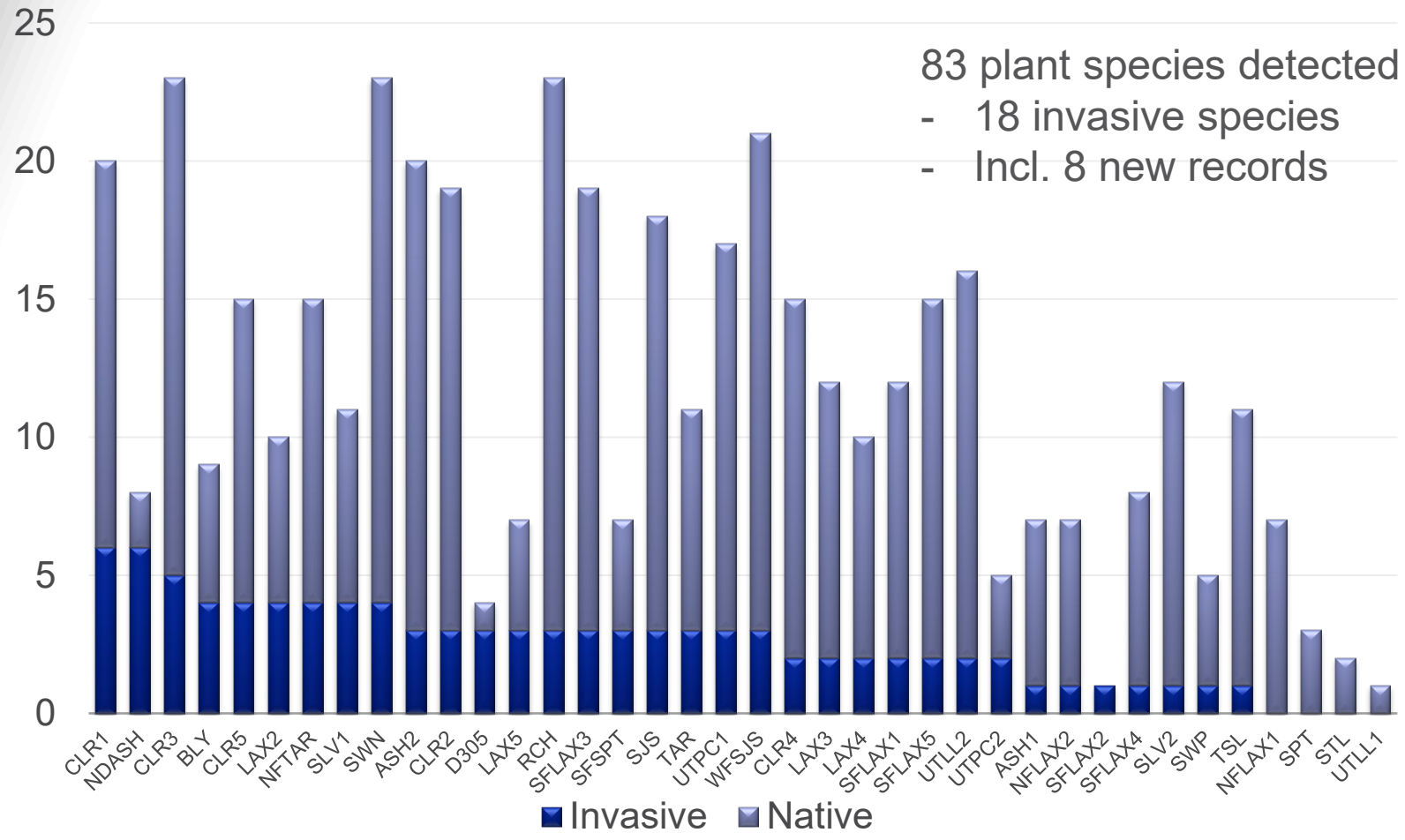
Freshwater aquatic communities at Fort McCoy, WI



18 invasive plants detected	New records
Black Cottonwood	
Climbing Nightshade	
Common Barley	Y
Crabgrass	
Garlic Mustard	
Giant-chickweed	Y
Glossy False Buckthorn	
Italian Ryegrass	
Knapweed	
Marshpepper Smartweed	
Meadow Fescue	
Perfumed Cherry	Y
Red-vein Dock	Y
Reed Canarygrass	
Scribner's Bluegrass	Y
Watercress	Y
Wavy Smoothcap Moss	Y
Woodland Bittercress	Y

Ft. McCoy summary:
5 days of sampling = 2,235 total species at 38 sites!

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eDNA Metabarcoding for Species Interactions

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Brook Trout and their parasitic copepods (*Salmincola edwardsii*)

Illustration: Duane Raver

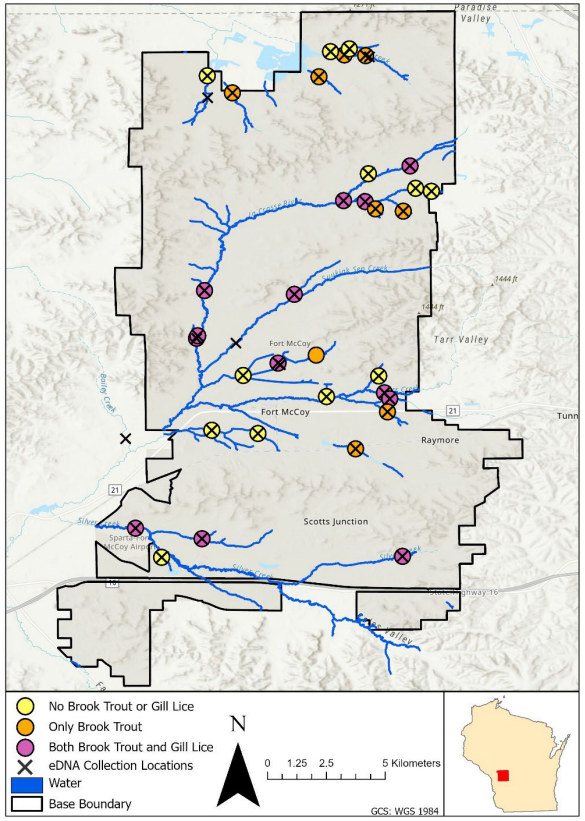
Photo: John Noble

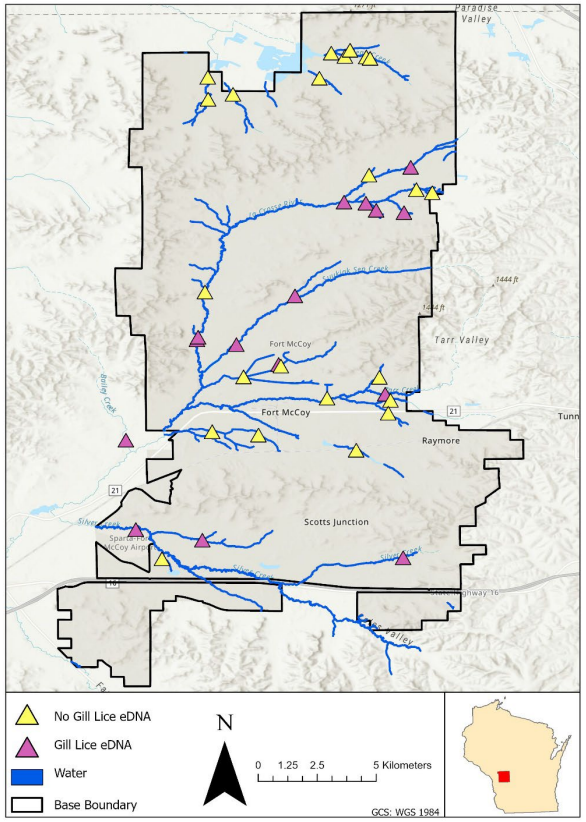
Photo: USGS

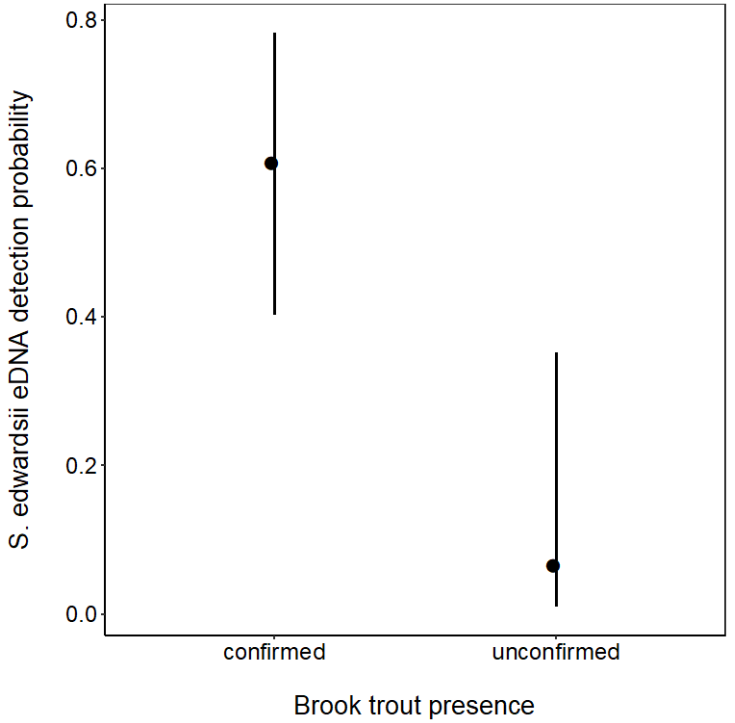
- eDNA metabarcoding data for gill lice, brook trout, and associated aquatic communities.
- qPCR to increase gill lice detection sensitivity
- Leveraged electrofishing survey data to validate eDNA approach
- Generated novel sequence data from gill lice collected at Ft. McCoy
  - qPCR assay development
  - metabarcoding taxonomic assignments
  - species confirmation

eDNA Metabarcoding for Species Interactions

Brook Trout and their parasitic copepods (*Salmincola edwardsii*)







No difference in gill lice detection probabilities:

- electrofishing vs. eDNA (p = 0.51)
- qPCR (15/38 sites) vs. metabarcoding (14/38 sites)

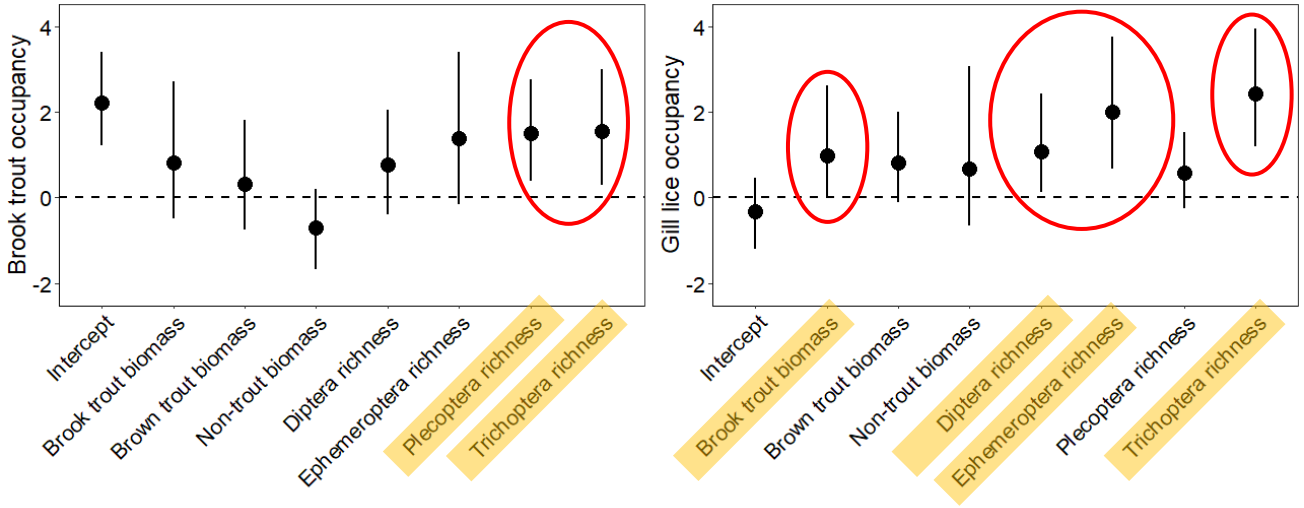
>21 times more likely to detect gill lice at sites with brook trout (p = 0.006)

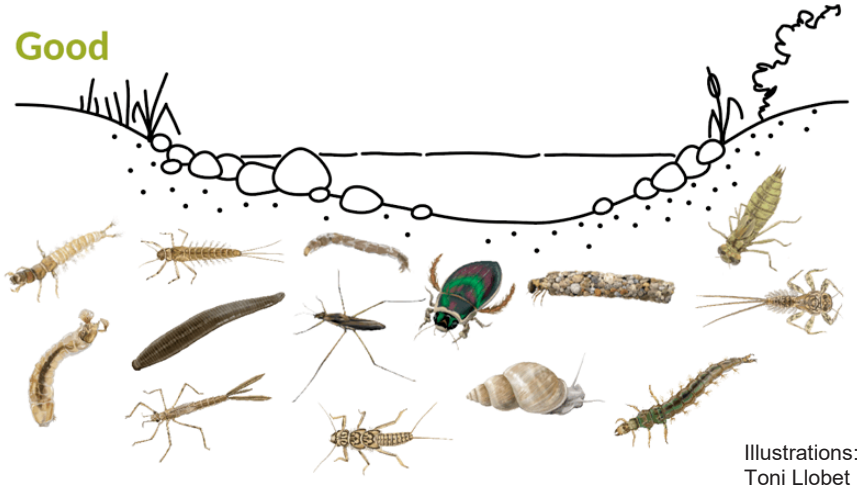
Katz et al., in prep

eDNA Metabarcoding for Species Interactions

Brook Trout and their parasitic copepods (*Salmincola edwardsii*)

Illustrations:
Duane Raver





Illustrations:
Toni Llobet

- Gill lice distribution dependent on host density
- Both related to aquatic insect diversity (i.e., host prey and habitat quality indicators)
- Brown trout not related to brook trout or gill lice occupancy

Tetzlaff et al., in prep.

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eDNA Metabarcoding for Species Interactions

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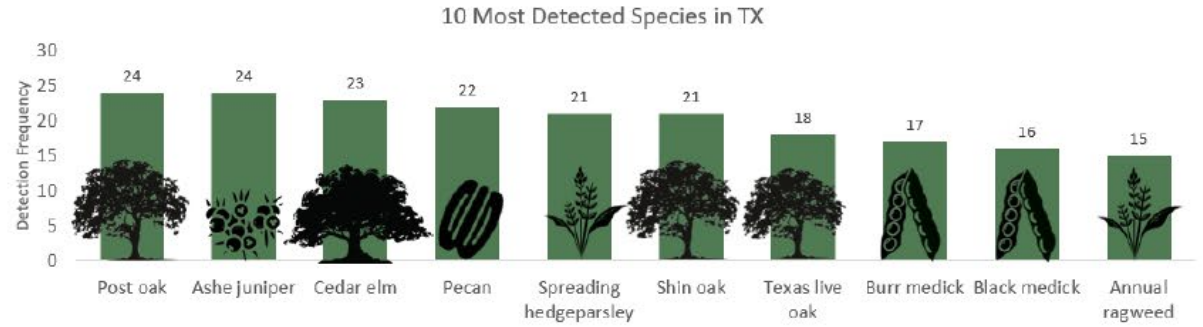
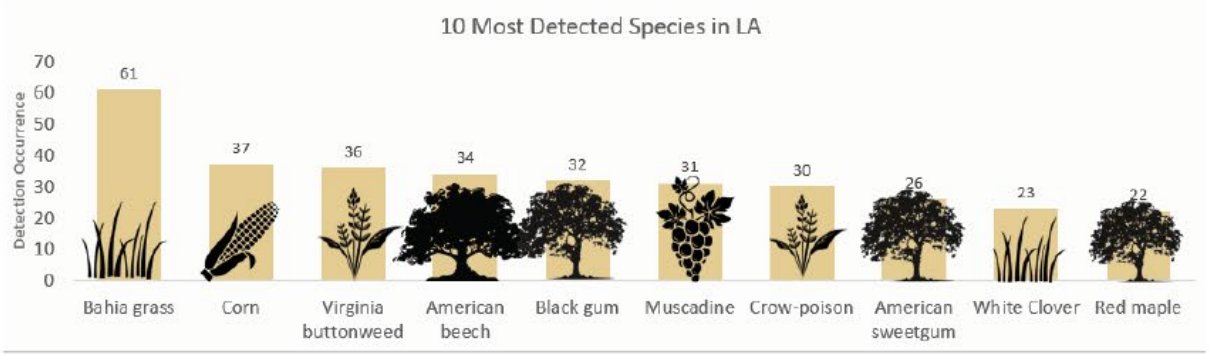
Fecal Metabarcoding of Feral Pigs at Fort Johnson and Cavazos

Species from Feces: Feral Hog Diet Analyses





187 species in Texas; 215 species in Louisiana

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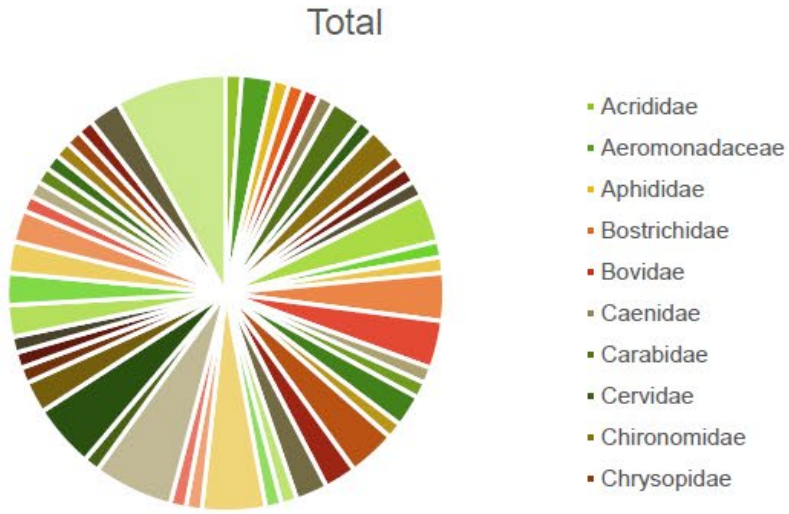
Fecal Metabarcoding of Bat Guano at Fort Johnson

Species from Feces: Bat species ID and diet analyses

- 114 guano samples
- 3 bat species
- Diet: 13 Orders, 47 Families, 18 Arthropod Species

No Pd (White-nose Fungus) DNA detected

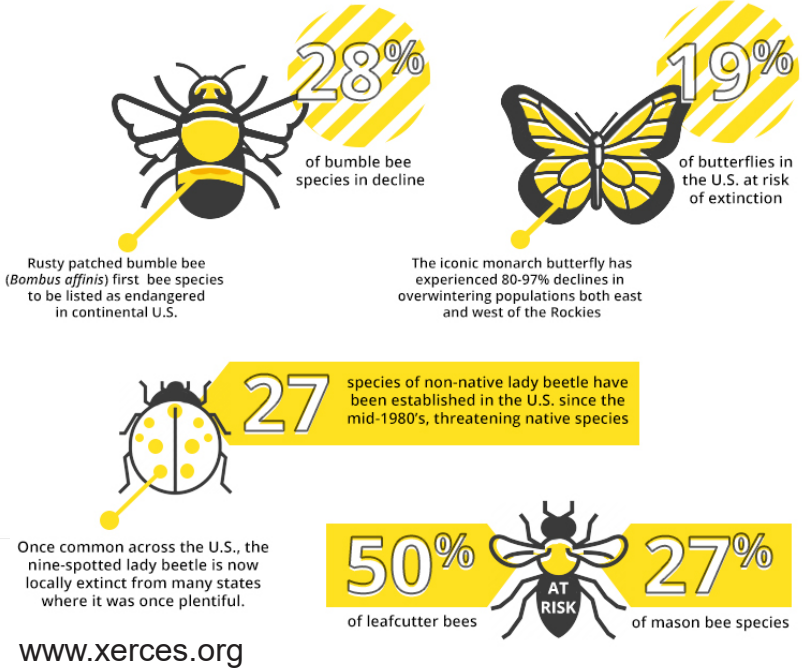


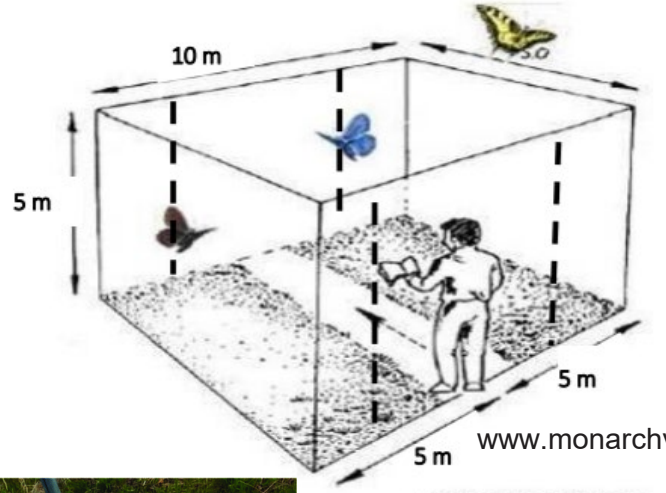
eDNA Metabarcoding for Species Interactions

Plant-Pollinator Interactions

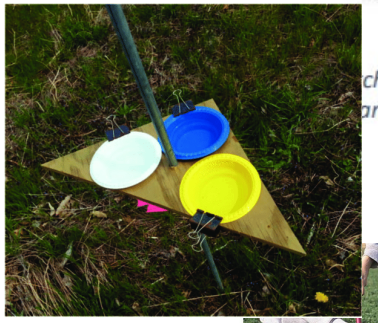
The New York Times

Decline of Pollinators Poses Threat to World Food Supply, Report Says





www.monarchventure.org



www.budburst.org

Joshi et al., 2015, Annals Ent. Soc.

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eDNA Metabarcoding for Species Interactions

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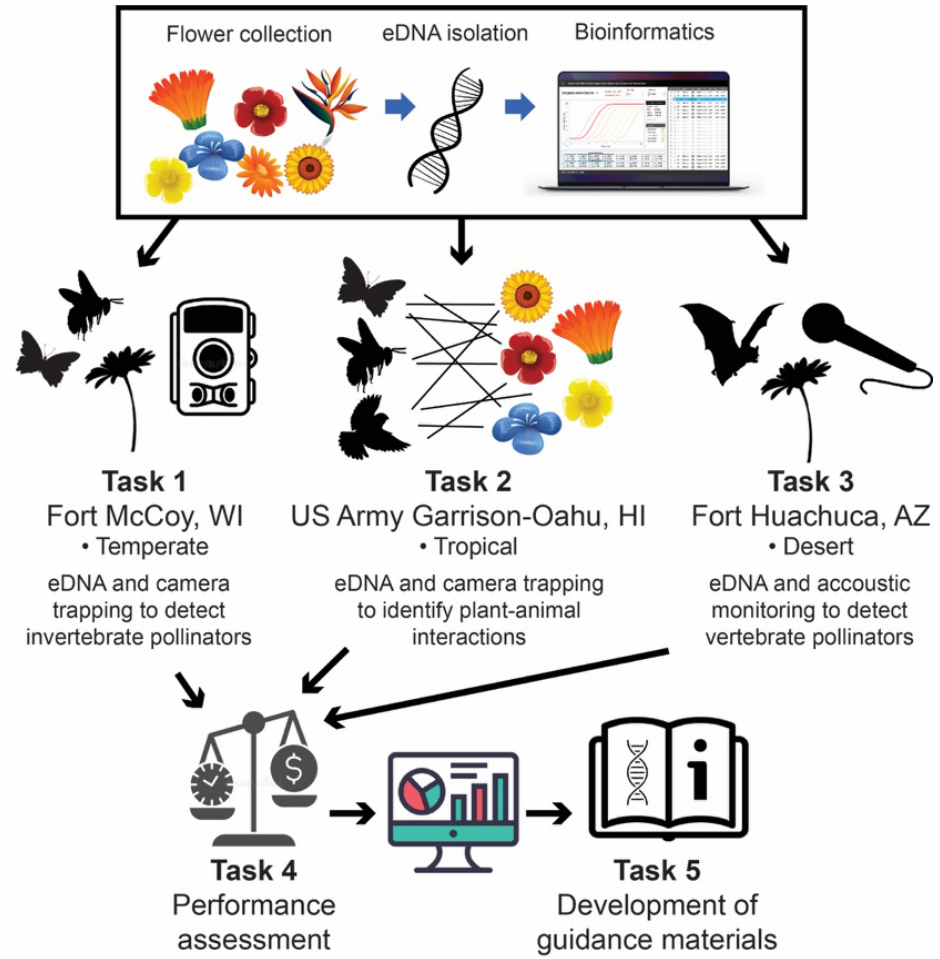
Plant-Pollinator Interactions

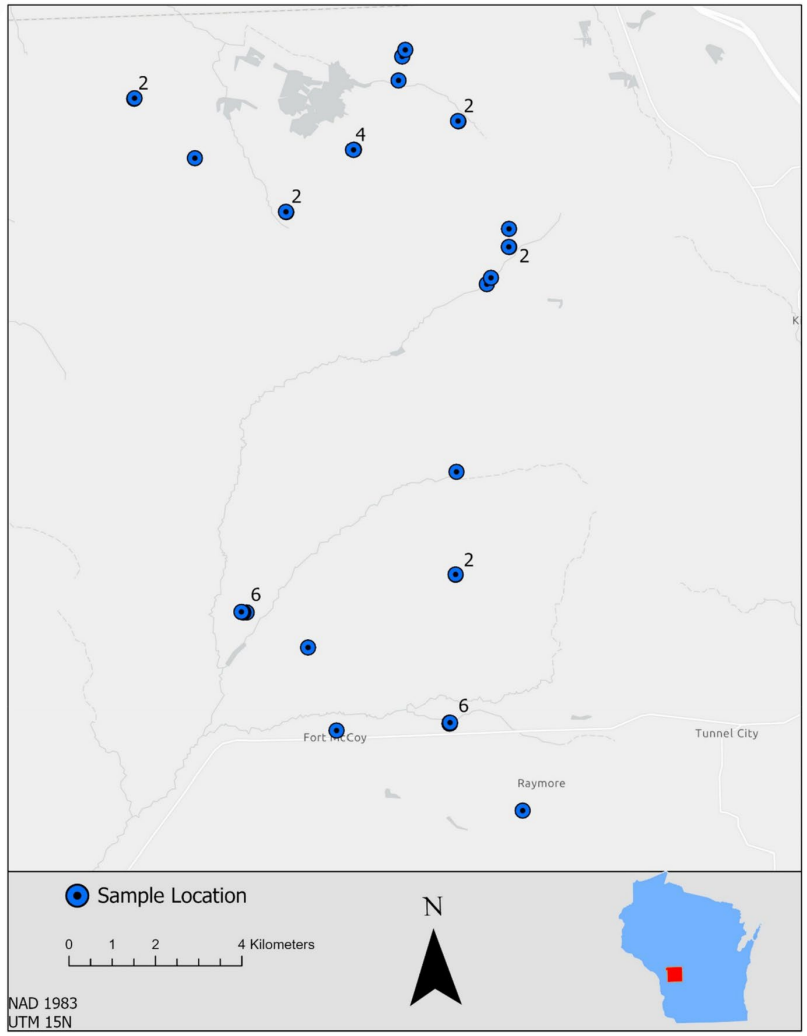


SERDP
DOD • EPA • DOE

ESTCP

eDNA Metabarcoding for Species Interactions

Plant-Pollinator Interactions at Fort McCoy, WI



37 flowers sampled

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eDNA Metabarcoding for Species Interactions

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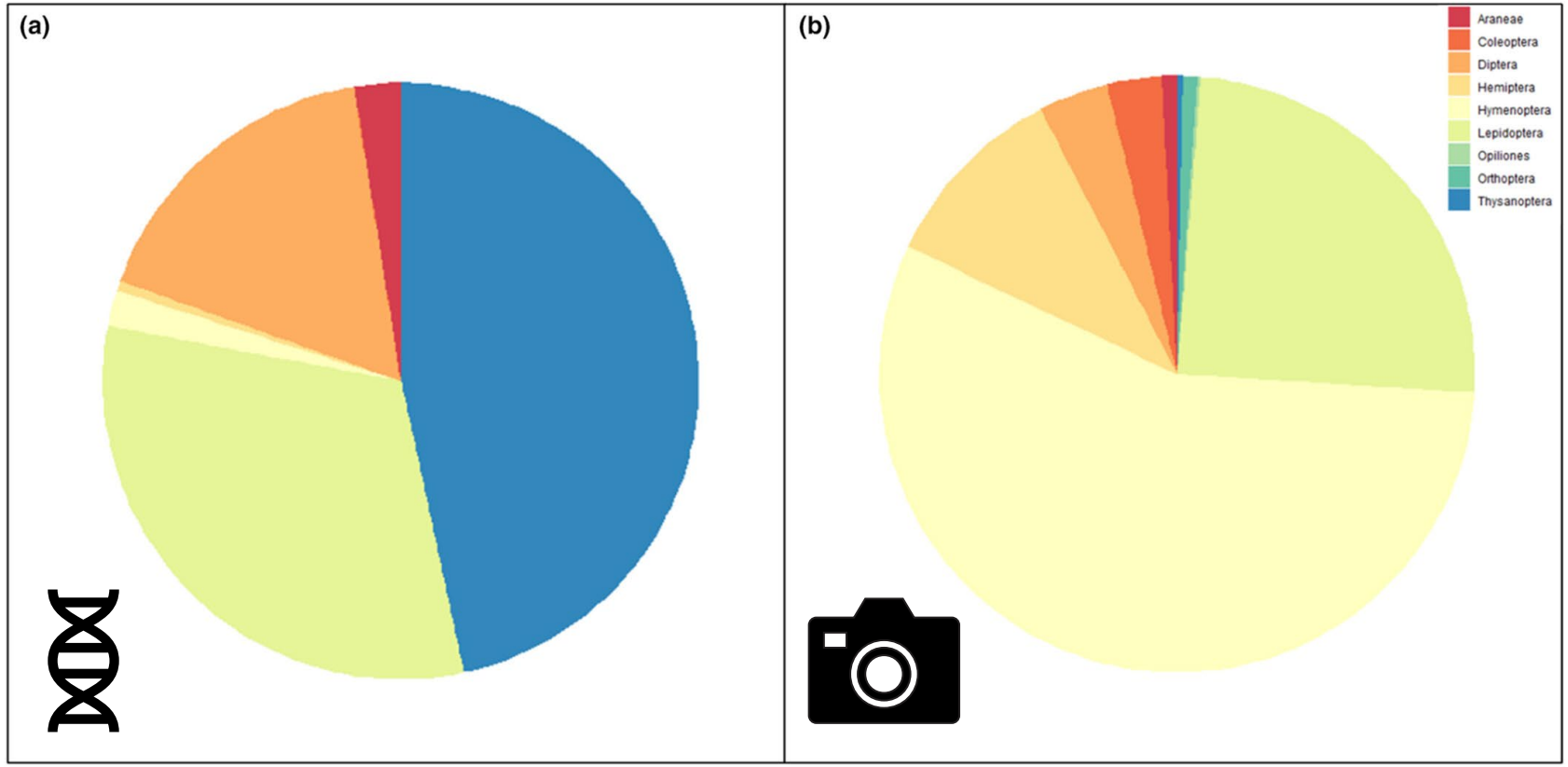
Plant-Pollinator Interactions at Fort McCoy, WI



61,152,771 reads

55 species, 68 genera, 48 families, and 11 orders

1583 individuals

21 species, 27 genera, 37 families, and 10 orders

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eDNA Metabarcoding for Species Interactions

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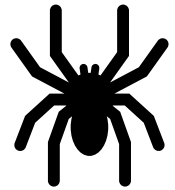

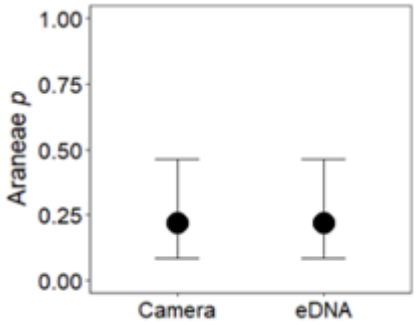

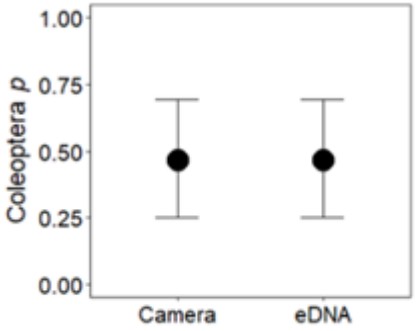

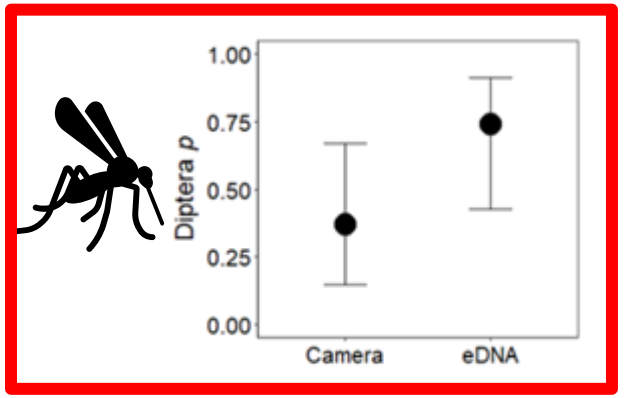

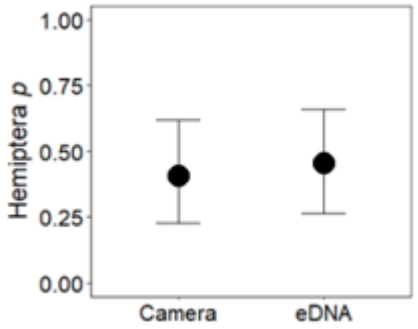

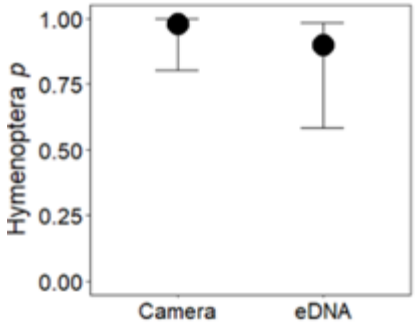

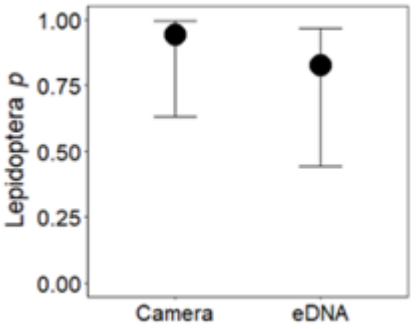

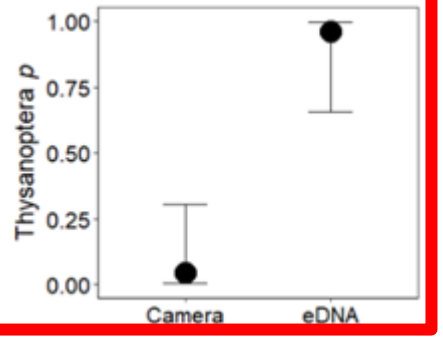

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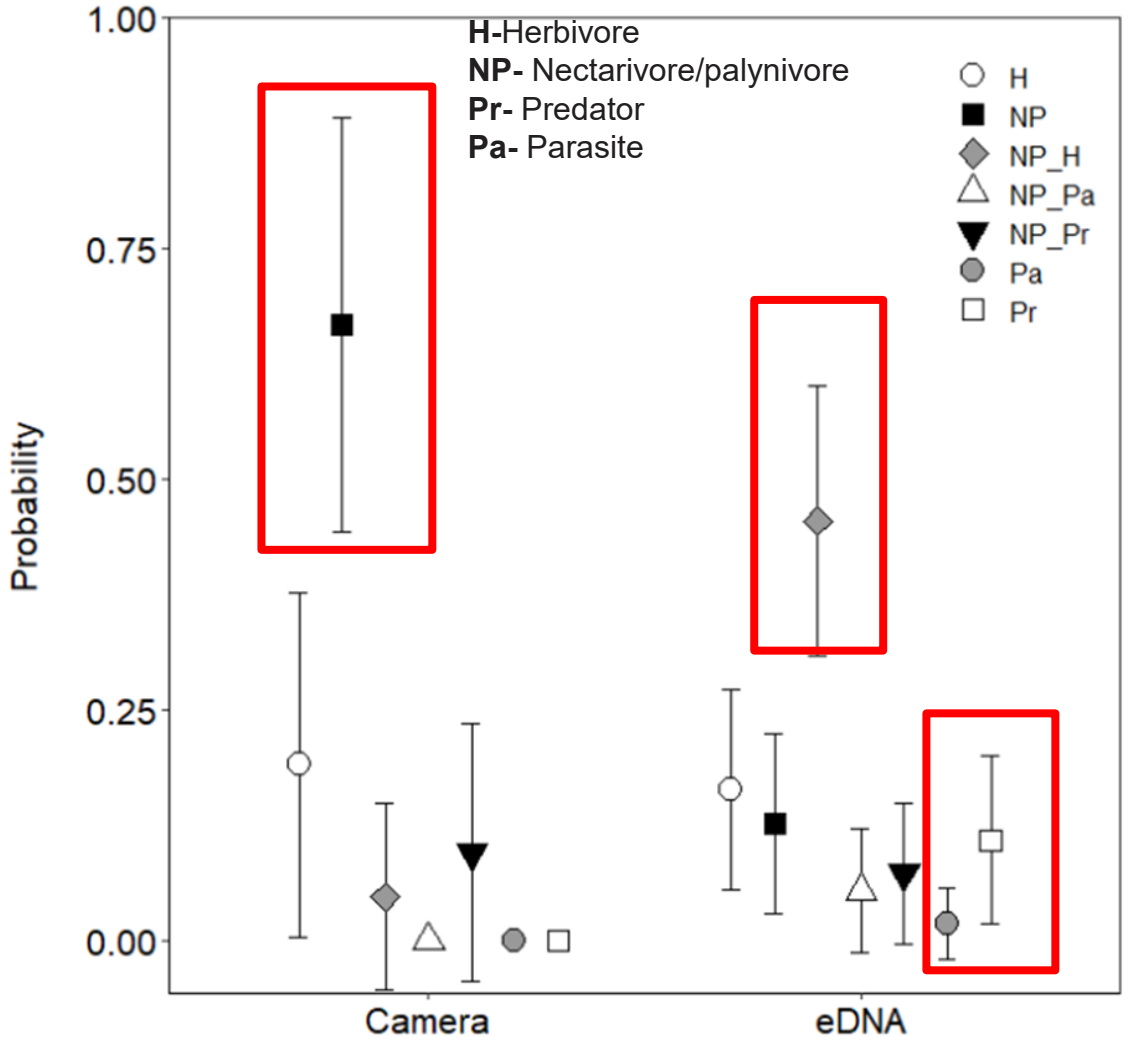

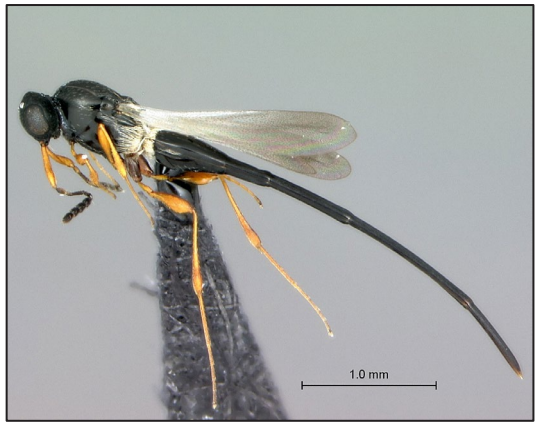

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Plant-Pollinator Interactions on Oahu

O'ahu Army Natural Resources Program

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eDNA Metabarcoding for Species Interactions

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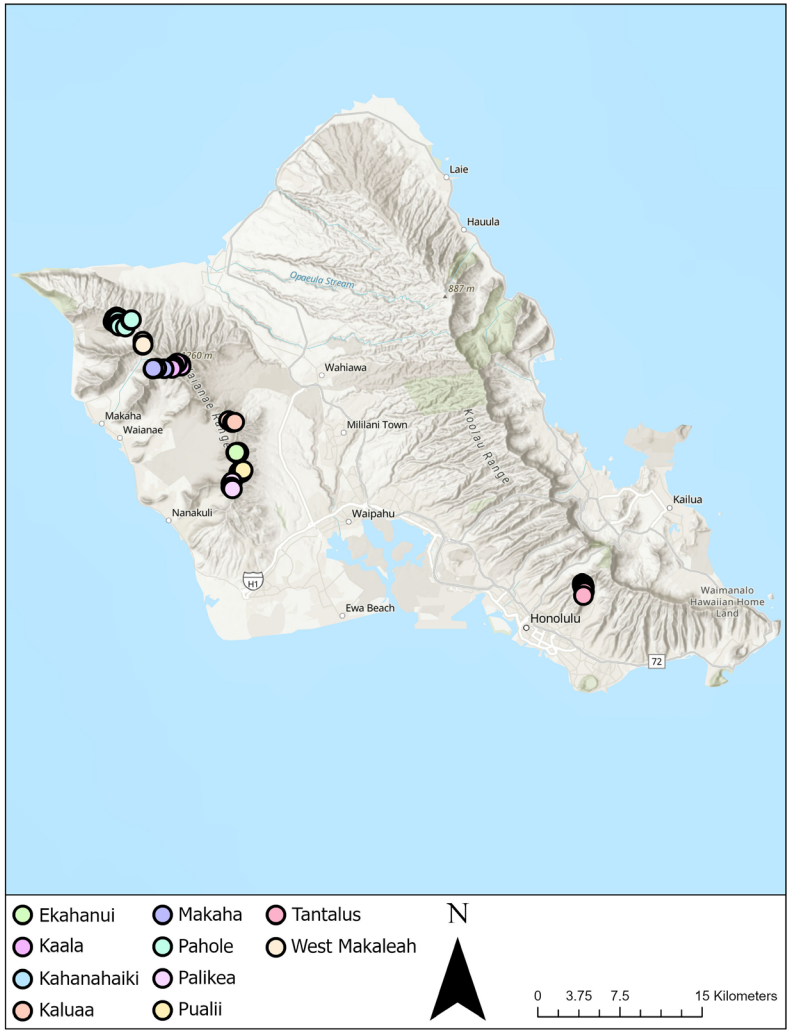
Plant-Pollinator Interactions on Oahu



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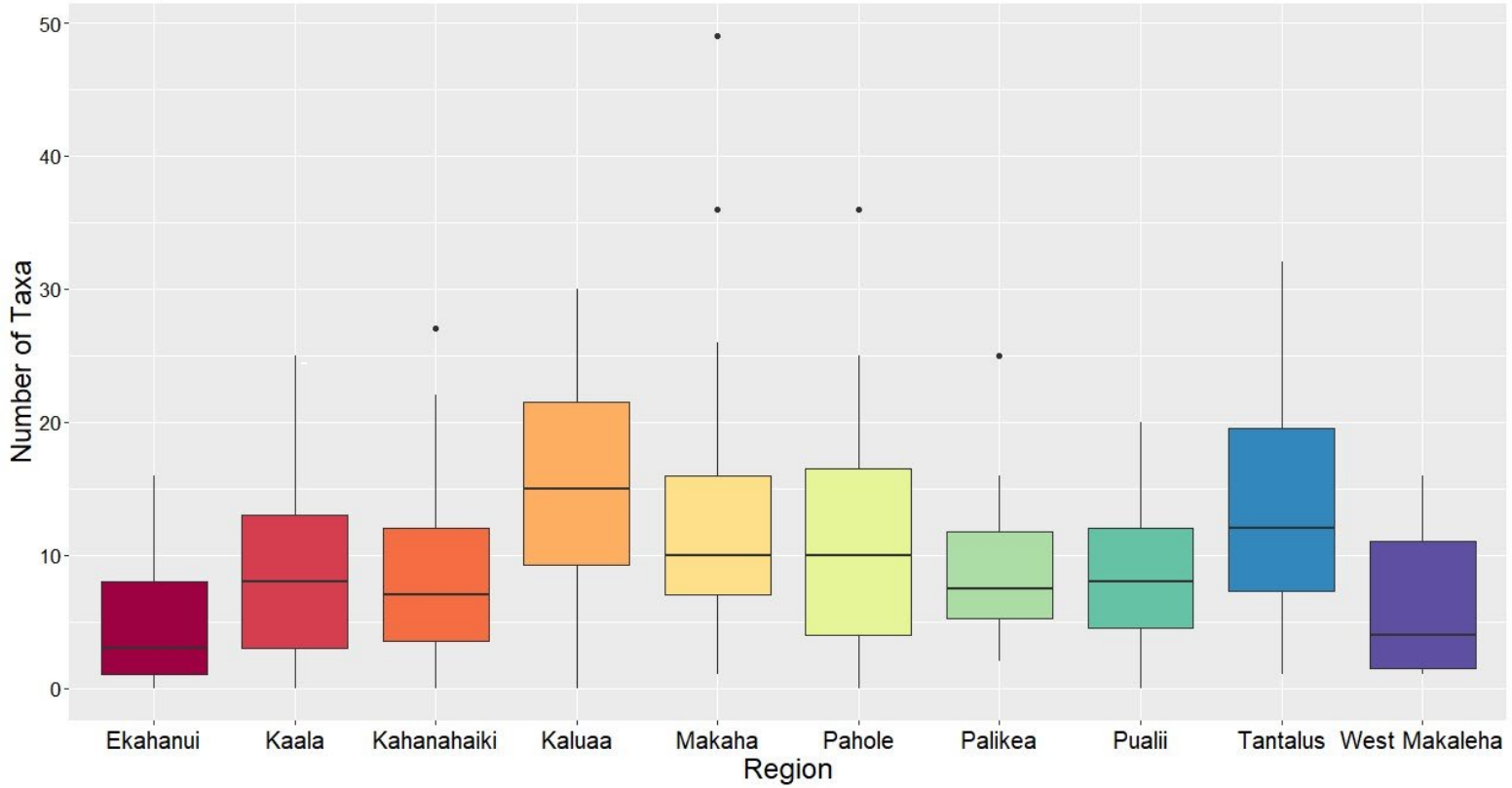
Plant-Pollinator Interactions on Oahu

eDNA:

- 276 samples total across 88 plant species
- 577,091,143 reads (307,689,503 match to family or lower)
- 157 families, 198 genera, and 180 species detected



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eDNA analysis for wildlife monitoring

Primary benefits

- Increase survey coverage and frequency
- Inform follow-up survey efforts, saving time and resources
- Detect rare/elusive species
- Rapidly assess entire communities
- Infer potential species interactions
- Can complements existing conventional approaches

Important considerations

- May require substantial R&D, bioinformatics, computing resources
- Always requires reference sequence data
- Difficult (or impossible) to ascertain abundance, health, population demographics
- Depends on molecular lab facilities
- Sampling bias is present, but different from conventional methods.
- eDNA presence/absence is not the same as species presence or absence

Detailed Guidance Document Available
Jinelle.Sperry@usace.army.mil

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QUESTIONS??