NEON Field and Lab Procedure and Protocol: TIS Soil Archiving

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A	05/09/2012	ECO-00409	Initial Release
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1 INTRODUCTION

1.1 Purpose

The primary purpose of this document is to provide a change controlled version of Observatory protocols and serve as the version used for external review by subject-matter experts. This document provides the content for training and collecting field-based materials for NEON staff and contractors. Content changes (i.e. changes in particular tasks or safety practices) occur via this change controlled document, not through field manuals or training materials.

This document is a detailed description of the field data collection, relevant pre- and post-field tasks, and safety issues as they relate to this procedure and protocol.

1.2 Scope

This document relates the tasks for a specific field sampling or laboratory processing activity and directly associated activities and safety practices associated with the field collection of- and archiving procedures for the TIS soil archive. This document does not describe:

- general safety practices (e.g., how to drive a boat)
- site-specific safety practices (e.g., how to safely walk in a stream)
- general maintenance (e.g., fill the car with gas)

1.3 Acknowledgements

Members of the FIU Soils Working Group (M. Cosh, D. Eissenstat, J. Harden, E. Kelly, V. Romanovsky, J. Tang, R. Vargas, C. Wells) and Luke Nave provided valuable comments on an earlier version of this protocol.

2 RELATED DOCUMENTS AND ACRONYMS

2.1 Applicable Documents

AD[01]	Soil pit protocol (TBD)
AD[02]	
AD[03]	
AD[04]	

2.2 Reference Documents

RD[01]	NEON.DOC.000008 NEON Acronym List
RD[02]	NEON.DOC.004316 EHS Safety Policy and Program Manual
RD[04]	NEON Sampling Design Document
RD[05]	Training Plan
RD[06]	QA/PA Plan
RD[07]	DOORS requirements database

2.3 Acronyms

NEON	National Ecological Observatory Network
P&P	Procedure and Protocol
TBD	To be determined
TIS	NEON Terrestrial Instrument System
USDA APHIS	United States Department of Agriculture Animal and Plant Health Inspection Service

3 BACKGROUND AND OBJECTIVES

3.1 Background

Archived soil samples are necessary to serve as a reference of current soil physical and chemical conditions, and for use with future analytical techniques that are yet to be derived or invented (Boone *et al.* 1999). Previous scientific studies using archived soils have produced a wide range of novel findings, including:

- assessing changes in lead pollution following the introduction of unleaded gasoline (Friedland *et al.* 1992),
- refining estimates of organic matter turnover using ^{14}C bomb carbon (Trumbore 1993),
- measuring the accumulation of atmospheric inputs such as sulfur (Lapenis et al. 2004) or pesticides, including DDT, in soils throughout the 20th century (Meijer et al. 2001).

In addition, archived soil samples and associated measurement data can be used to calibrate new techniques/instruments/models to data collected with earlier methods or technologies.

The Terrestrial Instrument System (TIS) soil archive will consist of air-dried soil. As noted by Boone *et al*. (1999), each soil storage condition (*e.g.*, dried, frozen, or refrigerated) has scientific limitations, and there is no perfect storage condition. Air-dried and sieved samples (air-dried, but not sieved, for O horizon samples) were chosen for the TIS Soil Archive because this storage condition:

- is suitable for a range of physical and chemical analyses, even after decades of storage (Boone et al. 1999),
- is less likely to be compromised by power failures (unlike frozen samples),
- is widely used in other soil archives (*e.g.*, USDA Natural Resource Conservation Service Soil Archive, Rothamsted Archive, Australian National Soil Archive, Scottish National Soil Archive, Hubbard Brook Sample Archive), thereby facilitating inter-comparisons,
- has proven to be appropriate for previous scientific studies, as evidenced by published studies that have made use of air-dried and sieved samples (*e.g.*, Trumbore 1993, Meijer et al. 2001).

3.2 Science Requirements

This protocol fulfills the following Observatory science requirements: NEON.TIS.4.1009

3.3 Data Products

There are currently no data products (DPs) associated with TIS soil archive samples because no measurements are currently planned to be made on these samples by NEON. The archived samples are a resource that NEON shall provide to the community and could provide significant scientific information in the future. There are however, chemical and physical analyzes preformed on samples collected in the same location, soil horizon, and time, which are associated with L1 DPs.

4 PROTOCOL

As described above, the samples in the TIS soil archive will constitute a valuable resource to the scientific community, as the samples provide information on soil conditions at the time of collection and an opportunity to calibrate new techniques/instruments/models to data collected with earlier methods or technologies.

This document describes the processing of samples for the TIS soil archive after they are received at NEON headquarters. A description of how soil samples for the FIU Soil Archive shall be collected in the field is given in AD[01]. Briefly, independent soil samples (~8 L at field moisture) will be collected from each soil horizon exposed in a single soil pit at each NEON core and relocatable site. At most sites, the soil pit will extend 2 m deep or to bedrock, however, it will extend 3 m deep at sites with permafrost. Because only one soil sample will be archived per soil horizon at each site, the TIS Soil Archive will be useful for regional and continental scale studies, but not local scale studies due to the lack of spatial sampling at that local-scale. The archived samples will be suitable for a range of physical and chemical analyses, but will typically not be suitable for assessments of soil biology or structure.

The processing of soil samples shall differ depending on soil horizon. Soil from the O horizon shall be air-dried, but NOT sieved prior to archiving. Whereas, soil from other horizons shall be air-dried and sieved (2 mm) prior to archiving.

To minimize contamination, all materials that come into contact with the soil samples (*e.g.*, trays for drying, spoon/spatulas, storage containers) shall be clean and un-corroded. Materials made of glass and corrosion resistant metal, not plastic, shall be used to process and store samples whenever possible. Glass and metal are recommended since they are unlikely to contaminate the soil samples or compromise any future analyses that may be performed on the samples. To avoid cross-contamination among soil samples, all materials shall be cleaned prior to processing the next sample. If soap is used for cleaning it shall be phosphate-free (e.g. Alconox). Quaternary ammonia must NOT be used for sterilizing soil or equipment used for the TIS Soil Archive to minimize the risk of contaminating the archive samples.

Once the sample has been dried, it will be processed and divided into four subsamples that will be stored in four amber glass jars. The purpose of the four jars is to provide redundancy, and to minimize the risk of losing an archived sample, *e.g.*, due to contamination, spillage, or loss.

Soil from several NEON sites (*e.g.*, Hawaii and Puerto Rico) must be quarantined according to USDA APHIS regulations. Furthermore, soils samples must be transported following USDA packing standards. To simplify this protocol and minimize the risk of accidental release of quarantined soil, NEON shall treat soil from all sites as quarantined and all parts of this protocol shall comply with NEON's USDA APHIS soil permit (see permit for details). This includes sterilizing or destroying, using an approved method, anything that came into contact with the soil. Refer to NEON's USDA APHIS permit for instructions to sterilize soil and material that came into contact with soil prior to disposal.

4.1 Protocol assumptions

For the purpose of this document, we assume that the TIS soil archive will be housed at NEON headquarters. We assume that the samples sent from the field site have not yet been dried, and that subsequent processing of the samples (including drying, sieving, subsampling, labeling, and entering metadata) will be conducted at NEON headquarters. For planning purposes, we assume 6 soil horizons/layers will be identified from each soil pit, which will result in 6 samples for the TIS soil archive from each NEON core and relocatable site.

Soils from several NEON sites is quarantined by USDA APHIS, including those from Hawaii, Puerto Rico, and several sites in the southeast US where fire ants are present. As a result, we assume all soil samples

for the TIS soil archive shall be treated as quarantined soil to simplify the processing procedures and minimize the risk of accidental release.

5 QUALITY ASSURANCE AND QUALITY CONTROL

Subsamples in the TIS soil archive shall be re-weighed every 3 years to ensure that the quantity of soil that is recorded as being in the archive matches the quantity that is actually present in the archive.

6 DECISION TREE

Table 1. Decision tree associated with processing samples for the TIS soil archive indicating how to respond to a delay in processing the samples once field collected, and consequences of the delay.

Delay	**Action**	**Adverse outcome**	**Outcome for TIS soil archive**
1-30 days	If delay occurs prior to starting the procedure, store samples at ~4 °C (*i.e.*, refrigerate)	None	None
	If delay occurs after sample(s) have reached an air dry state, do nothing	Increased potential for contamination or spillage of sample(s)	None if an incident does not occur
>30 days	If delay occurs prior to starting the procedure, store samples at ~4 °C (*i.e.*, refrigerate)	Aspects of the sample, including chemical composition, may be altered. Particularly as the length of the delay increases.	Uncertainty in some measured properties of the sample(s) will be increased by an unknown amount.
	If delay occurs after sample(s) have reached an air dry state, do nothing	Increased potential for contamination or spillage of sample(s).	None if an incident does not occur

7 SAFETY

Personnel working at a NEON site should be familiar with and practice safe field and lab work as outlined in the EHS Safety Policy and Program Manual. Additional safety issues associated with this field procedure are outlined below. The Field Operations Manager and the Lead Field Technician have primary authority to stop work activities based on unsafe field conditions; however, all employees have the responsibility and right to stop their work in unsafe conditions.

Laboratory safety training is required prior to operating laboratory equipment, including the drying ovens.

8 PERSONNEL REQUIREMENTS

Demonstrated ability to follow standard operating procedures in a laboratory setting. Experience using standard laboratory equipment (*e.g.*, balances, drying ovens). Experience working with environmental samples (*e.g.*, soil, plant, or water samples) is desirable. Experience working with quarantined samples is desirable.

9 TRAINING REQUIREMENTS

The training plan for this procedure is to read this document, read NEON's USDA APHIS soil permit, discuss the procedure with FIU personnel, and contact FIU personnel when issues or concerns arise. It is essential that any personnel undertaking this procedure are familiar with the current version of NEON's USDA APHIS soil permit, as this permit dictates how to transport soil samples, and how quarantined soil, and material that has come into contact with quarantined soil, can be safely disposed of or sterilized. Any personnel undertaking this procedure for the first time shall be supervised by personnel that are familiar with the procedure. Note that the USDA APHIS soil permit can be updated at any time, therefore, close communication between NEON EHS and personnel performing this protocol is required.

10 FIELD STANDARD OPERATING PROCEDURE

The procedure for collecting samples for the TIS soil archive in the field is described in AD[01].

11 LAB STANDARD OPERATING PROCEDURE

The procedure shall always fully comply with all restrictions outlined in NEON's USDA APHIS soil permit. If a discrepancy is identified between the procedure outlined in this document (or other NEON documents) and the USDA APHIS soil permit, the permit has precedence. If this occurs, contact a FIU Manager through the NEON Problem Reporting and Tracking system.

The laboratory procedure has the following goals: 1) dry and sieve (if necessary) the soil samples; 2) weigh the predetermined amount of soil for each of the subsamples into labeled containers; 3) store the samples in the TIS soil archive; 4) record and enter metadata associated with each sample in the TIS archive; and 5) monitor and record conditions and use of archived samples.

Under no circumstances should the floor (or walls) of the soil archive room be either waxed, or have the use of wax strippers (NH4, ammonia) be used for the life of the soil archive. This is a requirement, and must be communicated to building managers, FCC, cleaning staff etc. Use of these materials can contaminate the entire archive.

11.1 Timing

- Begin drying of soil samples within 30 days of the arrival of the samples at NEON headquarters (preferably within 1-3 day).
- While samples are drying, their weight shall be measured at least every 3 days (preferably daily) to determine whether they are dry (i.e., they shall be dried to constant weight).
- Once samples are dry, they shall be placed within sealed glass containers within 4 hours to minimize the risk of spillage or contamination if processing cannot begin before then.

- Once samples are dry, they shall be sieved (if necessary), subsampled, and added to archive jars within 7 days.
- Metadata relating to any step in the processing of samples for the TIS soil archive shall be entered within 1 day of completion of that step to minimize mistakes in the metadata (*e.g.*, forgetting how a particular sample was processed).

11.2 Lab Procedure

11.2.1 Equipment and Materials

Table 2. Materials and supplies required for the Procedure.

Item Description	Quantity per sampling event	Hazardous Chemical
Scissors/blades to open shipping containers	2	No
Laboratory gloves (*e.g.*, latex or nitrile)	12/sample	No
Printable labels	1/sample	No
Corrosion resistant metal drying tray (approx. 50 x 30 x 5 cm; L x W x H)	1/sample	No
Drying oven(s) for samples	Space as needed	No
Calibrated mass balance (≤0.1 g accuracy; up to ≥6 kg capacity)	1	No
Set of calibration standards for mass balance	1	No
Sieves with 2 mm mesh	4	No
Wire brush	1	No
Container to dispose of rocks, roots, and other debris	1	No
Wide-mouth glass container with screw-cap lid. Glass shall be amber and container shall meet EPA's performance based specifications for metals (Group 1) and semi-volatiles/ pesticides/ PCB's (Group 2) analysis.	4/sample	No
Metal spatulas or spoons	4	No
Shelves with storage boxes	Space as needed	No
Data sheets	1 set	No
Pencil	1/technician	No
Faucet, sink, and drying rack/space	1	No
Detergent	1 bottle	No
Scrubbing brush	2	No
Parchment (baking) paper	1 roll	No
Bleach for sterilizing materials	1 liter	Yes
Buckets/tubs for sterilizing materials	4	No
Oven for sterilizing materials and samples for disposal	1	No
Ethanol (95%)	0.5 liters	Yes
Sample splitter	1	No

11.2.2 Preparation

Ensure that you are familiar with the current version of NEON's USDA APHIS soil permit. Ensure that all storage, processing, and cleaning locations comply with NEON's USDA APHIS permit at all times (see permit for details).

Ensure that the processing area is clean and that there is sufficient space (including in drying ovens, drying racks) prior to beginning sample processing. This includes making sure that the floor has been swept so that if a sample is spilt, it can be recovered with minimal contamination.

At least 10 days prior to the expected receiving date for TIS soil archive samples, check the inventories of all laboratory consumables (e.g., labels, gloves, sterilizing supplies). Keep enough supplies to process at least 24 samples available at any time in case samples arrive unexpectedly.

When the archive storage reaches 80% of capacity, consider ordering additional storage shelves/cabinets and storage boxes. Acquire additional storage capacity if the current NEON construction schedule implies that storage capacity will be exceeded within 1 year.

11.2.3 Sample Processing in the Lab

Always wear gloves when handling soil for the TIS soil archive. Gloves can be removed and destroyed using an appropriate method as defined in NEON's USDA APHIS soil permit, whereas it is much harder to appropriately dispose of quarantined soil on your hands.

It is preferable that all data and metadata recorded during this procedure be entered directly into electronic form (rather than a data sheet, which must then be transcribed to electronic form), as this will eliminate a potential source of errors. If a data sheet is used, the original data sheet shall be archived within the NEON soil archive.

All materials (including trays, sieves, and spoons/spatulas) must be cleaned in between processing each sample to avoid cross-contamination. If soap is used for cleaning it shall be phosphate-free. Do not use quaternary ammonia for sterilizing soil or equipment used for the TIS Soil Archive to minimize the risk of contaminating the archive samples. Equipment or material used in procedures where quaternary ammonia is used must not be used for processing or storing TIS Soil Archive samples. Immediately prior to use, all materials shall be wiped with 95% ethanol.

1. Put on lab gloves. Make sure the drying trays, data sheets, pencil, scissors/blades, and shipping container are accessible.

2. Open the shipping container(s) and remove the soil samples. Ensure that none of the samples have leaked during transit. If a leak or leaks have occurred note the affected sample(s) on the data sheet, and collect the sample if possible (*i.e.*, if the sample appears uncontaminated).

3. All packing material, including shipping containers, shall be sterilized or destroyed by an approved method according to NEON's USDA APHIS soil permit. Suggested method that

complies with NEON's USDA APHIS permit as of Jun 2012 is to generously spray container with ethanol (≥70%) to the point of runoff.

4. Ensure that each soil sample is labeled correctly. If discrepancies in labeling are identified, contact an FIU team member or the FIU contractor who collected the samples for clarification through the NEON Problem Reporting and Tracking system. Follow-up until resolved.

5. Remove and correctly dispose of lab gloves. Then, print out labels corresponding to each soil sample (sample label format: "Domain_Site_Location_Top-Bottom depth (cm)_Sample_Horizon", e.g., Woodworth_Pit1_16-32_Archive_2Bw). Ensure that the labels comply with the current version of NEON's USDA APHIS soil permit. For example, as of February 2012, the permit specifies that all containers of quarantined soil include "*Quarantine Soil-Sterilize Before Disposal*" on their label.

6. Put on lab gloves and then label a drying tray with a label corresponding to one of the soil samples.

7. Ensure that the balance is level and that it has passed a traceable calibration within the last year. Generally, balances need to be re-leveled any time they are moved and re-calibrated after receiving a physical shock (*e.g.*, being dropped, or abruptly moved).

    a. Weigh the empty tray on the balance and record the weight (in grams) on the data sheet.

    b. Empty the soil from the appropriate soil sample into the drying tray. Break up any large clumps of soil and spread the soil out evenly across the tray. To ensure rapid drying, make sure that soil depth is ≤5 cm (if necessary, additional labeled trays can be used).

    c. Weigh the tray and soil and record the weight on the data sheet. If your gloves become coated in soil (which will occur if the soil is moist/wet), change gloves before handling the next soil sample to avoid contamination. Dispose of gloves according to the specification in NEON's USDA APHIS soil permit.

8. If the soil is moist or dry, cover the tray with a single layer of parchment (baking) paper to minimize the risk of contamination from spillage of other samples. Place the tray on a shelf or bench where it is unlikely to be knocked off. [If water has pooled in the tray, do not place parchment paper over the sample as it may disintegrate when it gets wet and contaminate the sample. Instead leave the tray uncovered until pooled water has evaporated.]

9. If shorter drying times are required (*e.g.*, due to a high volume of samples over a short period of time), the trays can be placed in a drying oven. If this is done, set the oven to 30 °C. If the oven has a cut-out temperature to prevent overheating, set the cut-out temperature to 34 °C. Soil for the TIS soil archive must not be heated above 35 °C, and the oven temperature is therefore set to 30 °C to allow for variability and uncertainty in oven temperature. Wear protective equipment (gloves, long-sleeves, etc) as necessary based on NEON lab safety training.

10. Mix the soil by hand at least every 3 days (preferably daily) to facilitate rapid drying. Wearing gloves, mix the soil and hand-crush larger aggregates. Change gloves before handling the next soil sample to avoid contamination. Dispose of gloves according to the specification in NEON's USDA APHIS soil permit.

11. Weigh the tray at least every 7 days (preferably more frequently) and record the weight on the data sheet to determine whether the soil has reached an air dried steady-state.

    a. Remove the parchment paper before weighing the soil. The soil shall be considered air dried steady-state when the weight changes by less than 0.5% over at least 7 days. The change in weight shall be calculated as:

$$\Delta s = \frac{(W_{t0} - T) - (W_{t1} - T)}{(W_{t1} - T)} \times 100 \quad (1)$$

    where Δs is the change in soil weight expressed as a percentage between time 0 (*t0*) and time 1 (*t1*), *W* is the weight of the soil and tray, *T* is the weight of the tray, and *t1* – *t0* ≥ 7 days. If Δs is ≥ 0.5 the soil needs to continue drying; if Δs is <0.5, the next step in the procedure can occur.

12. If the soil is from the O horizon (defined as any soil horizon where the first letter of the horizon designation is a capital "O", e.g., Oa, OA, O/A, 2O, or O'i1) proceed directly to Step 13 (i.e., do not sieve the O horizon soil). If the soil is from any other horizon (e.g., A1, Bw, C2, AO), once it is air dry it shall be sieved within 7 days.

    a. Examine the 2 mm mesh sieve to make sure that it is clean and that it does not have any gaps larger than 2 mm. Sieves often wear out where the mesh attaches to the rim, so make sure that the mesh is not starting to detach, which would allow soil aggregates larger than 2 mm to pass through the sieve. Wipe down sieve with 95% ethanol.

    b. Prior to beginning sieving, put on gloves and make sure there is container nearby where you can place debris that does not pass through the sieve. Also, label 4 clean glass jars with the appropriate sample code. Both the jar and jar lid shall be labeled to minimize the chance of the lids being mixed up among the jars.

    c. Gather all of the soil at one end of the tray, and pour the soil into the sieve sitting in another labeled tray. Shaking and tapping the sieve helps the soil to pass through more quickly. Larger lumps of soil can be broken up by hand or pushed through the sieve by hand. Material (*e.g.*, rocks, roots) that collects on the sieve surface should be placed in the container for appropriate disposal. The entire sample shall be sieved.

    d. Weigh out 1200 g soil and pass that soil through the sample splitter twice to create four subsamples of ~300 g soil each. The sample splitter must be cleaned before processing soil from different soil horizons. Cleaning with compressed air followed by rinsing with 95% ethanol should be sufficient to remove soil particles in most cases.

e. Weigh each of the 4 jars and jar lids and record the weight on the data sheet. Take care to keep the lid associated with the correct jar – the jar and lid should have the same label.

f. Add a ~300 g soil subsample to a jar and re-weigh the jar to determine the weight of soil in the jar. Ensure that each jar receives 300 ± 1 g soil. If the weight of soil added to the jar is outside 300 ± 1 g add or remove soil using a metal spatula/spoon. Record the weight of soil added to each jar in grams to one decimal place (*e.g.*, 300.3 g) on the data sheet.

g. If 300g of soil cannot fit into the amber jars, fill the jars and weigh. Then, add additional jars until 1200g of soil is to be stored.

h. Proceed directly to Step 14.

13. [**Organic soil only**] Gather the soil in one corner of the tray, remove rocks ≥10 mm and roots ≥5 mm diameter by hand, and mix it thoroughly by hand to ensure that the sample is homogenous.

    a. Once mixed, avoid tapping or shaking the tray as this will lead to fractionation of the soil aggregates (*e.g.*, small aggregates will move toward the bottom of the pile and larger aggregates will remain on top). This is important as it will ensure that the subsamples are as similar to one another as possible.

    b. Weigh each of the 4 jars and jar lids and record the weight on the data sheet. Take care to keep the lid associated with the correct jar – the jar and lid should have the same label.

    c. Use a clean metal spoon or spatula to add the soil to the jars. Add one spoonful to each jar sequentially and then repeat. This is important as it will minimize differences among the 4 subsamples. In contrast, if you completely fill each jar before moving on to the next jar it is more likely that the subsamples will differ, because, for example, small aggregates may fall towards the bottom of the pile during the procedure, hence the last jar will contain disproportionately more small aggregates and fewer large aggregates.

    d. If necessary the soil sample can be re-mixed by hand to ensure it remains homogenous while being weighed into the jars.

    e. Ensure that each jar receives 300 ± 1 g soil. Record the weight of soil added to each jar in grams to one decimal place (*e.g.*, 300.3 g) on the data sheet. Based on this step, ~1200 g soil will be archived from each soil horizon/layer.

    f. If 300g of soil cannot fit into the amber jars, fill the jars and weigh. Then, add additional jars until 1200g of soil is to be stored.

14. Seal the jar, ensuring that the correct lid was used, and remove any soil particles or dust on the outside of the jar. Place the jar in the TIS soil archive cabinets and record the location on the

data sheet. If the jars must be moved between rooms to get to the soil archive, they shall be placed inside a secondary sealed container (*e.g.*, cooler) to minimize the risk of spillage during transit.

15. Weigh and then dispose of excess soil, rocks, roots and other material from the sample and record the weight of disposed material on the data sheet. Clean and sterilize all sieves, trays, spoons, and other equipment that came into contact with the soil in accordance with the procedures described in NEON's USDA APHIS soil permit. Wear protective gear (gloves, eye-protection, etc) as required based on NEON lab safety training. Return all equipment to its storage location once clean/sterilized so that it is easily accessible and available when the next set of samples arrives.

16. If a sample is found whose jar label does not match its lid label, the label on the jar shall be considered correct unless there is strong evidence to the contrary. Record any label mismatches in the metadata associated with the sample(s), and indicate how the mismatch was resolved. For example: "An archive sample was found with a different label on the jar (XXXX) than on the lid (XXXO). The jar label was assumed to be correct and a new lid was added with a label matching the jar label."

11.2.4 Sample Preservation

Samples shall be stored in the TIS soil archive when not in use. Samples shall always be returned to their allocated location in the archive (*e.g.*, shelf 4, cabinet 2). This is important to ensure that samples can be found quickly and easily whenever requested.

Samples shall never be stored on the floor to minimize the risk of loss due to flooding. To minimize degradation of soil samples due to light exposure:

- Amber glass jars shall be used to store archive samples,
- Light shall be blocked from storage shelves (*e.g.*, sealed cabinets),
- Lights shall be switched off when the archive is not in use,
- Windows shall be covered with blinds or curtains.

11.2.5 Sample Shipping

The shipping procedure shall comply with NEON's USDA APHIS Permit to Receive Soil. The procedure for shipping TIS soil archive samples to people/organizations requesting them is TBD.

11.2.6 Data Handling

It is preferable that all data and metadata recorded during this procedure be entered directly into electronic form (rather than a data sheet, which must then be transcribed to electronic form), as this will eliminate one source of errors.

If physical datasheets are used, all data and metadata shall be entered in electronic form to the appropriate database (TBD) within 1 day.

11.2.6.1 Metadata

Metadata shall be stored for each soil sample in the TIS Soil Archive. The NEON metadata strategy is TBD, but it is likely that metadata for each TIS Soil Archive sample will fit into a hierarchy of NEON metadata components (Figure 1). As a result, it is not necessary to capture all metadata relating to a Soil Archive sample in its metadata, since much of that information will be captured at higher levels in the hierarchy (*e.g.*, latitude and longitude of the soil pit where the sample is collected shall be captured in the TIS Soil Pit metadata, while climate statistics shall be captured in the NEON site metadata).

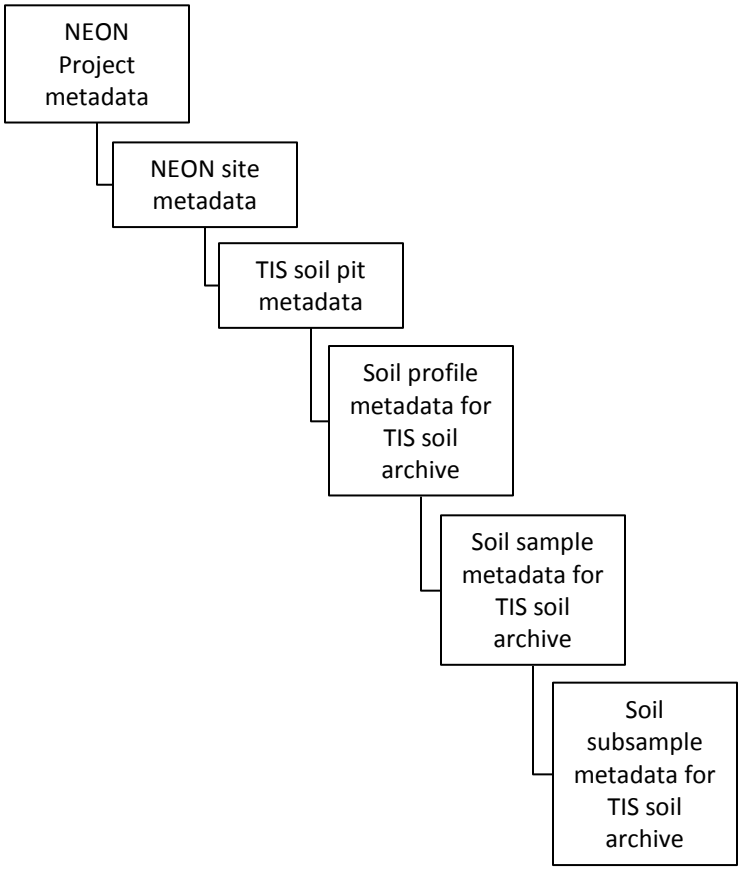

Figure 1. Metadata for the TIS Soil Archive samples fits within the hierarchy of NEON metadata. Note that NEON's metadata strategy is TBD and this figure is illustrative of an anticipated structure.

Metadata fields for the TIS soil archive are shown in Tables 1-3 and are adapted from Boone et al. (1999).

Table 3. Metadata fields for soil profile for TIS soil archive.

Metadata field	Units	Example
Sample identification code	Domain_Site_Location	9_Woodworth_Pit1
Sample description	Text	Soil samples extracted from the TIS soil pit for the TIS soil archive.
Metadata created	MM/DD/YYYY	09/11/2012

Metadata last updated	MM/DD/YYYY	05/17/2018
Extractors name(s)	Text	John Smith (NEON TIS soil pit contractor)
Extractors affiliation(s)	Text	A Government Agency
Extractors contact information	Text	john.smith@government.gov
Soil profile extraction description	Text	Soil was extracted from each soil horizon/layer from the soil surface to the bottom of the soil pit (2 m) for the TIS Soil Archive. The horizontal dimensions of the extracted soil were 0.2 m by 0.2 m, and the height was determined by the height of the soil horizon/layer. For each horizon/layer, the extracted soil was broken up and mixed by hand to homogenize the sample. A 2 kg subsample of mixed soil was placed in a labeled sealable plastic bag, which in turn was placed inside another sealable plastic bag to minimize the chance of spillage. The subsamples representing each soil horizon/layer were placed inside an insulated box and mailed to NEON headquarters. The temperature of the samples was not controlled after the soil was extracted.

Table 4. Metadata fields for soil sample for TIS soil archive.

Metadata field	**Units**	**Example**
Sample identification code	Domain_Site_Location_Top-Bottom depth (cm)_Sample_Horizon	9_Woodworth_Pit1_16-32_Archive_2Bw
Sample description	Text	Soil from 0.21 to 0.65 m (soil horizon: A) collected from the TIS soil pit.
Metadata created	MM/DD/YYYY	09/11/2012
Metadata last updated	MM/DD/YYYY	05/17/2018
Sampling depth	Centimeters	21 to 65 cm
Soil horizon/layer description	Text	A horizon
Date soil sample extracted	MM/DD/YYYY	09/02/2012
Date soil sample mailed	MM/DD/YYYY	09/02/2012
Date soil sample received at NEON headquarters	MM/DD/YYYY	09/05/2012
Date soil sample processing began	MM/DD/YYYY	09/06/2012
Date soil sample processing finished	MM/DD/YYYY	09/11/2012
Processers name(s)	Text	Edward Ayres
Processers affiliation(s)	Text	NEON
Processers contact	Text	eayres@neoninc.org

information		
Sample processing description	Text	Field moist soil was mixed by hand and spread out in a large metal tray to dry. The soil was air dried at ambient room temperature until weight varied <1 % over 24 hours. It was subsequently sieved through 2 mm mesh. Material remaining on the sieve was discarded. The sieved soil was thoroughly mixed by hand and four 300 g subsamples were placed into labeled glass jars for archiving.
Storage location	Text	NEON HQ TIS Soil Archive

Table 5. Metadata fields for soil subsample for TIS soil archive.

Metadata field	Units	Example
Subsample identification code	Domain_Site_Location_Top-Bottom depth (cm)_Sample_Horizon_Subsample	9_Woodworth_Pit1_16-32_Archive_2Bw _3
Subsample description	Text	Subsample 2 of 4
Originators name(s)	Text	Edward Ayres
Originators affiliation(s)	Text	NEON
Originators contact information	Text	eayres@neoninc.org
Metadata created	MM/DD/YYYY	09/11/2012
Metadata last updated	MM/DD/YYYY	05/17/2018
Storage location	Text	NEON HQ TIS Soil Archive, Cabinet 2, Shelf 1
Storage container	Text	Amber glass jar
Initial subsample weight	Grams	300.7 g
1 Requesters name(s)	Text	Jane Doe
1 Requesters contact information	Text	jane.doe@university.edu
1 Quantity removed	Grams	1 g
1 Date removed	MM/DD/YYYY	05/17/2018
1 Requesters research description	Text	Measured pyrolysis-molecular beam mass spectrometry profiles of soil to characterize its chemical composition.
1 Link to dataset(s)	Text	TBD, e.g., www.neoninc.org/abc123
1 Citation for publications featuring this subsample	Text	Doe J (2019) The chemical composition of soil based on pyrolysis-molecular beam mass spectrometry. Advances in Soil Ecology 1, 1-5.
Additional metadata fields relating to sample removal (added as	NA	NA

needed)		

11.2.7 Refreshing the Laboratory Supplies

Keep enough supplies to process at least 24 samples available at any time in case samples arrive unexpectedly.

11.2.8 Laboratory Maintenance, Cleaning, Storage

Prior to beginning this procedure and at the end of every day when samples are processed, all work surfaces shall be cleaned with 95 % ethanol and the floor shall be cleaned (*e.g.*, swept).

All materials shall be placed in labeled locations (*i.e.*, draws, cupboards, racks) when not in use.

12 DEFINITIONS

N/A

13 REFERENCES

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Lapenis, A. G., G. B. Lawrence, A. A. Andreev, A. A. Bobrov, M. S. Torn, and J. W. Harden. 2004. Acidification of forest soil in Russia: From 1893 to present. Global Biogeochemical Cycles 18.

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APPENDIX A Field Data Sheets

See AD[01]

APPENDIX B Lab Data Sheets

The following data sheets serve as a backup procedure for times when electronic data collection devices are not available. An ongoing digital datasheet will be kept, data should either be directly entered into this datasheet or transcribed as soon as possible.

TIS SOIL ARCHIVE DATA ENTRY SHEET - Air drying soil

Name:

	Date:											
Soil sample ID	Tray Label ID	Tray weight (g)	Tray + soil weight (g)	Tray + soil weight (g)	Soil weight (% change)	Tray + soil weight (g)	Soil weight (% change)	Tray + soil weight (g)	Soil weight (% change)	Tray + soil weight (g)	Soil weight (% change)	Notes

NOTES

Date format: MM/DD/YYYY

Weights recorded in grams to at least one decimal place

Additional sheets can be used if more space is required

Figure A1. Sample data sheet for air drying soil.

TIS SOIL ARCHIVE DATA ENTRY SHEET - Weighing out subsamples for archiving

Name:

Date (MM/DD/YYYY):

Soil sample ID	Soil subsample ID	Jar ID	Jar lid ID	Jar weight (g)	Lid weight (g)	Jar + soil weight (g)	Soil weight (g)	TIS archive cabinet #	TIS archive shelf #	Notes

NOTES

Weights recorded in grams to at least one decimal place

Additional sheets can be used if more space is required

Figure A2. Sample data sheet for subsampling soil.

APPENDIX C Considerations for implementation

If an entire sample is spilt, try to salvage parts of the sample that are least likely to have been contaminated. For example, if a subsample jar is dropped and smashes and the soil is in a pile on the floor, collect the soil from the top of the pile (*i.e.*, that does not appear to have come into direct contact with the floor). Record details of the incident in the metadata and note the quantity of sample that was salvaged.