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Soil pH measurements (adapted from NEON protocols)

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We use this protocol and it's working

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Abstract

This protocol is designed to collect metadata about soil pH using soil collected for metadata (NOT the sample reserved for metagenomic sequencing) during the sampling protocol. Students will suspend a small amount of the soil sample in water, allow it to settle, measure pH in water, then add a Calcium Chloride solution and repeat the measurement. This will generate metadata that should be included with soil microbiome data to describe soil conditions, and adapts the National Ecological Observatory Network's best practices intended for student use.

Materials

- pH meter and calibration solutions
- kimwipes and rinse solution/distilled water
- balance
- serological pipets and pipet-aid and/or graduated cylinder
- Glass stir rod
- Scoopula or clean spoons
- 50 ml conical tube for each sample
- tube rack for 50 ml conical tubes
- 100 ml glass beaker (may or may not be necessary)
- distilled or deionized water
- 0.02 M CaCl_2 solution, pH between 5 and 6.5 (if adjusting the pH is necessary, use 1N HCl or 6N Ca(OH)_2)



Safety warnings

! Handle glass stir bars and (if relevant) pH probes gently to prevent breakage.

- 1 Wearing gloves, place a 50 ml conical tube in a foam rack onto the balance, and tare the balance.
- 2 If you have an M horizon soil, measure 10 g into a fresh 50 ml conical tube. If you have an O horizon soil, measure 5 g of soil into a fresh 50 ml conical tube using a clean scoopula.
- 3 Record soil mass on your logsheet  Soil sampling log sheet.docx 15.9KB .
- 4 Add 10 ml of diH₂O to M-horizon samples, or 20 ml of diH₂O to O-horizon samples. Record water volume in your log.
- 5 Allow soil to absorb water by thoroughly mixing with a clean glass stir rod. (Clean stir rod with a kimwipe and DI water when you are done).
- 6 Allow to settle. If there is not a 1 cm layer of water on top of the settled soil (there may be a floating layer over it especially with O-horizon samples) then add another 10 ml aliquot of water. Add water in 10 ml aliquots until the soil is saturated and there is enough standing water on top for submerging the pH meter, recording the total volume of water added in your log. (note: if you have an O-horizon sample and need to add a lot of water, you may need to transfer the soil and water to a clean glass beaker).
- 7 Let sample sit for 30 minutes with the cap off. Stir with the stir rod, and let sit for 30 minutes again with the cap off.
- 8 Meanwhile, turn on and calibrate the pH meter. Follow directions using buffers 4, 7, and 10.
- 9 Stir the sample before measuring, let sit for 1 minute, then measure by pushing the probe in just deep enough. Recording your reading as soil+water pH.

Hints from NEON: How deep to insert the probe tip depends on the soil horizon type:

*For M horizon samples, measure in the top 1/3 of the slurry, e.g., only submerge probe tip ~ 1.5 cm. This more dilute part of the slurry will avoid most settled particles.

*For O horizons, submerge probe tip into the solution below the layer of floating organic material that will be present. It is OK if some flocculated soil is floating in this region.

- 10 Add an equal volume of 0.2 M CaCl₂ as the total volume of water you added. (example: if you added 20 ml of water, add 20 ml of CaCl₂). If there isn't enough room in the tube, transfer to a beaker first.



- 11 Record the amount of CaCl_2 added into your log.
- 12 10. Stir with a stir rod for 20 seconds, then let the sample sit for 1 minute.
- 13 11. Immediately measure the pH and record measurement as Soil+water+ CaCl_2 pH.
- 14 Discard the sample according to local regulations and procedures. You may need to let the samples settle overnight to discard the liquids and solids separately.

Protocol references

Stanish, L. (2024). TOS Protocol and Procedure: SLS – Soil Biogeochemical and Microbial Sampling. NEON.DOC.014048 version P. NEON (National Ecological Observatory Network).