

APR 05, 2023

# OPEN ACCESS

dx.doi.org/10.17504/protocol s.io.x54v9d5yzg3e/v1

**Protocol Citation:** Grayson Huston 2023. [Modified] Lake ABPS Protocol - University of Maine. protocols.io

https://dx.doi.org/10.17504/p rotocols.io.x54v9d5yzg3e/v1

License: This is an open access protocol distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working We use this protocol and it's working

Created: Mar 09, 2023

Last Modified: Apr 05, 2023

**PROTOCOL** integer ID:

78446

**Keywords:** Sedimentary DNA, SedDNA, Fish

# ( Modified Lake ABPS Protocol - University of Maine

## Grayson Huston<sup>1</sup>

<sup>1</sup>Maine Center for Genetics in the Environment, University of Maine



**Grayson Huston** 

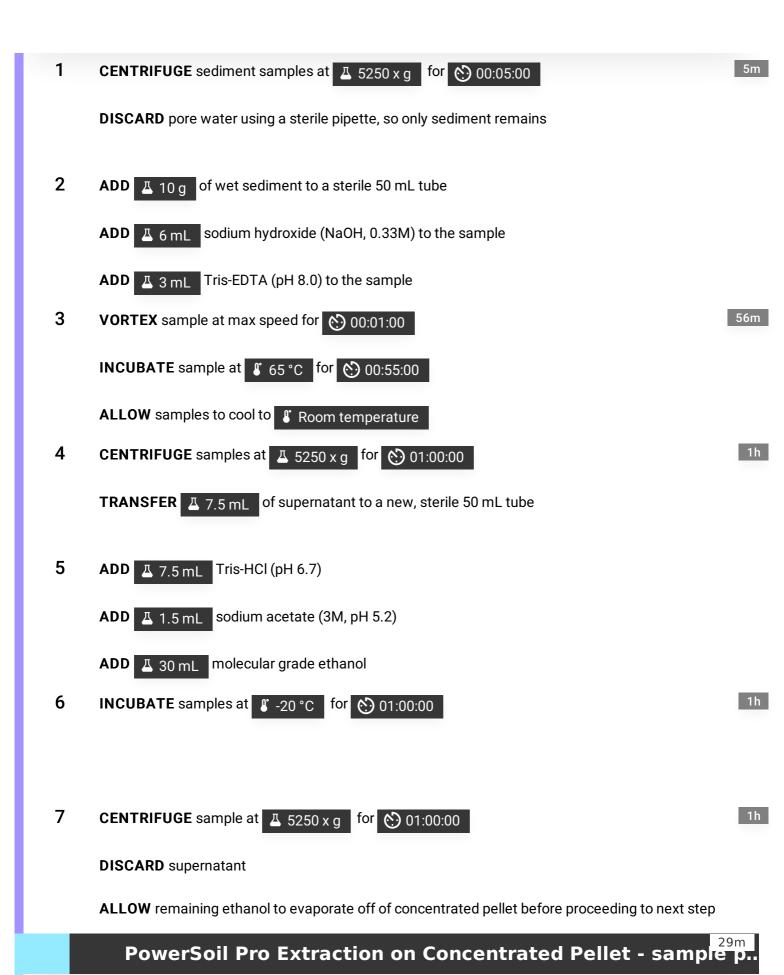
**ABSTRACT** 

A modified version of the Lake ABPS protocol as described in Thomson-Laing et al. 2022

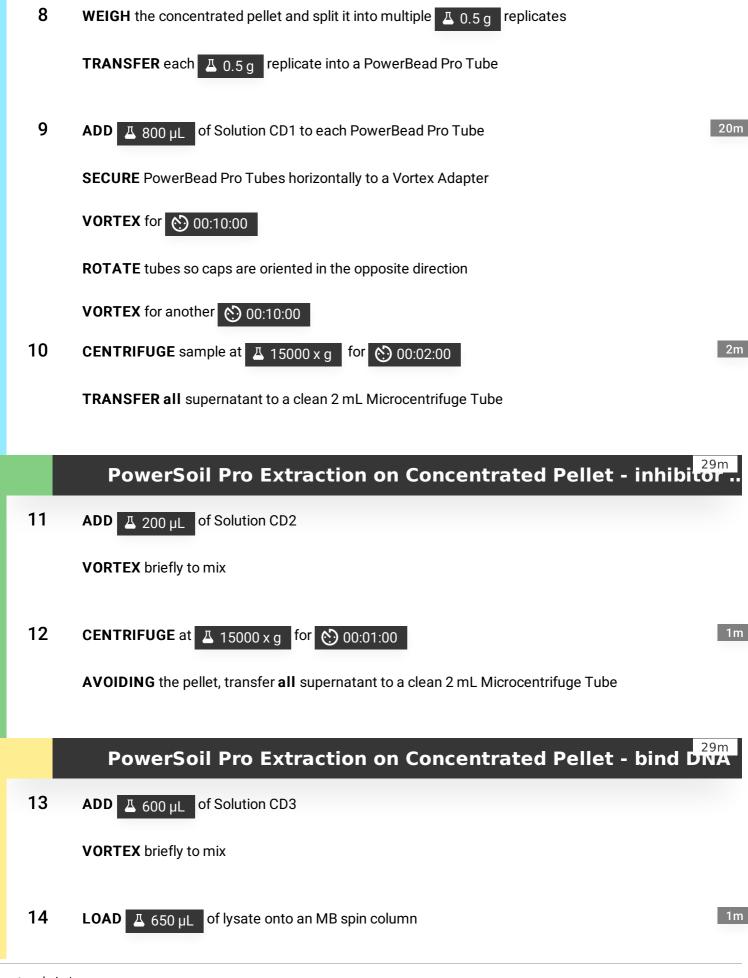
Protocol successful at detecting fish sedDNA collected from lake surface sediments, as well as river sediments during an anadromous fish sea-run migration

**Alkaline Lysis & Ethanol Precipitation** 

4h 1m



protocols.io | https://dx.doi.org/10.17504/protocols.io.x54v9d5yzg3e/v1



**CENTRIFUGE** at <u>■ 15000 x g</u> for 00:01:00

**DISCARD** the liquid flow-through

15 REPEAT step 14 to ensure all the lysate has passed through the MB Spin Column

CAREFULLY place the MB spin column into a clean 2mL collection tube

## PowerSoil Pro Extraction on Concentrated Pellet - wash spi

16 ADD 4 500 µL of Solution EA to the MB spin column

**CENTRIFUGE** at <u>■ 15000 x g</u> for 00:01:00

**DISCARD** the liquid flow-through and place the MB spin column into the same 2 mL Collection Tube

17 ADD A 500 uL of Solution C5 to the MB spin column

**CENTRIFUGE** at <u>■ 15000 x g</u> for 00:01:00

DISCARD the liquid flow-through and place the MB Spin Column into a new 2 mL Collection Tube

18 **CENTRIFUGE** at <u>■ 16000 x g</u> for 00:02:00

2m

CAREFULLY place the MB spin column into a new 2mL Collection Tube

19 A 50-100 uL of Solution C6 to the center of the white membrane in the MB Spin Column

#### Note

Adjust the amount of Solution C6 added to each replicate so that the final volume, once all replicates are pooled together (step 21), totals 200ul

For example, if at Step 8 sample A weighed 1.0g and was split into two 0.5g replicates: A1 and A2. At this step (Step 19), A1 and A2 would each receive 100ul Solution C6, so that when they are pooled together, their total volume is 200ul

If sample A was split into three 0.5g replicates (A1, A2, and A3), each would receive approximately 66ul of Solution C6

# 20 CENTRIFUGE at Δ 15000 x g for ○ 00:01:00

**DISCARD** the MB Spin Column

POOL all replicates into a sterile 1.5 mL Microcentrifuge Tube

### DNA is now ready for downstream applications

Note

For best results in qPCR, use  $\sim$   $\_$  6  $\mu$ L of extracted DNA template per PCR reaction