B.C. SOIL HEALTH ASSESSMENT LABORATORY PROCEDURES

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Recommended laboratory procedures for measuring soil health indicators in British Columbia.

Sustainable Canadian Agricultural Partnership
British Columbia | cleanBC
Canada

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Mineralizable Nitrogen

Nitrogen is the main growth-limiting nutrient in many agricultural soils. The largest pool of nitrogen in soil is found in organic matter. However, before plants can take up nitrogen, it must be converted from its organic form to an inorganic form. This process, known as nitrogen mineralization, is a key part of soil fertility.

Potentially mineralizable nitrogen (PMN) measures the “active” fraction of soil organic nitrogen that is primarily responsible for releasing mineral nitrogen through microbial activity (Curtin and Campbell, 2007). Microbial activity depends on environmental factors such as moisture, temperature, and soil aeration, which interact with vegetation and management practices like tillage and the type and amount of soil amendments applied.

Because of these influences, nitrogen mineralization rates vary across space and time and are difficult to predict. However, good management practices that reduce soil disturbance, maintain living roots and crop residues, and increase the use of organic nutrient sources can enhance a soil’s ability to recycle and supply nitrogen to plants (Liptzin et al., 2023).

Method

The nitrogen mineralization potential of a soil is determined by measuring the amount of ammonium produced when the soil is incubated under controlled conditions for a defined period. Various methods exist for this assessment, differing in incubation time, temperature, and whether the incubation occurs under aerobic (with oxygen) or anaerobic (without oxygen) conditions.

The (anaerobic incubation) method proposed here, adapted slightly from Curtin and Campbell (2007), was selected for its practicality and operational efficiency. It involves incubating the soil at 40°C for one week. While nitrogen mineralization can theoretically continue over an extended period, most of the measurable mineralization occurs within the first few days once optimal conditions are established.

Material

- Centrifuge
- Cabinet/incubator capable of maintaining temperatures of up to 40°C (and humidity near 100% during incubation or air tight caps to prevent moisture loss during incubation).
- 2.5 M KCl
- Filter paper (Whatman No 42)

Procedure

1. Weigh 2.5 g of sieved (< 2 mm) soil into a 25 mL plastic, screw-cap centrifuge tube. Add 5 mL of distilled water to submerge the soil, stopper the tube, and place in a constant temperature (40°C) cabinet/incubator for 7 days.
2. Remove tube from incubator and add 20 mL of 2.5 *M* KCl (after dilution with water in the sample, final KCl concentration is 2 *M*). Mix contents of tube, centrifuge at 1900 *g*, for 60 minutes and filter the supernatant (prewashed Whatman No. 42).
3. Determine NH_4-N in the supernatant. Measure the amount of NH_4-N in the soils before incubation by extracting a separate sample with KCl. Mineralized N is estimated by deducting this preincubation NH4-N value from the amount measured in the incubated sample.

Comments

- Because most arable soils contain little or no ammonium nitrogen (NH_4–N), the initial NH_4–N measurement may be omitted; however, preliminary checks should be conducted to confirm that native NH_4–N levels are negligible.
- Although anaerobically mineralizable nitrogen (AMN) is correlated with nitrogen mineralized under aerobic incubation, this relationship is often weak or inconsistent.
- This test is not intended for use in fertilizer nitrogen recommendations.
- Different amounts of soil may be used, but it is critical to maintain a 1:10 soil-to-extractant ratio. For example, using 5 g of sieved soil requires 10 mL of distilled water and 40 mL of 2.5 M KCl; in this case, a 50 mL centrifuge tube is appropriate.

Reference

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Carbon Mineralization Potential

Carbon mineralization refers to the conversion of organic carbon into inorganic carbon, which under mostly aerobic conditions occurs primarily as carbon dioxide (CO_2). In soils, this process takes place during the decomposition of organic matter as a result of microbial activity.

The metabolic activity of the soil microbial community responds strongly to management practices such as tillage, fertilization, and crop rotation. However, it is also influenced by soil–climate interactions and other environmental factors. As a result, carbon mineralization is a highly variable soil health indicator—both seasonally and spatially—and can show inconsistent patterns (Liptzin et al., 2022; Hargreaves et al., 2019).

Despite this variability, assessing the carbon mineralization potential provides a biological measure of the active fraction of soil carbon, in contrast to purely chemical fractionation methods. Nevertheless, results must always be interpreted in conjunction with other soil health indicators and within the context of site-specific environmental and management conditions. They should not be used to compare soils across different fields, farms, or regions.

Method

When air-dried soil is rewetted, it triggers a rapid increase in microbial respiration known as the "Birch effect" or "CO_2 flush upon rewetting."

This method measures the amount of carbon dioxide (CO_2) released within 24 hours after rewetting air-dried, sieved soil. CO_2 production can be quantified using different techniques, such as an alkaline trap or gas analysis following a 24-hour incubation. Results are expressed as milligrams of CO_2–C per kilogram of dry soil per 24 hours.

The procedure is adapted from the Cornell Soil Health Laboratory Protocol (2022) with minor modifications. The description below provides a brief overview; detailed instructions, including material preparation and safety guidelines, are available in the full Cornell Soil Health Laboratory Procedure (2022).

Material

- 2-mm (10 mesh) sieve and 8-mm sieve (2.5 mesh)
- Air-drying facility (minimum temperature: 30 °C)
- Coarse-grinding apparatus (e.g., rolling pin, flail mill)
- Analytical balance (±0.1 g sensitivity)
- Perforated aluminum weigh boat (see reference)

- Filter paper (55 mm)
- Weighing paper

Half pint mason jar (1 per sample)

- Incubator (capable of 20°C)
- 10-ml pipettors and tips
- Double-deionized (dd) water or, better, ultra-pure water (18 μΩ)
- EC Meter
- Trap Assembly (which is a 10-ml beaker stuck to plastic tripod 'pizza' stools (Cornell Soil Health Laboratory Procedure, 2022) See picture 1.
- 0.5 M KOH

Picture 1 CO_2 trap assembly

Procedure

1. Transfer the sample to a paper bag or spread out on a tray before drying. Dry the soil sample at temperatures below 30°C to constant weight at ambient temperature. Break up soil and pass through an 8-mm sieve before drying if necessary. Forced air circulation is recommended to speed up the drying process.
2. Pass air-dried sample through a 2-mm sieve. When working with large samples (e.g., composite samples), a subsample may be taken prior to sieving using a sample splitter or by spreading the soil sample across a flat surface, dividing the soil into sections (e.g., a rectangular grid or pie), and collecting all material within multiple non-adjacent sections. Visually verify that the proportions of large to small aggregates/particles are similar for the subsample and the bulk soil sample, as soil aggregates will separate by size during sample handling.
3. Place two filter papers to the bottom of the jar, offset from each other.
4. Weigh 20g of dried and sieved soil into the weigh boat. Since the weigh boat is perforated, place some weigh paper on scales pan to catch soil that falls through the perforations.
5. Gently place weigh boat onto the filter paper in the jar. Any soil that was caught on the weighing paper can be added now.
6. Add 9 ml 0.5 KOH to the trap beaker in the jar.
7. Dispense 7.5 mL of dd water inside the glass jar, but not inside the beaker.
8. Seal the jar tightly with a compatible lid.

9. Place the jar into an undisturbed location with room temperature > 20°C, ideally an incubator (set at 20°C) for 24 hours. Note the time when the samples were placed in the room or incubator.
10. After 24 hours (± 1 hour), remove the jar from the incubator and open the jar.
11. Place EC probe into the trap beaker giving a brief gentle stir.
12. Let reading stabilize and record EC.
13. Discard soil and clean material. See Cornell Soil Health Laboratory Procedure (2022) for a detailed description of a safe disposal of soil and chemicals and cleaning of materials.

CO_2 Measurement

Two primary approaches are commonly used to quantify CO_2 produced during incubation. The method outlined in this protocol relies on an alkaline trap—potassium hydroxide—to capture CO_2 within the sealed jar. The absorbed CO_2 is then quantified by measuring changes in electrical conductivity.

The alternative approach uses analytical instruments capable of detecting CO_2 concentrations, such as a gas chromatograph or an infrared gas analyzer. In this case, the change in CO_2 concentration before and after incubation is measured and converted to milligrams of carbon using the ideal gas law. This method requires a jar lid that accommodates gas sampling by syringe or direct connection to the measuring instrument.

Although both methods produce highly correlated results, switching between them within the same study is not recommended.

Report results as CO_2-C mg per kg soil.

Calculations

This calculation applies to the proposed method.

Pure 0.5 M KOH solution has an electrical conductivity of 112 µS cm^{-1}. As it absorbs CO_2, it is gradually converted to 0.25 M K_2CO_3. When fully saturated, 9 mL of 0.5 M KOH can absorb approximately 99 mg of CO_2. A pure 0.25 M K_2CO_3 solution has an electrical conductivity of 42 µS cm^{-1}.

These conductivity values make it possible to calculate how much CO_2 has been trapped in the KOH solution using the following relationship:

$$\frac{EC_{raw}-EC_{sample}}{EC_{raw}-EC_{sat}} = f$$

where
f = fraction of maximum CO_2 that can be absorbed
EC_{raw} = electrical conductivity of pure 0.5 M KOH ("blanks")
EC_{sample} = electrical conductivity of the trap associated with the particular sample
EC_{sat}= electrical conductivity of fully saturated 0.5 M KOH

Example

- Initial solution: 0.5 M KOH, electrical conductivity (EC) = 112 mS cm^{-1}
- Fully saturated solution (all KOH converted to K_2CO_3): 0.25 M K_2CO_3, EC = 42 mS cm^{-1}
- Maximum CO_2 capacity of 9 mL of 0.5 M KOH = 99 mg CO_2
- Assume that the EC of the solution changes linearly between these two endpoints as CO_2 is absorbed
- Suppose that, after incubation, the measured EC of the KOH solution is 90 mS cm^{-1}

Calculation

Step 1: Calculate the fraction of maximum CO_2 absorbed

$$\text{Fraction of capacity (f)} = \frac{EC_{raw}-EC_{sample}}{EC_{raw}-EC_{saturated}} \quad \longrightarrow \quad f = \frac{112-90}{112-42} = \frac{22}{70} \approx 0.314$$

→ the solution has absorbed about 31.4% of its maximum CO_2 capacity.

Step 2: Convert this fraction to mg of CO_2

$$CO_2 \text{ trapped (mg)} = f \times \text{maximum } CO_2 \text{ capacity}$$

$$CO_2 \text{ trapped} = 0.314 \times 99 \text{ mg} \approx 31.1 \text{ mg } CO_2$$

Step 3: Convert to CO2-C mg per kg of (dried) soil

- 1 mg CO_2 = 0.273 mg of CO_2-C
- Sample size is 20 mg -> 1 kg of dried soil = 50 x 20 mg. Thus,

$$31.1 \text{ mg} \times 0.273 \text{ mg } CO_2\text{-C mg mg}^{-1} CO_2 \times 50 \approx 425 \text{ mg } CO_2\text{-C kg}^{-1} \text{ soil}$$

QA/QC

Accept when within Quality control sample value ±2 standard deviations

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Soil Organic Matter

Soil organic matter (SOM) includes all materials derived from living organisms, whether alive, recently dead, or in advanced stages of decomposition. It plays a vital role in soil function at many levels: supplying and storing plant nutrients and water, improving soil structure to enhance water infiltration, supporting biodiversity that helps suppress pathogens, and contributing to long-term carbon sequestration.

Increasing and maintaining soil organic matter has long been a central focus of soil management aimed at improving soil quality and resilience. Practices such as applying organic amendments (e.g., compost), planting cover crops, rotating crops, and reducing tillage can all help build SOM. For many decades, measuring SOM was therefore viewed by some as nearly synonymous with assessing soil health. However, the "ideal" SOM content is site-specific and varies among environments, soil types, management systems, and vegetation (Franzluebbers, 2002). SOM is thus one—albeit important—indicator of soil quality, but it cannot fully represent soil health on its own.

SOM can be measured in several ways, each with strengths and limitations. One commonly used method involves determining the weight loss of a dried soil sample after strong heating, which volatilizes organic components. However, this method can introduce errors in soils containing appreciable calcium carbonate ($CaCO_3$), as carbonates may also decompose during ignition.

Some laboratories instead estimate SOM indirectly by measuring total soil carbon (typically using dry combustion) and subtracting inorganic carbon (often determined after acidification and sparging). The resulting organic carbon value is then multiplied by the "van Bemmelen factor" of 1.724, based on the assumption that dry organic matter contains 58% carbon. Recent work has challenged the universality of this factor, suggesting a wider possible range of 1.4 to 2.5 and a median of 1.9 (Pribyl, 2010). Data from the B.C. Ministry of Agriculture and Food, based on more than 50 sites across diverse commodities and regions, indicate a factor close to 2 with little variation (Geesing and Lasisi, 2025).

Despite these limitations, soil organic matter remains a valuable and historically important metric. There continues to be a meaningful role for measuring SOM directly—at least for the foreseeable future—provided its constraints are understood and complemented with additional indicators of soil quality

Method

There is no universally accepted standard method for Loss-on-Ignition (LOI), particularly with respect to ignition temperature, which can range from 360 °C to 600 °C. However, temperatures between 375 °C and 400 °C are most commonly used, and 375 °C is

recommended for this protocol. The procedure below is adapted from Kalra and Maynard (1991).

Material

- Muffle furnace
- Porcelain crucibles
- Desiccator

Procedure

1. Oven-dry about 10 g of sieved soil (2-mm) soil at 105°C for 4 hrs.
2. Heat porcelain crucibles for one hour at 375°C.
3. Cool in open to about 150°C. Place in a desiccator, cool for 30 minutes, and weigh.
4. Weigh about 5 g of the oven dried sample (0.001 g accuracy) into each crucible.
5. Place crucibles containing samples in muffle furnace at room temperature. Heat slowly (increase temperature about 5°C minute·') to 375° ± 5°C.
6. 375°C ±5°C overnight (16 hours).
7. Tum furnace off and let temperature drop to about 150°C.
8. Remove crucibles and place in desiccator for 30 minutes. Weigh to the nearest milligram.

Calculation

$$\text{Loss on ignition (approximate organic matter (\%)} = \frac{\text{oven dried sample (g)-LOI sample (g)}}{\text{oven dried sample}} \text{ x } 100$$

Comments

1. This method should be indicated as loss-on-ignition (LOI). It gives an estimate of organic matter sufficiently accurate for most descriptive purposes.
2. The procedure is subject to error as weight loss may include C from carbonates and water and hydroxyl groups from clay. Error is also caused by combustion of inert C compounds and volatilization of substances other than organic material.
3. There is incomplete oxidation of carbonaceous materials in some soils at 375°C.

4. The method is most suitable for well-aerated samples (e.g., sandy and peaty soils) with low clay mineral and inert carbon (charcoal) content but it is not suitable for calcareous soils.

References

Franzluebbers, A.J. 2002. Soil organic matter stratification ratio as an indicator of soil quality. Soil&TillageResearch66(2002)95–106.

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Plant Available Water

Water storage is an essential soil function. However, not all water held in the soil is available to plants. Plant-available water is defined by two soil moisture limits. The upper limit is *field capacity*, which represents the amount of water retained in the soil after gravitational drainage—typically several hours after rainfall or irrigation. As plants use water and evaporation occurs, the soil dries and the remaining water is held more tightly. Eventually, the soil reaches a point at which plants can no longer extract water. This lower limit is known as the *permanent wilting point*.

The amount of water held between field capacity and the permanent wilting point is called *plant-available water*. Once this water is depleted, plants wilt and cannot recover. In practice, most plants begin exhibiting water stress well before the plant-available water is fully exhausted, often resulting in reduced growth or yield.

Soil texture is a major factor influencing plant-available water, but sustainable soil management practices—such as increasing organic matter and reducing soil disturbance—can significantly improve a soil's capacity to store and supply water to crops.

Method

Field capacity and permanent wilting point can be measured by applying controlled pressure to soil samples placed on a porous surface (pressure plate). The applied pressure simulates the suction force that plant roots would need to exert to extract water from the soil. The method is adapted from Kalra and Maynard (1991).

The unit for pressure under the International System of Units is *Pascal* (Pa). For the purpose of measuring the pressure needed to extract soil water, the pressure is usually reported as hectopascal (hPa, 100 Pa) or kilopascal (kPa, 1,000 Pa). Other units that are frequently used are bar (1,000 hPa = 100 kPa = 1 bar), or pounds per inch (psi) (1,000 hPa= 100 kPa = 14.5 psi).

Field capacity (FC) is commonly estimated by measuring the moisture retained at the following pressures:

Texture	Pressure
Coarse-textured soil (sand, sandy loam, loamy sand)	10 kPa
Medium-textured soil (loam, silt loam)	33 kPa
Fine-textured soil (clay loam, silty clay loam, silt clay, sandy clay, clay)	50 kPa

Permanent wilting point (PWP) is commonly estimated by measuring the water content at a pressure of 1,500 kPa, regardless of the texture.

Available water (AW) or **available water capacity (AWC)** is estimated by the difference in soil water content between FC and PWP.

$$AWC\ (\%) = FC\ (\%) - PWP\ (\%)$$

For this protocol, moisture content will be assessed at 10 kPa, 33 kPa, 100 kPa and 500 kPa across all textures. An additional measurement at 1,500 kPa is optional but needs to be clearly communicated with the laboratory conducting the soil moisture retention assessment.

Plant-available water is defined as the amount of water held in the soil between field capacity and a matric potential of 1,500 kPa. In practice, however, crops begin to experience substantial drought stress well before this point. For example, at a matric potential of about 500 kPa, more than 50% of plant-available water has typically been depleted in most soils (USDA, 1997), often resulting in significant moisture stress. Consequently, most irrigation practices aim to prevent depletion of plant-available water beyond 50%, and many crops benefit from maintaining depletion below 30%.

The positive effect of best management practices on plant-available water is driven mainly by increases in water content at field capacity rather than changes in water content at the permanent wilting point. From a practical soil management perspective, the matric potential of 500 kPa is therefore far more relevant than the 1,500 kPa standard. Measuring soil water content at 1,500 kPa primarily increases analytical costs without offering substantial operational value. Nonetheless, when budgets allow, determining water content at 1,500 kPa remains useful for research and scientific comparisons, as it is still widely used in the literature.

Material

- Pressure plate extractor (Soil Moisture Equipment Co., Santa Barbara, California)
- 15-bar pressure plate extractor (=pressure extractor vessel) (Soil Moisture Equipment Co., Santa Barbara, California)
- 1-bar (100 kPa) ceramic plates
- 15-bar (1,500 kPa) ceramic plates
- Pressure plate rings (usually about 5-cm diameter, 1-2 cm height where 1-cm height rings are used for 1-bar ceramic plates, and 2-cm heigh rings for 15-bar rings)
- Compressed gas (N_2) cylinder with regulator, manifold, and gauge (0-2 MPa) (Pressure regulator must be capable of controlling in the range 0.1-1.6 MPa)
- Triangular supports for extractor
- Plate spacers (plastic)
- Burette
- Scales
- Knife
- Zip-ties
- Large beaker or bucket
- Porous filter paper (for example Whatman Grade 4)
- Drying oven

- Disposable aluminum dishes or soil-moisture cans
- Wax paper or plastic sheet
- Syringe or disposable pipette or siphon
- Desiccator

Procedure

Preparation

1. Soak the ceramic plates in water for 24 hours to fully saturate them.
2. Place the soaked plates on the workbench.

Preparing Soil Samples

3. Insert a pressure plate ring into the soil inside the soil-sampling ring. Soil should be moist—not wet.
4. Remove the ring with soil by cutting around it and gently lifting it from below with a knife.
5. If soil sticks out below the ring, trim the bottom so it is level with the rim.
6. Place coarse filter paper on the bottom of the ring and secure it with a zip tie to prevent soil loss (see picture 2).
7. Label the rings and place them onto the ceramic plate.

Picture 2: Pressure plate ring with filter paper secured at the bottom (*Picture credit: Reza Habibiandehkordi*)

Pre-Wetting the Samples

8. Add water onto the plate between the rings so the samples are wetted from below. Water should reach at least half the height of the rings.
9. Cover the samples with wax paper or plastic.
10. Let them stand overnight to fully saturate.
11. The next morning, remove extra water on the plate (using a syringe, pipette, or siphon).

Running the Pressure Extractor (≤ 100 kPa)

12. Place the triangular support at the bottom of the pressure extractor.

13. Install the plate with samples in the lowest position, then add the middle and top plates with spacers.
14. Connect the outflow tubes to the burette, and place a beaker or bucket under the burette (the burette will likely overflow initially).
15. Close and tighten the extractor evenly. Make sure the O-ring is seated correctly.
16. Apply pressure slowly (up to, but not exceeding, 100 kPa).
    - Keep pressure constant
    - When water flow has nearly stopped (usually 18–20 hours, but sometimes up to 48 hours), close the burette and wait until flow fully stops
17. Disconnect the outflow tubes.
18. Release pressure from the extractor.
19. Open the extractor.
20. Quickly transfer the soil from the ring into a pre-weighed dish using a wide spatula.
21. Weigh the wet soil immediately (to 0.01 g accuracy).
22. Return the rings to the plate and add water again before continuing to the next pressure step.
23. Repeat steps 12–22 for each pressure level up to the maximum below 100 kPa.

For Pressures Above 100 kPa (Using 15-Bar Plates)

24. Move the soil rings to 15-bar plates.

- *Note: Soak the 15-bar plates the day before, the same way as Step 1.*

25. Repeat steps 12–22 using the 1,500-kPa extractor, applying pressure above 100 kPa.
26. Keep samples under pressure until all flow stops, which may take several days or weeks.
    - A stable burette reading means equilibrium has been reached.
    - Disconnect the burette to prevent backflow.

Final Steps

27. Release pressure from the extractor.
28. Quickly transfer the moist soil to a pre-weighed dish.
29. Immediately weigh the wet soil (0.01 g accuracy).
30. Dry the sample in an oven at 105 °C for 24 hours.
31. Place dried samples in a desiccator to cool, then weigh.

Calculation

$$\text{moisture (\%)} = \frac{\text{wet sample (g)-ovendry sample (g)}}{\text{ovendry sample (g)}} \text{ x } 100$$

Comments

1. **Never remove extractor lid with pressure in the container**.
2. If the outlets of the plates continue to bubble after a few hours of applied pressure, the plates are probably defective and should be replaced.
3. Pressure should not be allowed to fluctuate during a run. It should be checked every 2-3 hours (and adjusted if necessary). If the pressure fluctuations are within the specified tolerance of the regulator, then no adjusting is needed.
4. The height of the sample in the ring should be as small as possible to reduce the time required to reach equilibrium, which is proportional to the square of the height of the sample in the ring.
5. 33 kPa

   For quality control, three ceramic plates can be used in the extractor for each run. One reference sample is used for each plate. Duplicates are done on approximately 5% of samples.

   Precision of moisture should be less than or equal to 5%.

   1,500 kPa
   For quality control, three ceramic plates can be used in the extractor for each run. One reference sample is used for each plate. Duplicates are done on approximately 5% of samples.

   Precision of moisture should be less than or equal to 10%.

6. AWC values for a particular crop computed from in situ observations of FC and PWP and those from laboratory methods may be quite different, partly because laboratory methods do not consider root distribution.

References

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Aggregate Stability

Soil particles are bound together by physical, chemical, and biological forces to form structural units known as aggregates—the basic building blocks of soil structure. Good soil structure enhances aeration, drainage, and infiltration; protects soil organisms and soil carbon; supports optimal germination and root growth; and contributes to erosion resistance, reduced compaction, easier soil workability, and improved nutrient cycling.

Aggregate formation is a dynamic process. Physical mechanisms such as freezing–thawing cycles and the swelling and shrinking of clay minerals rearrange soil particles. Chemical mechanisms, including the aggregating effects of divalent cations (especially calcium) and the formation of organo-clay complexes, help bind particles together. Biological activity also plays a major role: soil fauna and plant roots can disrupt or stabilize aggregates, fine roots and fungal hyphae enmesh particles, and root exudates and microbial secretions act as organic "glues" that cement particles and organic matter.

Aggregates are commonly grouped into macroaggregates (0.25–5 mm), which consist of smaller microaggregates (0.002–0.25 mm). Particles smaller than 0.002 mm typically consist of very fine minerals and tiny organic matter fragments (Brady & Weil, 2017; Scheffer & Schachtschabel, 2002).

Aggregate stability describes the ability of aggregates to resist breakdown under environmental stresses such as wetting, raindrop impact, or mechanical disturbance. It is often improved by management practices such as reduced tillage, cover cropping, organic amendments, and residue retention (Rieke et al., 2022). Because aggregate stability reflects both the physical resilience of the soil and the influence of biological activity and organic matter dynamics, it serves as a valuable indicator of soil functioning and management impacts.

However, aggregate stability is also strongly shaped by inherent soil properties—especially texture and mineralogy—and by climatic factors that influence natural wetting–drying or freezing–thawing cycles. For this reason, aggregate stability results should be compared only among similar soils within similar climate regions (SHI, 2023). Comparisons across very different soil types or climates can be misleading and do not meaningfully reflect management effects.

Method

The method described here is adapted and slightly modified from the **Wet Aggregate Stability by Image Quantification** procedure developed by the Soil Health Institute (SHI, 2023), which itself builds on the method published by Fajardo et al. (2016). When compared

with more sophisticated and costly laboratory techniques, the SLAKES method has been shown to perform equally well, even though—like all methods—it has its own advantages and limitations (Rieke et al., 2022).

This low-cost method is based on quantifying the dispersion of aggregates after a 10-min submersion in water. Pictures of the soil aggregates taken before and after the submersion. The area of soil in both images is estimated automatically using a pixel thresholding algorithm in the free programming software *R* (see R-code provided in the appendix).

Alternatively, a smart phone application called **"Slakes"** is available for android and iPhone. The application can be found in google Play Store or in the AppStore, respectively (see icon in Figure 1). **The app also contains a detailed description of the procedure.**

Figure 1: Icon of the Slakes application or iPhone or android

The method produces a dimensionless aggregate stability index that is determined as the ratio of initial to final soil area. Values of the index range from ~0.1 to 1, with higher values indicating greater aggregate stability.

A) Slakes Test Using Smartphone Application

Material

- Air-dry soil aggregates (4–8 mm diameter)
- Transparent or white dishes, about 90 mm Petri dishes are recommended
- SLAKES smartphone application installed
- Mount for a smartphone
- Water
- Tweezers or small forceps (those used for entomology are preferred)
- Labeling materials
- Backlighting is recommended

Procedure

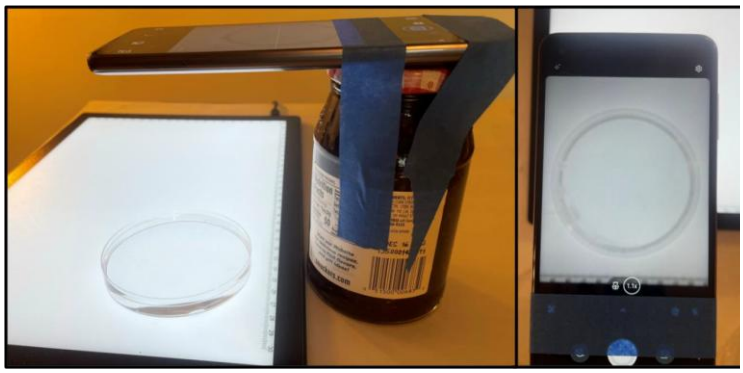
Sample preparation

1. Gently sieve air-dry soil to obtain **three aggregates** between **4 and 8 mm** (pea-sized). Avoid crushing the aggregates.
2. Select representative aggregates free of large stones, obvious root fragments or other debris.

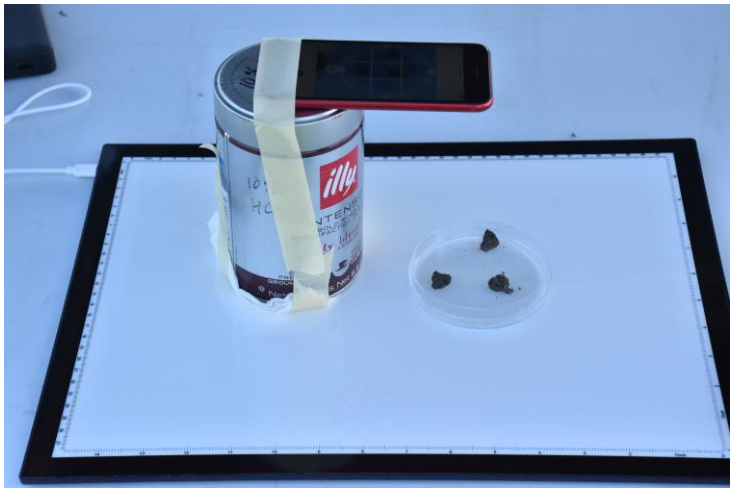
Test setup

(see pictures 3 – 5)

3. Place the three aggregates in the dish so that they do not touch each other and are not too close to the edge of the dish.
4. Position the smartphone directly above the dish using a stable mount. Ensure even lighting (backlighting helps).
5. Fill a second dish or container with enough water to fully submerge the aggregates (at this stage, do not yet place the aggregates into the second dish).
6. Open the **SLAKES app** and follow the on-screen instructions to run the test.



Picture 3: The phone positioned over the Petri dish (left) with no outside objects in the view (right). (Picture credit: Soil Health Institute)



Picture 4: Phone positioned over aggregates after first photo is

Picture 5: Phone positioned over transparency to take a series of initial and then final photos (left). Cross hairs on an example camera app are aligned with guide marks on transparency to aid in repositioning the phone (right). (Picture credit: Soil Health Institute)

B) Slakes Test Using camera and R Code

(This method is efficient when analyzing many aggregates at the same time.)

Materials

- Air-dry soil aggregates (4–8 mm diameter)
- Transparent or white dishes (90 mm Petri dishes recommended)
- Digital camera or smartphone used as a fixed camera
- Camera mount or tripod
- Tweezers or small forceps
- Labeling materials
- Backlighting (recommended)
- Computer with R installed
- Prepare a dedicated folder for SLAKES images
- R script for image analysis (provided below)

Procedure

Follow **Steps 1–4** from the smartphone app sample preparation.

5. Place the dish containing the dry aggregates (but no water) under the camera.
    - Mark three small points outside the dish (e.g., on the background) to help reposition the dish consistently
6. Take a reference image of the dry aggregates before wetting.
7. Replace the first dish with the second dish that contains water, ensuring the dish is positioned as in the reference image using the points marked outside the dish.
8. Using tweezers or forceps, gently place the aggregates into the water-filled dish.
9. Immediately take a photo (t = 0).
10. Take a second photo after 10 minutes (or at the interval required by the protocol).
11. Save all images in a single folder using a consistent naming convention (e.g., SampleID_time.jpg).
12. Import the images into R.
13. Run the SLAKES R script. The script will perform image analysis and calculate a stability index for each aggregate.

References

Brady and Weil. 2017. *The Nature and Property of Soils*. 15th Edition. Pearson

Fajardo, M., McBratney, A. B., Field, D. J., & Minasny, B. 2016. *Soil slaking assessment using image recognition*. Soil and Tillage Research, 163, 119–129. https://doi.org/10.1016/j.still.2016.05.018

Rieke et al. 2022. *Evaluation of aggregate stability methods for soil health* https://www.sciencedirect.com/science/article/pii/S0016706122004633

Scheffer and Schachtschabel. 2002. Lehrbuch der Bodenkunde. 15th Edition. Spektrum

SHI 2022. Soil Health Institute: *Comparing Aggregate Stability Methods*. Posted 13Oct2022. https://soilhealthinstitute.org/news-events/comparing-aggregate-stability-methods/ accessed 03Mar2024.

SHI 2023. Soil Health Insitute. *Wet Aggregate Stability by Image Quantification*. Version 1, published 23 January 2023. https://soilhealthinstitute.org/app/uploads/2022/06/SOP_AggStability_ImageRecogn_simplified_v1.2.pdf

Total Soil Organic Carbon

Method

Dry combustion method (Flash, 2000).

If soil pH $\geq$ 6.0 ($CaCl_2$), determine the inorganic carbon by acidification and sparing at 150°C (Skalar Method P05-001). Soil organic carbon is the difference between total soil carbon and inorganic carbon.

Comment

pH_{water} is higher than pH_{CaCl2}. The differences can range between 0.6 – 1.2 (DPIRD-WA, 2024)

References

DPIRD-WA. Western Australia Department of Primary Industries and Regional Development's Agriculture and Food division https://www.agric.wa.gov.au/soil-acidity/soil-ph?nopaging=1 [retrieved 29 Oct 2024]

Flash 2000 Elemental Analyzer Operating Manual. Thermo Fisher Scientific Revision D 2010

Skalar Method: Total Inorganic Carbon Catnr: P05-001

Soil Nutrients

Method

Dry combustion method (Flash, 2000).

Reference

Flash 2000 Elemental Analyzer Operating Manual. Thermo Fisher Scientific Revision D 2010

Extractable Nitrate, Phosphorus and Potassium

Nitrate

Method

2N KCl extraction and auto-analyzer (Maynard et al. 2007)

Phosphorus

Method

Mehlich III extraction (Ziadi, N. and Sen Tran, T. 2007) and measurement by either UV-Vis or ICP-OES.

1. Report analytical method.
2. Report values as is (Mehlich III) and by conversion as Kelowna values (Poon and Schmidt, 2010).
3. Kelowna-P = 0.72 x Mehlich-3 P (all pH)

Kelowna-P = 0.74 x Bray-1 P (at pH < 7.2)

Kelowna-P = 1 x Bray-1 P (at pH ≥ 7.2

Kelowna-P = 0.99 x Bicarbonate (Olsen)-P (at pH < 7.2)

Kelowna-P = 1.21 Bicarbonate (Osen)-P (at pH ≥ 7.2)

Potassium

Method

Mehlich III extraction (Ziadi, N. and Sen Tran, T. 2007) and measurement by ICP-OES.

Report values as is (Mehlich III) and by conversion as Kelowna (Poon and Schmidt, 2010)

Kelowna-K = 0.75 x Mehlich-3

Kelowna-K = 0.80 x Ammonium Acetate K

References

Maynard et al. 2007. Nitrate and exchangeable ammonium nitrogen. In: Carter, M.R. and Gregorich, E.G. (Eds) Soil Sampling and Methods of Analysis, 2nd Edition CRC press, pp: 599 – 606

Poon, D. and Schmidt, O., 2010. Understanding Different Soil Test Methods. Nutrient Management Factsheet – No. 3 in Series (https://www2.gov.bc.ca/assets/gov/farming-natural-resources-and-industry/agriculture-and-seafood/agricultural-land-and-environment/soil-nutrients/600-series/631500-9_soil_test_methods_factsheet_no3_sep2010.pdf)

Ziadi, N. and Sen Tran, T. 2007. Mehlich 3-extractable elements. In: Carter, M.R. and Gregorich, E.G. (Eds) Soil Sampling and Methods of Analysis, 2nd Edition CRC press, pp: 81 - 88

pH

Soil pH can be measured using either distilled water (H_2O) or a calcium chloride solution ($CaCl_2$), and the two methods often produce slightly different results. pH in water reflects the soil's active acidity under field-moist conditions and is more sensitive to short-term changes such as recent fertilization, drying, or seasonal variation. pH in $CaCl_2$, typically measured in a 0.01 M solution, provides a more stable reading because the added calcium reduces the influence of soluble salts and temporary fluctuations in soil moisture. As a result, pH values in $CaCl_2$ are usually 0.5 to 1.0 units lower than pH measured in water. Measuring pH in both media can improve interpretation, as the difference between the two values can offer insights into soil salinity, ionic strength, and buffering capacity.

Method

$CaCl_2$, pH meter (Hendershot et al., 2007)

References

Hendershot, W.H., Lalande, H., and Duquette, M. 2007. Soil Reaction and Exchangeable Acidity. In: Carter, M.R. and Gregorich, E.G. (Eds) Soil Sampling and Methods of Analysis, 2nd Edition CRC press, pp: 173 – 174

Electrical Conductivity

Method

1:2 soil to water ratio, conductivity meter (Kalra and Maynard, 1991)

References

Kalra, Y.P. and Maynard, D.G. 1991. Electrical Conductivity and Soluble Salts. In: Methods manual for forest soil and plant analysis. Forestry Canada, pp: 37

Appendix

R Code for Aggregate Stability Assessment

#This sample code is Appendix A for the Soil Health Institute's Standard Operating Procedure for Wet Aggregate Stability by Image Quantification.

#The code was last updated on May 16th, 2022. Email **erieke@soilhealthinstitue.org** with questions.

#The code uses two images of a petri dish containing three soil aggregates. One image is taken at the start and one is taken at the end of a 10 minute

#submersion in water. The code calculates the mean aggregate stability for the three aggregates in the petri dish by 1) cropping the images, 2) converting them to

#grayscale, 3) converting to binary by using a threshold obtained using Otsu's method (this value represents the cutoff value between water and aggregate pixels)

#and then 4) dividing the total initial area of aggregates (not individual aggregate areas) by the total final are of aggregates. In step 5), an example of how to save data is given.

#You will need to update file paths to locate the images on your computer lines and to save output.

library(EBImage)#Helpful resource to download EBImage
https://bioconductor.org/packages/release/bioc/html/EBImage.html

library(dplyr)lib

```
# path to folder where results are/will be stored -- need forward slash
results_path <- 'C:/Users/SlakeItEasy_results/'

# read in the initial and final images and plot to visualize

# you don't have to plot, but it can be helpful to confirm you have the correct image img_Initial <-
readImage("C:/Users/Slake it easy/Images/InitialTestImage.jpg")
        plot(img_Initial)

img_Final <- readImage("C:/Users/Slake it easy/Images/FinalTestImage.jpg") plot(img_Final)

#get the image dimensions to inform where to crop
        dim(img_Initial)

        dim(img_Final)
```

1) crop images so they only contain the three aggregates you intend to analyze. #It is good practice to leave the sample ID and replicate number in the image

#If you already cropped your images before reading in, you need to rename them.

#To rename your image, you need to comment out lines 39 and 41 and uncomment lines 34 and 35 and run them:

```
#img_Initial_crop <- img_Initial
#img_Final_crop <- img_Final

#No other aggregates should be in the image and no petri dish edges should be present. #you may have to guess and check to get the right crop.

img_Initial_crop <- img_Initial[350:870, 250:650,]#

plot(img_Initial_crop)#plot to see if you got a good trim, if not, change numbers
and try again. Note that images have three bands and the first is plotted by default.
We will average over bands at the end.

img_Final_crop <- img_Final[370:900, 250:700,]
plot(img_Final_crop)

#2 make images gray scale

colorMode(img_Initial_crop) <- Grayscale

colorMode(img_Final_crop) <- Grayscale

#3 get the threshold values for the images so we can make them binary, these are essentially the cutoffs for what values will be classified as soil

threshold_Initial <- otsu(img_Initial_crop)

threshold_Final <- otsu(img_Final_crop)

#make binary versions of the images by selecting values less than the threshold (found in lines 48 and 49) and then summing the pixels that are

#greater than the threshold (lines 48 and 49). These are done for each of the three image bands.

img_binary_Initial <- EBImage::combine( mapply(function(frame, th) frame <= th,
getFrames(img_Initial_crop), threshold_Initial, SIMPLIFY=FALSE) )

plot(img_binary_Initial)

img_binary_Final <- EBImage::combine( mapply(function(frame, th) frame <= th,
getFrames(img_Final_crop), threshold_Final, SIMPLIFY=FALSE) )

plot(img_binary_Final)

area_Initial <- apply(img_binary_Initial, MARGIN = 3, sum, na.rm = T)

area_Final <- apply(img_binary_Final, MARGIN = 3, sum, na.rm = T)

#4 The stability index for this petri dish is initial area divided by the final area. The mean is taken across the three image bands.

StabilityIndex<-mean(area_Initial/area_Final)
```

```
        StabilityIndex

#5 Example of how to save results

        #This will name results with the year, month, day, hour, and minute  processing_datetime <-
format(Sys.time(), "%Y%m%d%H%M")
        #this will save the value to a CSV

        write.csv(StabilityIndex, paste0(results_path, 'slaking_image_results_',          processing_datetime,
'.csv'), row.names = F)
```