

# Identifying Bats via eDNA in Water

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# Can we develop a method to identify bats from environmental DNA in water sources?

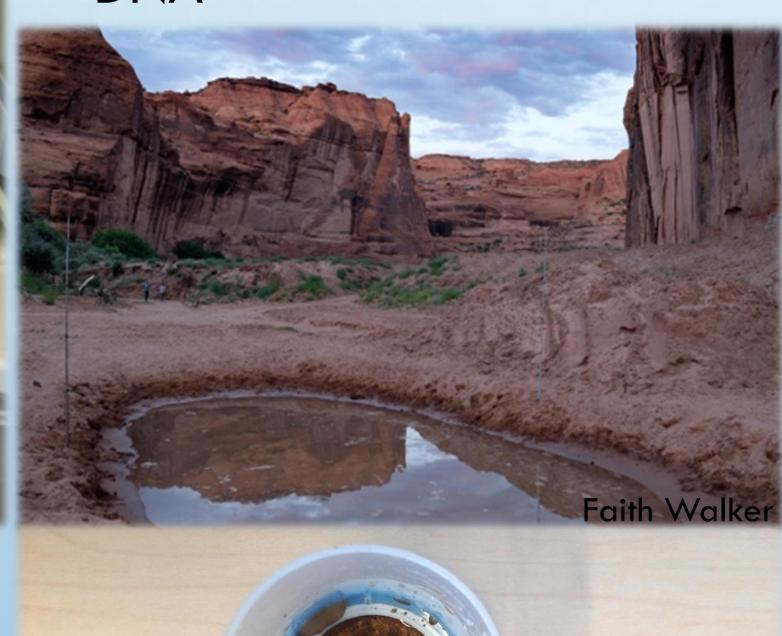
### Development

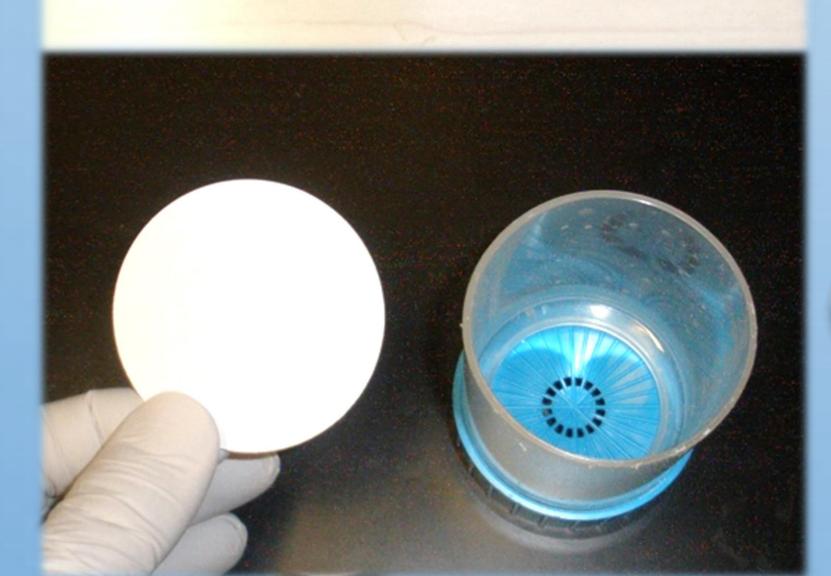


No bats, but bat DNA

Troubleshooting and Lab Testing Problem: Turbid water

clogging filter Solution: Used two filter sizes; large pore first to remove most of the debris and small pore second to capture remaining cells and DNA





Question: Can we detect trace amounts of bat DNA?

Tests: 2 dilution tests:

- 1) Small amounts of guano were diluted in DI water (lowest proportion = 0.000628g/L) 2) Individual buccal swabs from
- known bat species were placed in 1 gallon of DI water.



## Dilution Test Results

All bat species were correctly identified at all dilutions.

They were:

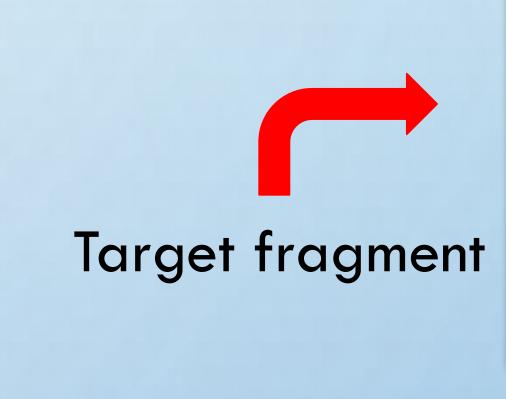
Eptesicus fuscus, Big brown bat

Myotis volans, Long-legged myotis

Myotis occultus, Arizona myotis

Species-level identification was not achieved for water from a mine that was subjected to next-generation amplicon sequencing or from water from a cattle tank that was Sanger sequenced, likely because of DNA degradation and nonoptimal sampling times.

Field Results and Future Directions



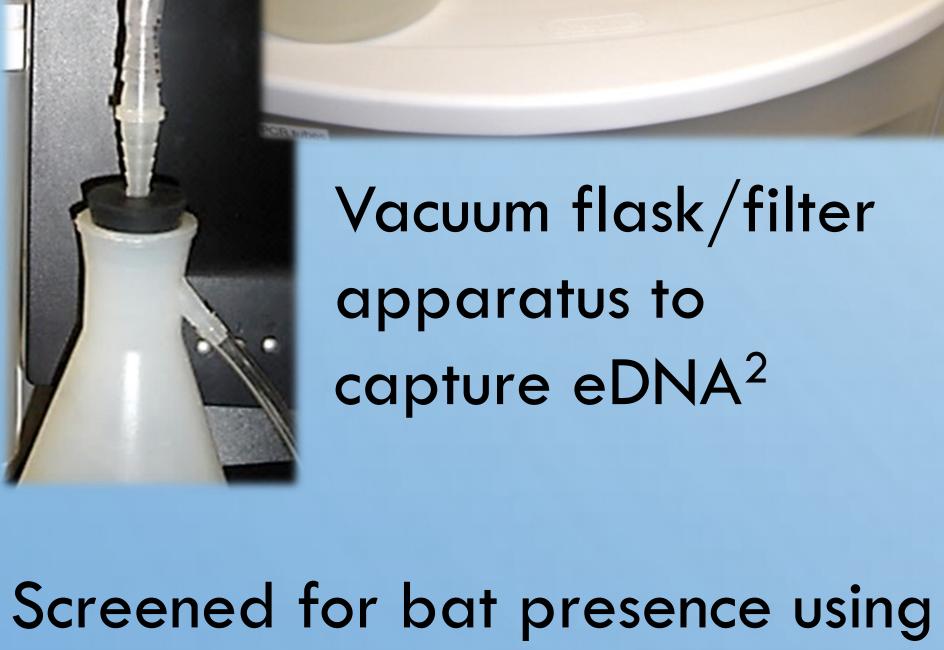


Next: 1. Apply and test non-filter based approaches such as centrifugation<sup>3</sup> to capture bat eDNA.; 2) Apply nextgeneration amplicon sequencing to recentlycollected water that was sampled during high bat activity.

#### References:

- 1) Walker FM, Williamson CHD, Sanchez DE, Sobek CJ, Chambers CL (2016) Species From Feces: Order-Wide Identification of Chiroptera From Guano and Other Non-Invasive Genetic Samples. PLoS ONE 11(9): e0162342. doi:10.1371/journal.pone.0162342
- 2) Laramie, M.B., Pilliod, D.S., Goldberg, C.S., and Strickler, K.M., 2015, Environmental DNA sampling protocol—Filtering water to capture DNA from aquatic organisms: U.S. Geological Survey Techniques and Methods, book 2, chap. A13, 15 p., http://dx.doi.org/10.3133/tm2A13.
- 3) Williams K.E., Huyvaert K.P., Piaggio A.J., 2016, No filters, no fridges: a method for preservation of water samples for eDNA analysis: BMC Research Notes. DOI: 10.1186/s13104-016-2104-5

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the Species From Feces DNA minibarcode assay<sup>1</sup> via Sanger and next-generation amplicon sequencing