# TOS PROTOCOL AND PROCEDURE: CORE SAMPLING FOR PLANT BELOWGROUND BIOMASS

| PREPARED BY | ORGANIZATION | DATE |
|---|---|---|
| Courtney Meier | FSU | 12/16/2016 |

| APPROVALS | ORGANIZATION | APPROVAL DATE |
|---|---|---|
| Andrea Thorpe | PROJ SCI | 1/27/2016 |
| Vladimir Aleksiev | PSE | 1/21/2016 |
| | | |

| RELEASED BY | ORGANIZATION | RELEASE DATE |
|---|---|---|
| Anne Balsley | CM | 1/28/2016 |

See configuration management system for approval history.



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# Change Record

| REVISION | DATE | ECO # | DESCRIPTION OF CHANGE |
|---|---|---|---|
| A | 03/25/2011 | ECO-00148 | Initial release |
| B | 01/20/2015 | ECO-02273 | Production release, template change, method improvements |
| C | 02/26/2015 | ECO-02702 | Migration to new protocol template |
| D | 1/28/2016 | ECO-03547 | Major changes to protocol include:<br>• All SOPs now implemented together every time protocol is executed, previously SOP D implemented 1X per site<br>• Timing information updated, and preservation of cores prior to core processing eliminated.<br>• Equipment list updates for lab work<br>• SOP C.1 sieving methods updated based on megapit sampling experience<br>• Roots from 2 cores within a clipCell are now pooled *after* weighing takes place and prior to grinding for chemical analysis / archive.<br>• “other” non-root biomass no longer quantified<br>• Method for calculating core `storageHours` now consistent with Herbaceous Biomass protocol.<br>• Updated Sample Shipment procedure (SOP F) to be consistent with Herbaceous Biomass protocol.<br>• To aid co-locating herbaceous clip and fine root coring, added maps of clip cells within plots to appendix G.<br>• References to mini-rhizotrons removed after descope. |

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# 1 OVERVIEW

## 1.1 Background

Belowground biomass represents a substantial component of the total plant biomass and plant carbon in terrestrial ecosystems, yet belowground biomass stocks and turnover remain very poorly understood both in space and in time. This is in large part due to the inherent difficulties associated with measuring plant parts that are obscured within soil. Developing a better understanding of how much belowground plant biomass there is, as well as how much of that biomass is produced and decomposed within a given year, is therefore crucial to improving our understanding of how terrestrial ecosystems respond to environmental changes. Here, we define fine roots to be roots with diameter ≤ 10 mm (Burton and Pregitzer 2008). In combination with the belowground biomass soil pit sampling conducted during site construction (RD[09]), the soil core sampling described here will enable estimation of the amount of belowground plant biomass ≤ 10 mm diameter within the same landsurface area from which NEON Tower eddy covariance data are derived; at many sites this will also be the dominant vegetation type(s).

Fine root frequency, biomass, and turnover rates differ substantially across size classes. In general, larger size classes constitute more of the biomass than smaller size classes, but larger roots also turn over much more slowly and therefore contribute less to annual belowground net primary productivity (BNPP) than fine roots do (Steinaker and Wilson 2005, Tierney and Fahey 2007). NEON will employ the most common and robust method to measure belowground biomass in both forest and grassland ecosystems: relatively large diameter (5–10 cm) cores (Tierney and Fahey 2007, Burton and Pregitzer 2008). Because large coarse roots occur infrequently in the soil, higher volume samples result in more accurate estimates of belowground biomass (Taylor et al. 2013). However, large sample volumes require a significant amount of time to sieve and sort in the laboratory. Given that time is limiting, there is therefore an inherent trade-off between the number and size of samples that must be resolved (Berhongaray et al. 2013). NEON will use a 3-inch outside diameter (66.5 mm inside diameter) soil corer for belowground biomass sampling, and samples will be cored to 30 cm depth in order to be consistent with the sampling depth used for soil biogeochemistry and microbe sampling (RD[07]). Within each clip "cell" selected for belowground biomass sampling, two 30 cm cores will be generated, for a total sample volume of 2722 $cm^3$ per clip cell. If roots up to 10 mm diameter exist at the site, sample volumes of this size should be sufficient to encounter them in the majority of soil samples (Taylor et al. 2013).

To account for differences in BNPP across the spectrum of fine root diameters, researchers typically sort roots within core samples into various size classes, and then calculate fine root production separately for each class. Following Burton and Pregitzer (2008), NEON will sort roots within each core to the following **sizeClass** categories: < 0.5 mm, 0.5–1 mm, 1–2 mm, and 2–10 mm.

Soil samples are sieved to remove soil, picked to separate roots from other organic material, and roots are then sorted to diameter size class. Picking and sorting roots is time consuming, and similar to other researchers, NEON will use a 1 cm length cutoff to limit the time spent searching for small root

fragments – i.e., root fragments < 1 cm length are ignored and discarded. However, root fragments < 1 cm length can contribute > 50% of the total root biomass in some samples (Koteen and Baldocchi 2013). To account for the biomass of root fragments < 1 cm length, NEON will employ a dilution technique on a subsample of cores every time fine root sampling occurs.

## 1.2 Scope

This document provides a change-controlled version of Observatory protocols and procedures. Documentation of content changes (i.e. changes in particular tasks or safety practices) will occur via this change-controlled document, not through field manuals or training materials.

### 1.2.1 NEON Science Requirements and Data Products

This protocol fulfills Observatory science requirements that reside in NEON's Dynamic Object-Oriented Requirements System (DOORS). Copies of approved science requirements have been exported from DOORS and are available in NEON's document repository, or upon request.

Execution of this protocol procures samples and/or generates raw data satisfying NEON Observatory scientific requirements. These data and samples are used to create NEON data products, and are documented in the NEON Scientific Data Products Catalog (RD[03]).

## 1.3 Acknowledgments

The author is grateful for time and detailed advice provided by Daniel Milchunas and Mark Lindquist at the Shortgrass Steppe LTER program. In addition, SOP D "Dilution Sampling for Fine Root Biomass Fragments" is based on the work of Koteen and Baldocchi (2013).

# 2 RELATED DOCUMENTS AND ACRONYMS

## 2.1 Applicable Documents

Applicable documents contain higher-level information that is implemented in the current document. Examples include designs, plans, or standards.

| | | |
|---|---|---|
| AD[01] | NEON.DOC.004300 | EHS Safety Policy and Program Manual |
| AD[02] | NEON.DOC.004316 | Operations Field Safety and Security Plan |
| AD[03] | NEON.DOC.000724 | Domain Chemical Hygiene Plan and Biosafety Manual |
| AD[04] | NEON.DOC.001155 | NEON Training Plan |
| AD[05] | NEON.DOC.050005 | Field Operations Job Instruction Training Plan |
| AD[06] | NEON.DOC.000914 | NEON Science Design for Plant Biomass and Productivity |
| AD[07] | NEON.DOC.014051 | Field Audit Plan |
| AD[08] | NEON.DOC.000824 | Data and Data Product Quality Assurance and Control Plan |

## 2.2 Reference Documents

Reference documents contain information that supports or complements the current document. Examples include related protocols, datasheets, or general-information references.

| | | |
|---|---|---|
| RD[01] | NEON.DOC.000008 | NEON Acronym List |
| RD[02] | NEON.DOC.000243 | NEON Glossary of Terms |
| RD[03] | NEON.DOC.002652 | NEON Level 1, Level 2, and Level 3 Data Products Catalog |
| RD[04] | NEON.DOC.001271 | NEON Protocol and Procedure: Manual Data Transcription |
| RD[05] | NEON.DOC.002135 | Datasheets for TOS Protocol and Procedure: Core Sampling for Plant Belowground Biomass |
| RD[06] | NEON.DOC.001925 | NEON Raw Data Ingest Workbook for TOS Belowground Biomass Soil Core |
| RD[07] | NEON.DOC.014048 | TOS Protocol and Procedure: Soil Physical, Chemical, and Microbial Measurements |
| RD[08] | NEON.DOC.000987 | TOS Protocol and Procedure: Measurement of Vegetation Structure |
| RD[09] | NEON.DOC.001708 | TOS Protocol and Procedure: Soil Pit Sampling for Plant Belowground Biomass |
| RD[10] | NEON.DOC.001717 | TOS Standard Operating Procedure: TruPulse Rangefinder Use and Calibration |
| RD[11] | NEON.DOC.014037 | TOS Protocol and Procedure: Measurement of Herbaceous Biomass |

## 2.3 Acronyms

| Acronym | Definition |
|---|---|
| BNPP | Belowground net primary productivity |
| OM | Organic material |

## 3 METHOD

The Standard Operating Procedures (SOPs) presented in this protocol describe tasks that, when taken together, allow estimation of plant belowground fine root biomass across four diameter size classes. These SOPs are:

- **SOP A: Preparing for Sampling.** Instructions to prepare for sampling for SOP B, SOP C, and SOP D.
- **SOP B: Soil Core Sampling in the Field.** Collecting soil core samples from peak herbaceous biomass clip harvest "cells" in the field, and recording required data and metadata.
- **SOP C: Processing Belowground Biomass Samples in the Laboratory.** Steps to wash, sieve, and separate roots ≥ 1 cm length from mineral soil and organic matter, and once roots are separated, steps to dry, weigh, grind, and sub-sample roots for chemical analysis.
- **SOP D: Dilution Sampling for Fine Root Fragments.** A sub-sampling procedure to quantify the amount of fine root biomass present in small root fragments < 1 cm length. By carrying out this SOP, it is possible to ignore root fragments < 1 cm length in SOP C while still generating accurate fine root biomass estimates, resulting in significant time savings.

Belowground biomass soil core sampling takes place in 400 $m^2$ sampling units located within Tower plots or subplots (Figure 1). Soil core sampling does not occur in Distributed or Gradient plots. In 20m x 20m Tower plots, two soil cores are sampled from one clip "cell" per bout. In larger 40m x 40m Tower plots (i.e. four 400 $m^2$ subplots per plot), soil core sampling occurs in each of the two subplots randomly assigned by Science Operations for sampling, and two soil cores are sampled from one clip cell per subplot per bout. This strategy means that:

- At sites with thirty 20m x 20m Tower plots, there will be n=60 soil core samples.
- At sites with twenty 40m x 40m Tower plots, there will be n=80 soil core samples.

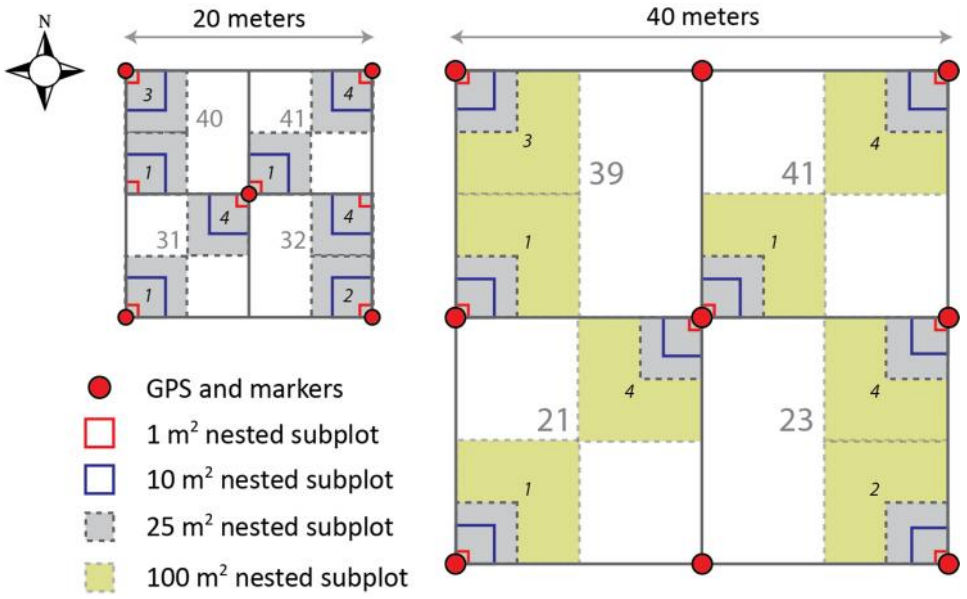


**Figure 1**. Illustration of two NEON plot sizes used for belowground biomass soil core sampling. Grey numbers indicate subplotIDs, but soil core sampling is only dependent on subplots for 40m x 40m plots. Italic black numbers show the location of nested subplots that are used for % cover and diversity measurements. Soil core sampling is prohibited within 1 $m^2$ and 10 $m^2$ nested subplots.

Within each 400 $m^2$ plot or subplot selected for belowground biomass core sampling, soil sampling locations in a given year are spatially co-located with the clip harvest "cell" used for the peak herbaceous biomass clip-harvest in that year. Clip cells are 0.5m x 3m, are sequentially numbered, and coordinates are assigned to the SW corner of a 0.1m x 2m clip-strip that is centered within each clip harvest cell. These coordinates are relative to the SW corner of the plot or subplot (i.e. the SW corner of the plot or subplot is defined as having coordinates [0,0]) (**Figure 2**, *left*). To determine soil coring locations, technicians consult a plot-specific "Clip List" to determine which clip harvest cell was (or will be) used for the peak biomass harvest in the current growing season. Within each clip harvest cell two 66.5 mm diameter (3" OD) x 30 cm length soil core samples will be harvested: one from each of the areas to the North AND South of the 0.1 m x 2 m clip-strip (**Figure 2**, *right*). To avoid roots and rocks, technicians may sample from anywhere within the North and South sampling areas shown in **Figure 2**.

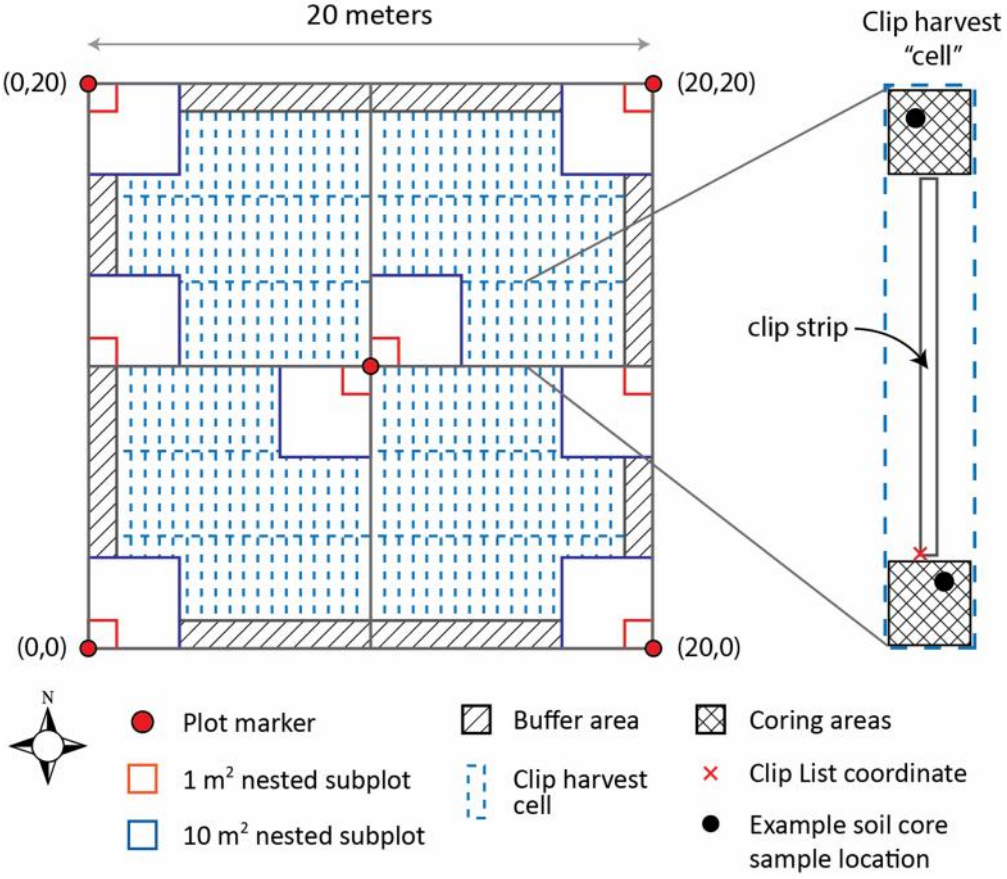


**Figure 2**. A 20 m x 20 m Tower plot showing the locations of 0.5 m x 3 m clip cells used for belowground biomass soil core sampling (*left*); the largest 25 $m^2$ nested subplot has been omitted for clarity. Within a clip cell selected for soil core sampling, one core is collected from each of the areas to the North and South of the clip-strip (*right*). The red "x" shows the coordinates provided in the Herbaceous Biomass Clip Lists.

Prior to driving the corer into the ground, crowns, corms, rhizomes, and other perennial belowground parts that are not roots are removed from the top 3 cm of soil and discarded. In some ecosystems, these non-root belowground plant parts may constitute a significant portion of the belowground biomass; however, the NEON protocol is solely focused on measuring fine root biomass.

After sampling from a given clip strip is completed, it is necessary to backfill the holes with a material approved by the site host (e.g. purchased sand, soil from another site-host approved location, etc.).

Section 4 of this document provides critical information with respect to sampling timing in both the field and the lab. Standard Operating Procedures (SOPs) in Section 7 provide detailed step-by-step directions, contingency plans, sampling tips, and best practices for implementing this sampling procedure. To properly collect and process samples, field technicians **must** follow the protocol and associated SOPs. Use NEON's problem tracking system to resolve any field issues associated with implementing this protocol.

The value of NEON data hinges on consistent implementation of this protocol across all NEON domains, for the life of the project. It is therefore essential that field personnel carry out this protocol as outlined in this document. In the event that local conditions create uncertainty about carrying out these steps, it is critical that technicians document the problem and enter it in NEON's problem tracking system.

The procedures described in this protocol will be audited according to the Field Audit Plan (AD[07]). Additional quality assurance will be performed on data collected via these procedures according to the NEON Data and Data Product Quality Assurance and Control Plan (AD[08]).

A number of protocol-specific QC checks may be used to ensure that:

- Equipment is used properly in the field
- Plant parts are sorted properly into functional groups in the field to remove non-root biomass
- Soil samples are processed in the lab according to the protocol, and
- Dried root biomass is properly weighed, ground, shipped, and tracked

# 4 SAMPLING SCHEDULE

## 4.1 Sampling Frequency and Timing

**Table 1.** Sampling frequency for belowground biomass soil core procedures on a per SOP per plot type basis.

| SOP | Plot Type | Plot Number | Sampling Events | Yearly Interval | Remarks |
|---|---|---|---|---|---|
| SOP B | Tower | All | 1X per sampling year | 5y | |
| | Distributed, Gradient | NA | NA | NA | Distributed and Gradient plots are not cored for belowground biomass. |
| SOP C | Tower | All | 1X per sampling year | Same as SOP B | SOP quantifies roots ≥ 1 cm length |
| SOP D | Tower | All | 1X per sampling year | Same as SOP B | Dilution sampling quantifies mass of root fragments < 1 cm length. |

A given sampling bout should ideally be concluded within 1 month of initiation so that the belowground standing crop does not change appreciably during the time that all target plots are sampled. This ensures that data collected across all plots within a given sampling bout are as comparable as possible.

At sites where plots may be seasonally submerged (e.g. D03 DSNY), core sampling must be timed to avoid standing water in potential soil core locations. If a plot is partially submerged but still accessible for terrestrial sampling, “cells” that contain standing water must be rejected for soil core sampling, and a new clip-location “cell” must be chosen.

After soil cores are sampled from a given clip strip, the following points are critical with respect to timing:

- Place soil core samples immediately into a cooler, and keep stored with re-usable cold packs until samples can be processed in the laboratory.

## 4.2 Criteria for Determining Onset and Cessation of Sampling

It is theoretically desirable to perform belowground biomass core sampling when the root crop is at peak biomass. However, peak belowground biomass does not necessarily correspond with peak aboveground biomass, and in some ecosystems, does not vary in a predictable manner within a growing season from year to year (Milchunas and Lauenroth 2001). Combined with the fact that belowground biomass timecourse data are unavailable for the majority of NEON sites, the timing of belowground biomass soil core sampling is guided by these two factors, listed in order of importance:

- **Date of peak biomass herbaceous clip harvest**: Perform belowground biomass soil coring either immediately before, during, or immediately after the herbaceous clip harvest associated with the greatest aboveground *peak biomass*.

- **Soil hardness**: At some sites, peak herbaceous biomass occurs during hot, dry parts of the year when soils are extremely hard and veritably impenetrable due to high clay content (e.g. D10 CPER). At sites where these conditions occur, the timing of soil core sampling may be moved to earlier in the growing season when soil moisture is more conducive to core sampling.
  - If soil hardness dictates the timing of core sampling, it is not important exactly when in the growing season sampling occurs, but once an acceptable sampling window is chosen for a given site, all future sampling within that site should be initiated within ± 2 weeks of that sampling window.

### 4.3 Timing for Scheduling Field Work and Laboratory Processing

Because root biomass continues to be biologically active after sampling, and because root structures are delicate and decompose easily, once soil samples are removed from the ground they must be kept cold at all times until they are processed in the laboratory according to SOP C. Acceptable methods include storing samples in:

- Coolers kept cold with re-usable cold packs. Cold packs should be exchanged for fresh cold packs every 12 hours.
- Refrigerator, 4–8 ˚C

Ideally, soil cores are processed in the laboratory within 24 h of collection in the field. However, it is acceptable to keep soil cores in cold storage for up to a maximum of 72 hours. Once laboratory processing is initiated on a given sample, processing should be carried all the way through without stopping.

The optimal strategy for allocating labor to field sampling and laboratory processing tasks, and completing the fine root biomass coring effort within 1 month, depends on the number of people available to complete the work:

- *Option 1:* One crew (minimum of 2 people) first performs the field sampling, then returns to the laboratory to process the cores just sampled in the field. Bear in mind that the number of consecutive field days is limited by the requirement that core samples do not remain in cold storage longer than 72 hours. Once cores have been processed in the laboratory and roots are in the drying oven, the crew returns to the field to sample more cores and repeat the cycle.

  *Option 2:* One crew performs field sampling, and another crew performs laboratory processing of cores delivered by the field crew.

### 4.4 Sampling Timing Contingencies

**Table 2.** Contingency decisions for belowground biomass fine root core sampling.

| Delay/ Situation | Action | Outcome for Data Products |
|---|---|---|
| Hours | If delay prevents sampling the second core from a given clip cell:<br>1. Bag and label first sampled core,<br>2. Place labeled bags into a cooler.<br>3. Resume core sampling in same clip cell ASAP | None |
| | If delay occurs between plots or subplots: Resume core sampling ASAP. | |
| 1-14 days | If delay prevents sampling second core from a given clip cell:<br>1. Bag and label first sampled core,<br>2. Place labeled bags into a cooler.<br>3. Process first core within 72 hours of sampling.<br>4. Resume sampling for second core in same clip cell ASAP | Increased uncertainty in belowground biomass estimates. |
| | If delay occurs between plots or subplots:<br>1. Process sampled cores within 72 hours.<br>2. Resume core sampling at additional required plots ASAP. | |
| 14+ days | If delay prevents sampling second core from a given clip cell:<br>1. Bag and label first sampled core,<br>2. Place labeled bags into a cooler.<br>3. Process first core within 72 hours of sampling,<br>4. Resume core sampling in same clip cell ASAP | Potentially substantial increases in uncertainty for belowground biomass estimates.<br><br>If delay prevents completing sampling from all plots or subplots within a 1 month window, belowground biomass may fluctuate substantially. |
| | If delay occurs between plots or subplots:<br>1. Process sampled cores within 72 hours.<br>2. Resume core sampling ASAP. | |

For QA/QC of the weighing and data entry portion of the laboratory work, select 10% of the previously dried, weighed samples for QA/QC per sampling bout. Technicians re-weigh and record the QA mass in the "qaDryMass" field of the "Lab Weighing Datasheet."

## 5 SAFETY

This document identifies procedure-specific safety hazards and associated safety requirements. It does not describe general safety practices or site-specific safety practices.

Personnel working at a NEON site must be compliant with safe field work practices as outlined in the Operations Field Safety and Security Plan (AD[02]) and EHS Safety Policy and Program Manual (AD[01]). 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.

For the field procedures, safety training is required to properly use the soil corer (e.g., use of gloves and ear plugs). Soils may contain fungi that may cause illness. Refer to the Operations Field Safety and Security Plan (AD[02]) for details on locations and appropriate precautions.

For the laboratory procedures, safety training is required to operate the grinding mill.

Additional safety issues associated with this field procedure include potential exposure to oils from roots of *Toxicodendron spp.* (discussed in Appendix F).

# 6 PERSONNEL AND EQUIPMENT

## 6.1 Equipment

The following equipment is needed to implement the procedures in this document. Equipment lists are organized by task. They do not include standard field and laboratory supplies such as charging stations, first aid kits, drying ovens, ultra-low refrigerators, etc.

**Table 3.** SOP B equipment list – Soil-core sampling belowground biomass in the field

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| **Durable Items** | | | | | | |
| MX103276 | R | Soil core sampling tube, 36” length, 3” OD | Generate soil core sample | All | 1 | N |
| MX103277 | R | Soil core drive head assembly | Works with slide hammer to drive soil core tube into soil | All | 1 | N |
| MX103278 | R | Soil core drive head pin, 3” length | Attach drive head assembly to core tube | All | 1 | N |
| MX103279 | R | Soil core quick relief bit, 3” OD* | Attach to soil core sampling tube | Standard bit for coring most soils | 1 | N |
| MX103280 | R | Soil core slide hammer, 16# | Drive sampling tube into soil | All | 1 | N |
| MX103281 | R | Soil core basket retainer, 3” adapter | Attach basket retainer system to sampling tube | Sandy soils that do not hold together | 1 | N |

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| MX103282 | R | Soil core basket retainer, 3” basket | Retain sandy soil in sampling tube | Sandy soils that do not hold together | 2 | N |
| MX103283 | R | Soil core basket retainer, 3” bit | Bit that works with basket retainer | Sandy soils that do not hold together | 1 | N |
| | S | GPS unit, pre-loaded with plot locations | Navigate to plots or subplots | All | 1 | N |
| MX100322 | R | TruPulse 360R laser rangefinder, current declination entered | Locate clip strip within a plot or subplot | Slope >20%, brushy | 1 | N |
| MX103218 | R | Foliage filter for laser rangefinder | Facilitates use of TruPulse in brushy conditions | Brushy vegetation | 2 | N |
| | R | Reflective surface (bicycle reflector or reflective tape on back of field notebook/clipboard) | Accurate location of clip strip with TruPulse in “FLT” mode | Used with TruPulse | 1 | N |
| | S | Extra battery for TruPulse (CR123A type) | Battery backup | Used with TruPulse | 2 | N |
| | R | Fiberglass meter tape (30m or longer) | Locate clip strip within plots or subplots | Plot slope <20%; grassland, savannah | 1 | N |

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| | R | Hand clippers, fine tip | Remove aboveground plant parts from soil coring location | All | 1 | N |
| MX100721 | R | Soil knife, hori-hori style | Loosen soil at surface to expose non-root plant parts | All | 1 | N |
| | R | Large chest-style cooler, with frozen cold packs | Keep core samples cool, slow down root decomposition | All | 2+ | N |
| | R | Sharpies | Label paper bags | All | 2 | N |
| MX104362 | R | Chaining pins, steel | Stretching tapes to enable location of target clip strip | Plot slope <20%; grassland, savannah | 2 | N |
| | R | Ruler or collapsible measuring stick, with 1 cm demarcations | Measure depth of the litter layer and depth of soil core bore hole | All | 1 | N |
| **Consumable items** | | | | | | |
| | R | 4”x 5” pin flags with PVC stakes | Accurate location of clip strip; PVC stakes avoid magnetic interference with compass or TruPulse | All | 6 | N |
| | R | Plastic freezer bags, 1.5 or 2 gallon | Store and organize soil core samples | All | 40+ | N |
| | R | Pencils | Record sampling metadata | All | 2 | N |

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| | R | Waterproof paper, Rite-in-the-Rain or equivalent | Material for making labels to record soil core metadata in the field | All | 10+ sheets | N |
| | R | Clip Lists (also used for Herbaceous Biomass sampling) | Identify clip cell associated with peak biomass clip harvest | All | Varies | N |
| | R | Random Tower Subplot Lists | Identify subplots for soil core sampling | Tower plots ≥ 1600 $m^2$ | Varies | N |
| RD[05] | R | Belowground biomass "Field Coring Datasheets" | Record sampling metadata | All | Varies | N |

R/S=Required/Suggested

* Bits with greater relief may be required to prevent soils with high clay content that are prone to expansion from getting stuck in the soil core tube. See http://www.soilsample.com/tooling/soiltubes.htm for available bits and soil core accessories.

**Table 4.** SOP C equipment list – Sieving belowground biomass cores, separating roots from soil organic matter, and drying root samples. Equipment listed is for 3 people working independently at a root washing station.

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| **Durable Items** | | | | | | |
| | R | Root washing station | Remove mineral soil from organic material | All | 1 | N |
| | S | 5-gallon plastic bucket | Soak core sample prior to sieving to break up cohesive clays and rehydrate roots | All | 6 | N |
| | R | Soil sieve, 2 mm stainless mesh, 8” or 12” diameter | Remove mineral soil from organic material | All | 6 | N |
| | S | Soil sive, 1 mm stainless mesh, 8” or 12” diameter | Remove mineral soil from organic material | Sandy soil sieving | 6 | N |
| | R | Soil sieve, 250 µm stainless mesh, 8” or 12” diameter | Remove mineral soil from organic material | All | 6 | N |
| | R | Rectangular enamel pan or equivalent, white (app. 30 cm x 20 cm, or 13”x 9”)† | Facilitates separating roots (which float) from mineral particles | All | 6 | N |
| | R | Forceps, blunt tip, stainless steel | Separate roots from organic material | All | 3 | N |
| | R | * Wire gauge with openings approx. 2mm, 1mm, and 0.5mm | Sort roots into size classes during sieving and picking | All | 3 | N |
| | R | Grinding mill, Wiley, 20 mesh | Grind larger fine root sample volumes | Sample masses > 750 mg | 1 | N |

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| | S | Porcelain mortar, 65 mL capacity, with pestle, | Grind smaller fine root sample volumes, avoid loss of small samples in mill | Sample masses < 750 mg | 1 set | N |
| | R | Sample microsplitter | Creates identical sub-samples from ground sample | Large root volumes | 1 | N |
| | R | Hi-back pans for sample microsplitter | 2 per splitter; receives split sub-sample | With micro splitter | 2 | N |
| | R | Sharpie, extra fine tip | Labeling envelopes and scint vials | All | 2 | N |
| | S | Toothbrush | Clean soil corer threads in field | Field | 2 | N |
| **Consumable items** | | | | | | |
| | R | Pencils | Record dry weight of root samples | All | 2 | N |
| RD[05] | R | Lab Weighing Datasheet | Record dry weight of root samples | All | Variable | N |
| | R | Scintillation vials with caps, 20 mL volume | Containers for ground split sub-samples | All | Variable | N |
| | R | Large plastic weigh boats | Weigh relatively large quantities of dried root samples | Large root quantities | 50+ | N |
| | R | Clasp envelopes, 6”x 9”, Kraft paper | Store and organize sieved roots during and after drying | Large root quantities | 50 | N |
| | R | Coin envelopes, 3⅜”x6”, Kraft paper | Store and organize sieved roots during and after drying | Small root quantities | 50 | N |

| Item No. | R/S | Description | Purpose | Conditions Used | Quantity | Special Handling |
|---|---|---|---|---|---|---|
| | R | Paper bag, 25# Kraft | Organize root samples in the drying ovens | All | 20 | N |
| | S | Small plastic weigh boats | Weigh relatively small quantities of dried root samples | Small root quantities | 50+ | N |

R/S=Required/Suggested

* Gauge 12 = 2.05 mm, gauge 18 = 1.02 mm, and gauge 24 = 0.51 mm; while not exactly the diameters desired, the gauges listed here are acceptable for this protocol.

† Note: the exact dimensions of the pan/tub are not critical, it serves as an aid for more easily spotting roots suspended in water. The only requirement is that it can safely contain liquid; a white material also makes identifying roots easier.

**Table 5.** SOP D equipment list – Dilution sampling for fine root biomass fragments < 1 cm

| Item No. | R/S | Description | Purpose | Quantity | Special Handling |
|---|---|---|---|---|---|
| **Durable Items** | | | | | |
| | R | Soil sieve, 53 µm stainless mesh, 8" or 12" diameter | Consolidate residual fraction from both cores per clip strip, rinse, and transfer to beaker for dilution | 2 | N |
| | R | Magnetic mixing plate, stir range 60 to 1200 rpm, 4 x 4 inch stirring surface | Randomize aqueous suspended residual fraction | 1 | N |
| | R | Magnetic stir bar, 2" to 3" length | Randomize aqueous suspended residual fraction | 2 | N |
| | S | Beaker, 1 L | Hold smaller volumes of aqueous suspended residual fraction | 2 | N |
| | S | Beaker, 2 L | Hold large volumes of aqueous suspended residual fraction | 2 | N |
| | S | Beaker, 4 L | Hold very large volumes of aqueous suspended residual fraction; e.g., for soils with thick O horizon | 2 | N |
| | R | Plunger, diameter approx. 1 cm less than beaker diameter | Stop mixing vortex, randomize aqueous suspended residual fraction | 1 | N |
| | R | Syringe, 40 – 60 mL, with tip cut off to make a 1 cm diameter aperture | Aspirate sub-sample from randomized aqueous residual fraction | 2 | N |
| | R | Plastic laboratory squirt bottle, filled with water | Rinse syringe following sub-sampling | 1 | N |

| Item No. | R/S | Description | Purpose | Quantity | Special Handling |
|---|---|---|---|---|---|
| | R | Aluminum weighing dishes, 65 mL (e.g. Fisher #: 08-732-102) | Hold and dry root and organic material from sub-samples. | 200 | N |
| | R | Forceps, fine tip | Pick small root fragments apart from organic material | 2 | N |
| | S | Sheet tray, baking or equivalent | Transfer aqueous samples in aluminum dishes to drying ovens | 1 | N |
| | R | Threaded Rod, 12” x ¼”, zinc | Plunger device for dilution sampling, rod | 1 | N |
| | R | Vinyl Laminate Wall Base Moulding | Plunger device for dilution sampling, plunger base | 1 | N |
| | R | Wood Dowel, 12” by ¾” diameter | Plunger device for dilution sampling, plunger handle | 1 | N |
| | R | Hex Nuts, ¼” | Plunger device for dilution sampling, fastening | 4 | N |
| **Consumable Items** | | | | | |
| None | | | | | |

R/S=Required/Suggested

**Table 6.** Equipment list – Minimizing exposure to toxic oils from roots of *Toxicodendron spp*. that may be encountered

| Item No. | R/S | Description | Purpose | Quantity |
|---|---|---|---|---|
| **Durable Items** | | | | |
| | R | Labeled clippers, dedicated to clipping *Toxicodendron spp.* (see Table 4) | Prevent spread of toxic oils to multiple clippers | 1 |
| | R | Labeled sieve set(s), dedicated to sieving samples containing *Toxicodendron.* (Set contains 2mm sieve and 250 µm sieve. See Table 4.) | Prevent spread of toxic oils to multiple sieves. | As needed |
| | R | Labeled forceps, blunt tip, stainless steel; dedicated to *Toxicodendron* samples | Prevent spread of toxic oils to multiple forceps. | As needed |
| **Consumable Items** | | | | |
| | R | Cleanser, urushiol-specific, Tecnu or equivalent (example) | Clean sieves and forceps after use. | As needed |
| | R | Cotton gloves, single use (example) | Prevent oil contact with hands while collecting cores in the field. | Box of 12 |
| | R | Disposable PPE outer-wear (example) | Prevent oil contact with skin and clothing while collecting cores in the field. | Case of 24 |
| | R | Large, single-use plastic bags (trash bag or large Ziploc-type bag) | Transport used gloves and PPE from the field and minimize toxic oil transfer. | Box |
| | R | Latex gloves, single use | Prevent hand exposure during sieving. | Box |
| | R | Envelopes, pre-weighed, labeled with envelope weight, coin or clasp type (see Table 4) | *Toxicodendron* roots never handled directly again after they are placed in pre-weighed envelope. | As needed |

R/S=Required/Suggested

### 6.2 Training Requirements

All technicians must complete required safety training and protocol-specific training for safety and implementation of this protocol as required in Field Operations Job Instruction Training Plan (AD[05]).

For the field component of this protocol, technicians must be trained in navigating to points in the field with a GPS and manual methods. Most critically, technicians must be trained to quickly identify commonly encountered types of belowground plant parts at the sites within the region of employment (e.g. crowns, corms, rhizomes, roots, etc.).

Training for both the field and laboratory work must emphasize the importance of consistent, detailed labeling of all samples. ***Improper or inconsistent labeling is the most common and problematic error associated with this work!***

### 6.3 Specialized Skills

For the field work, a minimum of 2 field technicians are required for harvesting soil cores due to weight of equipment and soil cores. Technicians must possess a demonstrated ability to identify crown material associated with perennial grasses.

For the laboratory work, technicians are required to wash, dry, weigh, grind, and sub-sample belowground biomass samples for shipment to external analytical or archive facilities.

### 6.4 Estimated Time

The time required to implement a protocol will vary depending on a number of factors, such as skill level, system diversity, environmental conditions, and distance between sample plots. The timeframe provided below is an estimate based on completion of a task by a skilled two-person team (i.e., not the time it takes at the beginning of the field season). Use this estimate as framework for assessing progress. If a task is taking significantly longer than the estimated time, a problem ticket should be submitted.

An experienced two-person team will require approximately 0.5 – 1 h to locate and delineate the target clip cell from within a given plot or subplot in the field, and extract two soil core samples from the target clip cell. In the laboratory, it requires between 0.5-3.5 h per soil core to perform wet-sieving and picking of root fragments down to 1 cm length. The exact time depends on soil type and vegetation type, and the average time required is approximately 2 h per soil core. Time requirements for the laboratory dilution method (SOP D) are based on published values, and range between 1.5-2.5 h per sample (Koteen and Baldocchi 2013).

# 7 STANDARD OPERATING PROCEDURES

## SOP A Preparing for Sampling

### A.1 Preparing for soil core sampling in the field (SOP B)

1. Make waterproof labels for tracking soil core sampling metadata in the field.
    - Cut waterproof paper (Rite-in-the-Rain or equivalent) into approx. 3”x5” rectangles.
    - Write metadata on the labels with Sharpie in the field, and place the labels inside the plastic bags with the cores.
    - The outside of plastic bags may also be labeled with Sharpie for easy visibility, but do **NOT** rely only on labeling the outside of bags; Sharpie can smear and become unreadable.

2. Using local knowledge of the soils present at the site, determine the type of soil coring bit that is required for the soil conditions at the site (i.e. the degree of relief needed inside the bit) (**Table 7**).

**Table 7**. Soil core bits and the soil types and conditions in which they should be used.

| Bit Type | Intended Soil Conditions or Soil Type |
|---|---|
| Standard taper | Dry soils |
| Quick relief | Clay/Loam soils (i.e. “typical soils”); relief inside bit allows for moderate expansion of core inside soil core tube, prevents sample from getting stuck |
| Heavy duty quick relief | Heavy clay soils; additional relief allows for additional expansion of core inside soil core tube |
| Extra heavy duty quick relief | Extra heavy expansive clay soils; allows for maximal expansion of core inside soil core tube |
| Basket retainer bit | Works with basket retainer and basket retainer adapter to retain sandy, non-cohesive soil samples inside the soil core tube |

3. Prepare equipment and material according to **Table 8** below.

**Table 8.** Actions required to prepare equipment and materials for belowground biomass soil core sampling in the field (SOP B). Equipment listed here are only those items that require preparation actions before sampling; the full equipment list is provided in **Table 3**.

| Item Description | Action(s) |
|---|---|
| GPS unit | • Charge<br>• Load target plot locations |

| Item Description | Action(s) |
|---|---|
| Compass, mirror-sight, adjustable declination | Check/set correct declination* |
| TruPulse 360R laser rangefinder and clinometer | • Check battery, charge (if possible)<br>• Clean lenses with lens cloth or lens tissue (if necessary)<br>• Check/set correct declination*. See RD[10].<br>• Calibrate tilt-sensor (only necessary after severe drop-shock; see RD[10]). |
| 3" OD (66.5mm ID) soil core tube and bit assembly | Measure 30 cm from the bottom of the bit, and mark on the tube with electrical tape. |
| Re-usable cold packs | Place in -20 ˚C freezer |
| Freezer, -20 ˚C | Clear sufficient space to freeze core samples after field sampling, and temporarily store until laboratory processing takes place. |
| Hand clippers | Clean and sharpen blades (if necessary) |
| Sand, or other site-specific material | Check with the Site Host to determine the desired back-fill material. Ensure supply is sufficient for backfilling soil core holes. |
| Belowground biomass core "Field Coring Datasheet" | Print as needed on waterproof copy paper |
| Herbaceous biomass Clip Lists | Print as needed on waterproof copy paper |
| Belowground biomass core "Random Subplot List" | Print as needed on waterproof copy paper |

* Declination changes with time and should be looked up annually per site: http://www.ngdc.noaa.gov/geomag-web/

### A.2 Preparing for processing soil cores in the laboratory (SOP C)

1. Empty and clean root washing station sediment traps.
2. Clear space in drying oven for drying root samples.
    a. Set oven 1 temperature to 65°C.
    b. Set oven 2 temperature to 105°C.
3. Print lab weighing datasheets as necessary.

### A.3 Preparing for dilution sampling for fine root fragments (SOP D)

| Item Description | Action(s) |
|---|---|
| Dilution Sampling Plunger | • Assemble plunger from items listed in **Table 5.** SOP D equipment list – Dilution sampling for fine root biomass fragments < 1 cm |

1. Assemble a plunger (**Figure 3**), with diameter suitable for the size of beaker selected from **Table 5**; plunger pieces can be assembled from locally available hardware store parts.

a. Use scissors or a utility knife to cut a circular section out of a piece of vinyl laminate wall base. The diameter of the circle should be approx. 1 cm less than the diameter of the beaker.
b. Create a small hole in the center of the cut out vinyl laminate wall moulding just large enough (1/4") to fit the threaded rod zinc rod through.
c. Tighten on one nut <1" from the bottom. Then slide the cut moulding on and fasten with another nut.
d. Drill a ¼" hole completely through the wooden dowel and cut length to a preferred size.
e. Repeat step 3 to attach the dowel using two nuts.

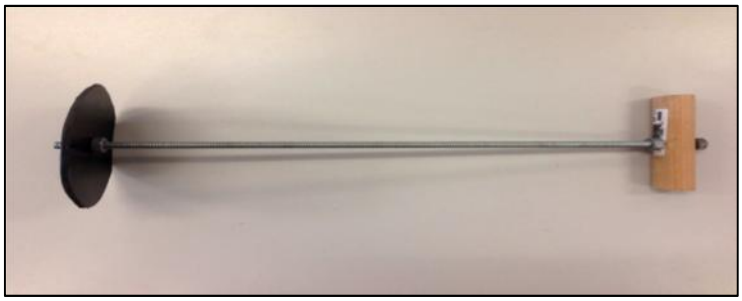

**Figure 3**. Assembled plunger used to randomize root fragment samples < 1 cm length as part of dilution sampling (SOP D).

2. Label aluminum weigh tins with unique tinIDs.

3. Print lab dilution datasheets as necessary.

**SOP B Soil Core Sampling in the Field**

1. Navigate to the plot or subplot to be sampled.
2. Use the plot or subplot-specific Clip List to identify the clip cell that was (or will be) used for the peak herbaceous biomass clip harvest in the current year. A pin flag may be left behind at the SW corner of the clip strip to aid co-location across protocols.
   - The Clip List provides the randomized list of potential clip cells per plot or subplot.
   - Coordinates provided for each clip cell correspond to the SW corner of the clip-strip – i.e. the area from which herbaceous biomass is harvested (Figure 2).
   - The Clip List indicates which clip cells have already been harvested or rejected; on the Clip List, mark cells selected for Belowground Biomass Core sampling with **status** = 5.
   - If the desired peak biomass clip cell is submerged by standing water, reject and work down the Clip List to choose an acceptable clip cell, and record "peak biomass cell submerged" in the "remarks" field of the "Field Coring Datasheet."
3. Locate the relative offsetEasting and offsetNorthing coordinates of the SW corner of the clip-strip within the target clip "cell". The procedure used to locate the offsetEasting coordinate depends on the value of the relative offsetNorthing coordinate:

*If the 'offsetNorthing' coordinate is < 10:*

a. Run a tape East/West along the south edge of the plot or subplot between the (0,0) → (20,0) plot markers (Figure 2), and stretch the tape taut.*
b. Place a pin flag at the desired relative X-coordinate.
c. Standing directly over the pin flag that was just placed, use the TruPulse in **HD** mode with a reflective surface to locate the Y-coordinate.
   - Make sure the azimuth is 0˚ (True North) when shooting the TruPulse to find the Y-coordinate (see RD[10] for detailed instructions for operating the TruPulse).
d. Place a pin flag at the clip-strip (X,Y) location – i.e. the SW corner of the clip-strip.

*If the 'offsetNorthing' coordinate is > 10:*

a. Run a tape* East/West from the plot or subplot centroid (10,10) to either the (0,10) position or the (20,10) position (Figure 2).

| offsetEasting coordinate | Tape Layout[1] |
|---|---|
| 1 < X < 10 | From (10,10) → (0,10) |
| 10 < X < 20 | From (10,10) → (20,10) |

[1] Use the TruPulse in **AZ** mode to guide the tape along the correct azimuth.

b. Place a pin flag at the desired relative offsetEasting coordinate.
c. Standing directly over the pin flag that was just placed, use the TruPulse in **HD** mode with a reflective surface to locate the Y-coordinate.

- Make sure the azimuth is 0˚ (True North) when shooting the TruPulse to find the Y-coordinate (see RD[10] for detailed instructions for operating the TruPulse).

d. Place a pin flag at the SW corner of the Clip Strip.

## TIPS

- If the plot slope is > 20%, or there is significant brush or obstacles that prevent accurately stretching a tape, the TruPulse laser rangefinder can be used in **HD** mode to place the initial pin flags relative to the plot markers.
- Plot slope can be quickly estimated using the inclinometer in the TruPulse (**INC** mode).

4. Mark the four corners of the South soil core sampling area within the clip strip "cell" with pin flags to delineate where the first of the two soil cores should be harvested (Figure 4).
   a. Place pin flag "A" 20 cm to the west of the coordinates provided in the Clip List (i.e. the red "x" in Figure 4) – use a meter tape or ruler to be accurate.
   b. Place pin flag "B" 50 cm to the east of pin flag "A"
   c. Place pin flag "C" 50 cm to the south of pin flag "A"
   d. Place pin flag "D" 50 cm to the south of pin flag "B"
   e. Domain staff may also pursue building a 50 cm by 50 cm square frame from small diameter PVC piping to reduce pin flag setup time.

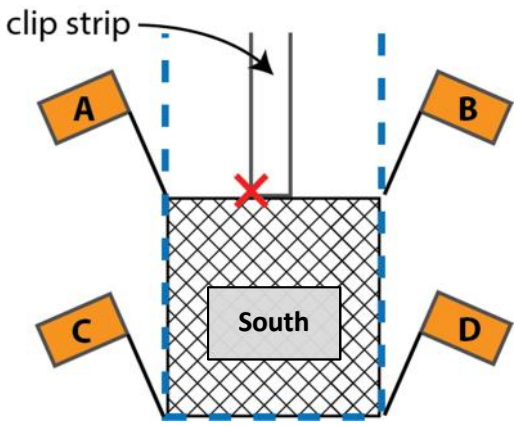


**Figure 4**. Delineating the South soil core sampling area (cross hatched) within a clip "cell" (dashed blue lines) with pin flags. The clip-strip (black lines) lies immediately to the north of the South soil core sampling area, and the red "x" marks the coordinates provided in the Herbaceous Biomass Clip List.

5. Mark the four corners of the North soil core sampling area within the clip strip "cell" with pin flags to delineate where the second of the two soil cores should be harvested (Figure 5).
   a. Place pin flag "E" 2 m to the north of pin flag "A"
   b. Place pin flag "F" 2.5 m to the north of pin flag "A"
   c. Place pin flag "G" 2 m to the north of pin flag "B"
   d. Place pin flag "H" 2.5 m to the north of pin flag "B"

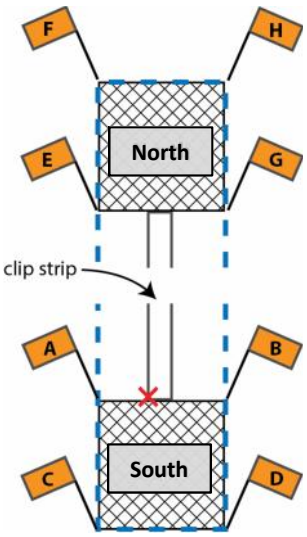


**Figure 5.** Delineating the North soil core sampling area with reference to the previously delineated South soil core sampling area (cross hatched) within a clip "cell" using pin flags (dashed blue lines indicate the clip cell boundary). The middle of both the cell and the clip-strip have been omitted for clarity.

6. For each of the 'N' and 'S' soil core sampling areas:
   a. Create a label on waterproof paper with the information below. The label and the core will then be placed in a large plastic freezer bag.
      - **coreDate**, *YYYYMMDD* format
      - **plotID** and **subplotID**,
        for 20m x 20m plots, subplotID = 31
        for 40m x 40m plots, subplotID = 21, 23, 39, or 41
      - **clipCellNumber**, the 3 digits to the right of the last "_" in the clipID on the Clip List
      - **coreID**, *N* or *S*
   b. Record on the "Field Coring Datasheet":
      - **plotID**
      - **subplotID**
      - **clipCellNumber**
      - **coreID,** *N* or *S*
      - **ltrDepth**, average litter depth for '*N*' or '*S*' core sampling area; nearest 1 cm
        - If litter depth is < 1 cm in depth, write "0.5" in **ltrDepth**

- **wstDist10cm**, distance to closest woody stem with DBH ≥ 10 cm; nearest 0.5 m
  - The closest qualifying woody stem > 10 cm DBH may occur outside of the delineated plot; record the distance irrespective of plot boundaries
  - If there are no qualifying woody stems within 20 m of the coring location, record "NA" in the **wstDist10cm** field
- **wstDist1cm**, distance to closest woody stem with 1 cm ≤ DBH < 10 cm; nearest 0.5 m
- **bareGround**, % of '*N*' or '*S*' core sampling area that is made up of soil (particles < 5 mm diameter) and / or rock (mineral particles > 5 mm diameter); nearest 10%)

7. Assemble the soil core tube, bit, retainer basket (if necessary), and drive head (see Appendix E).
8. Within the targeted soil core sampling area, determine the exact location from which the soil core will be harvested
   a. To avoid rocks and roots that may interfere with coring, probe the ground within the target sampling area with a chaining pin or pin flag to determine a suitable location.
   b. Use hand clippers to remove aboveground plant leaves and stems from the exact 3-inch diameter area to be cored, and remove litter down to the soil surface.
   c. Clip all vegetation down to the soil surface.
9. For each core, remove non-root belowground plant parts from the top 3 cm of soil:
   a. Score the ground with the soil core bit so it is clear exactly where the soil will be cored.
   b. Loosen the soil with a soil knife, and remove the soil from around any perennial non-root plant parts growing within the scored area (e.g. corms, rhizomes, crowns, biological soil crust, etc.).
      - If perennial graminoid crowns are present, remove soil until the transition from crown to root is visible.
      - If biological soil crust is present, score the soil just below the moss/lichen layer and carefully remove the crust. The crust can be placed back over the coring hole on top of the backfill material.
   c. Clip all non-root material from within the bit-scored area, and discard.
10. For each sampling area ('N' and 'S'), harvest one 66.5 mm ID (3-inch OD) soil core sample to 30 cm maximum depth:
    a. Position the soil core bit back over the scored area, and make sure the soil core assembly is vertical. If the plot is sloped, the soil core assembly should still be vertical.
    b. Use the slide hammer to pound the soil core tube to 30 cm maximum depth (*which should be marked on the soil core tube with electrical tape or similar*).

**Once the soil corer is in the ground, do not turn the unit counter-clockwise, as this will unscrew the bit from the core tube underground, resulting in loss of the bit.**

    c. Push the core tube back and forth sharply several times to loosen it within the soil profile.

d. Remove the core tube from the ground, and carefully extract the core into a labeled plastic bag.

**TROUBLESHOOTING**

- If obstacles are encountered that prevent coring to 30 cm depth, a minimum core depth of 20 cm is acceptable.
- If a minimum 20 cm depth core cannot be obtained, select another location from within the target 'N' or 'S' sampling area, but do not attempt more than 3 alternate locations.
- If a minimum 20 cm depth core cannot be sampled within the target 'N' or 'S' sampling area, record "20 cm coreDepth not achieved" in the **remarks** field of the 'Field Coring' datasheet, and collect a core with the greatest depth possible.
- If no core can be obtained from a representative cell, record `coringPossible = N' in the **remarks** field, and move on to the next plotID or subplotID on the list.

11. Place the core into the cooler immediately for cold storage until cores can be processed in the laboratory.
    - Remember to refresh cold packs every 12 h or transfer cores to a refrigerator in the lab.
12. Measure the average depth of the bore hole, and record on the "Field Coring Datasheet":
    - **coreDepth**, the total depth of the core; nearest 1 cm
    - **time**, the time the core is placed into the cooler in the field; *HHmm,* 24-h format
    - **remarks**, e.g. "20 cm coreDepth due to root"
13. Backfill the bore hole with site-host approved material.

## SOP C Processing Belowground Biomass Samples in the Laboratory

***Overview***

Fine root samples generated from this procedure are analyzed for isotopes ($^{13}C$ and $^{15}N$); as such, disposable latex or nitrile gloves are required during sieving, sorting, and grinding tasks to prevent contamination of the sample with your hands.

Soil cores must be sieved in the laboratory within 72 h of sampling in the field. Use time estimates for lab processing steps provided in Section 4 to plan field work so that a backlog of cores does not develop, and the 72 h requirement can be met. Time sensitive processing steps include:

1. Wash and sieve soil cores to separate mineral soil from root biomass and organic matter, and separate roots ≥ 1 cm length from the residual fraction. The residual fraction is defined as root fragments < 1 cm length and non-root soil organic matter.
2. Set aside the residual fraction from a random subset of 20 cores for processing with SOP D.
    - See SOP D, step (1) for guidance on randomly selecting cores for dilution sampling.
3. Dry fine root biomass ≥ 1 cm length to constant weight.

Once roots are dry, time is no longer of the essence, and the following may be completed as time allows:

4. Weigh and record dry weight biomass.
5. Grind fine root samples for chemical analyses.

### C.1 Sieving soil cores for fine root biomass

Prepare a soil core for sieving:

1. If the soil cores have a large amount of root mass, finely textured soils, or the soil is difficult to break apart by hand without fragmenting roots, wet-sieving may be the most efficient procedure for separating roots from soil; follow instructions in the wet sieving section below.
2. If the soil cores have little root mass, are coarsely textured, or the roots are very brittle then dry-sieving soils may be the most efficient procedure; follow instructions in the dry sieving section below.

The goal for the sieving procedure is to isolate fine roots and sort to sizeClass. Non-root material encountered during sieving are discarded (rhizomes, corms, bulbs, perennial graminoid crowns, etc.).

#### C.1.1 *Wet Sieving Soils*

1. For wet sieving:
    - Soak the core in water for 1 hour in a 5 gallon plastic bucket to facilitate breaking up clays. Water depth should be sufficient to cover the core.

2. For each soil core sample, label up to 8 coin envelopes with the information below. You will not need all 8 envelopes if all **status** x **sizeClass** combinations are not present in the core sample. If there is a large amount of root biomass within a given size class, use a clasp envelope instead.
   - **coreDate**, date roots were sampled in the field; *YYYYMMDD* format
   - **plotID** and **subplotID**,
     for 20m x 20m plots, subplotID = 31;
     for 40m x 40m plots, subplotID = 21, 23, 39, or 41
   - **clipCellNumber**, the 3 digits to the right of the last "_" in the clipID on the Clip List
   - **coreID**, either '*N*' or '*S*'
   - **status,** '*Live*' or '*Dead*'
   - **sizeClass** (<0.5, 0.5-1, 1-2, 2-10)
3. Use a 250 µm sieve, a white enamel pan, and the root washing station to begin separating roots and organic material from mineral soil particles in the bucket.
   a. Massage the sample in the bucket with gentle manual pressure to break up large aggregates and organic matter pieces.
   b. Thoroughly mix the slurry in the bucket by hand to separate small roots from mineral soil particles. At this point, roots and small pieces of organic material (OM) should be floating on the surface.
   c. Remove and sort floating roots from the surface of the slurry.
      i. Skim the surface of the slurry with the 250 µm sieve, then rinse the sieve contents with the root washer.
      ii. Transfer the sieve contents to the enamel pan by inverting the sieve over the pan and rinsing with the root washer nozzle. Be careful not to overfill / overflow the pan!
      iii. In the pan, pick and separate root fragments from organic material, sorting to **status** and **sizeClass** combinations as you go.
4. Separate fine roots remaining in the bucket from mineral soil and organic matter. Use the root washing station, a 2 mm sieve, a 250 µm sieve, and a white plastic or enamel tray.
   a. Pass no more than 10%-20% of the slurry through the top of the sieve stack.
      - You must avoid overloading / overflowing the 250 µm sieve.
   b. Wash fine mineral soil particles through the sieve stack using the root washer nozzle; mineral soil particles > 250 µm diameter, roots, and organic matter should be retained in both sieves.
      - Break up aggregates and organic matter pieces using gentle manual pressure.
      - BE CAREFUL NOT TO OVERFLOW THE 250 µm SIEVE!
   c. Manually remove larger rocks from the top of the 2 mm sieve – but don't spend more than several minutes.

d. Turn the 2 mm sieve upside down over one of the enamel pans, and use the root washer nozzle to transfer material from the 2 mm sieve to the pan. Roots often float, and mineral particles sink.

e. Place the 2 mm sieve back on top of the 250 µm sieve, and decant the sample from the enamel pan back through the sieve stack, retaining mineral particles in the enamel pan.

f. Discard mineral particles retained in the enamel pan, and rinse the pan.

g. Repeat (d)-(f) until enough mineral particles have been removed from the sample that it is possible to begin picking ≥ 1 cm root fragments from remaining organic matter.

h. Use forceps to pick all roots ≥ 1 cm length from the enamel pan, sorting to **sizeClass** and **status** either as you go; alternatively, you may sort to **sizeClass** and **status** after all roots > 1 cm in length have been picked.
   - Use a wire gauge to determine the **sizeClass;** the largest diameter of a root fragment should be used to classify the size.
   - If only part of a root is alive, categorize the entire root as "live." Live roots are most readily distinguished from dead roots on the basis of color and friability; dead roots are often dark brown or black and brittle, while live roots are often lighter in color and flexible – i.e., they can typically be bent into a "U" shape without breaking.

i. Place sorted roots into the pre-labeled envelopes created in step (2).

j. Rinse out the enamel pan, and repeat steps (d) to (i) for the 250 µm sieve.

k. **If the sample has been randomly selected for dilution sampling**, set aside the residual fraction (i.e., root fragments < 1 cm mixed with organic material) for processing via SOP D.
   - See SOP D, step (1) for guidance on randomly selecting cores for dilution sampling.

l. Clean the 250 µm sieve, mix the remaining slurry in the bucket by hand, and repeat all of step (4) until the entire sample has been processed through the sieve stack.

5. Thoroughly clean the sieves and enamel pan with water between core samples.

6. Check sediment traps in the root washing station; if traps are full, dispose of sediment in an approved receptacle.

7. Gather samples from the same core together to keep them organized. For example:
   - Place envelopes containing root samples into a paper bag to keep samples organized (lunch sack size works well); OR
   - If there are very few roots, coin envelopes may be paper clipped together.

### *C.1.2 Dry Sieving Soils*

1. Process soils using the different sized sieves as you would with the wet-sieving procedure, but do not apply water.
2. Use a 2 mm sieve, a 250 µm sieve, and a pan bottom. Work your way through the various sieves to pick out roots > 1 cm in length.
    a. The 2 mm sieve is useful for catching and removing large rocks from the sample, as well as larger roots
    b. The 250 µm sieve is useful for capturing any roots that have passed through the 2 mm sieve. Roots > 1 cm in length are not likely to pass through this finer mesh.
    c. The white pan can be used to more easily differentiate small roots in the 250 µm soil fraction.
3. For each soil core sample, label up to 8 coin envelopes with the information below. You will not need all 8 envelopes if all **status** x **sizeClass** combinations are not present in the core sample. If there is a large amount of root biomass within a given size class, use a clasp envelope instead.
    - **coreDate**, date roots were sampled in the field; *YYYYMMDD* format
    - **plotID** and **subplotID**,
        - for 20m x 20m plots, subplotID = 31;
        - for 40m x 40m plots, subplotID = 21, 23, 39, or 41
    - **clipCellNumber**, the 3 digits to the right of the last "_" in the clipID on the Clip List
    - **coreID**, either '*N*' or '*S*'
    - **status,** '*Live*' or '*Dead*'
    - **sizeClass** (<0.5, 0.5-1, 1-2, 2-10)
4. Separate fine roots remaining in the bucket from mineral soil and organic matter.
    a. It is often helpful to pass no more than 10 – 20% of the soil through the sieve stack, as it makes it easier to spot roots.
    b. Break up aggregates and organic matter pieces using gentle manual pressure.
    c. Manually remove larger rocks from the top of the 2 mm sieve – but don't spend more than several minutes.
5. Use forceps to pick all roots ≥ 1 cm length from the enamel pan, sorting to **sizeClass** and **status** either as you go; alternatively, you may sort to **sizeClass** and **status** after all roots > 1 cm in length have been picked.
    a. Use a wire gauge to determine the **sizeClass;** the largest diameter of a root fragment should be used to classify the size.
    b. If only part of a root is alive, categorize the entire root as "live." Live roots are most readily distinguished from dead roots on the basis of color and friability; dead roots are often dark brown or black and brittle, while live roots are often lighter in color and flexible – i.e., they can typically be bent into a "U" shape without breaking.

c. Place sorted roots into the pre-labeled envelopes created in step (2).
d. **If the sample has been randomly selected for dilution sampling**, set aside the residual fraction (i.e., root fragments < 1 cm mixed with organic material) for processing via SOP D.
e. Clean the 250 µm sieve and repeat all of step (4) until the entire sample has been processed through the sieve stack.

6. Once all roots >1 cm in length have been picked and sorted, wash sediment from roots by using a clean 250 um sieve. Sediment clinging to roots can significantly inflate weighed root biomass; thus the importance of gently washing dry roots once they are sieved.
    a. Place a sorted group of roots into the 250 um sieve and gently run water over the roots.
    b. Use forceps to transfer the roots to a labeled coin envelope.
    c. Repeat the above steps (a-b) for the remaining root samples.
7. Thoroughly clean the sieves and enamel pan with water between core samples.
8. Gather samples from the same core together to keep them organized. For example:
    - Place envelopes containing root samples into a paper bag to keep samples organized (lunch sack size works well); OR
    - If there are very few roots, coin envelopes may be paper clipped together.

### C.2 Drying, weighing, and processing belowground biomass samples

Washed roots should be placed in the drying oven as soon as possible following sieving.

1. Label each 25# bag containing washed root samples with the date and time it is placed in the drying oven.
    - These data are the ovenInDate and time required during data entry.
    - ***Critical step:*** Labeling bags allows assessment of how long different batches of bags have been in the oven, especially when roots sampled on different days occupy the same oven.
2. Place labeled 25# bags into a drying oven for a minimum of 48 h (longer is okay, but not required).
    - Dry all root diameters at 65 ˚C.
3. Remove bags of dried biomass from the drying oven, and label with **ovenOutDate** / Time.
    - Dried plant material should be weighed immediately after removing from the drying oven, as it will absorb moisture from the air if left in ambient room conditions (particularly in humid environments).
        - If using this method, it is helpful to remove bags from the oven and weigh one at a time.

- Dried samples may also be stored for up to 30 days in ambient room conditions prior to weighing. Samples treated in this manner must be returned to the drying oven for 24 h prior to weighing, and must be weighed as above after removal from the oven.

Group all samples from the same **plotID**, and weigh each fine root sample using a mass balance (minimum 0.01 g accuracy) and a weigh boat. Grouping samples in this step will greatly facilitate assigning **poolID** and **subSampleType** in SOP C.4 (see

4. **Table 9**).
    - Record **dryMass** on the 'Lab Weighing' datasheet; nearest 0.01 g, plant material ONLY (without the bag).
    - For large quantities of biomass that do not readily fit into a large weigh boat, use the following strategies:
        - Use a large plastic tray (or equivalent) instead of a weigh boat.
        - Crush or chop the biomass to reduce volume so it will fit into a weigh boat.
        - *Avoid splitting the biomass into subgroups for weighing, as uncertainty values must be added each time a subgroup is created.*
    - For very small quantities of biomass, use a microbalance (0.0001 g accuracy) and a small weigh boat.
5. Record required metadata for the sample in the 'Lab Weighing' datasheet.
    - **coreDate**, date fine roots were sampled in the field
    - **plotID**, unique ID of the sampled plot
    - **subplotID**, unique ID of the sampled subplot
    - **clipCellNumber**, the last three digits of the sampled clipID
    - **coreID**, either '*N*' or '*S*'
    - **ovenInDate/ Time**, date and time sample was placed in drying oven; 24 h format
    - **ovenOutDate/ Time**, date and time sample was removed from drying oven; 24 h format
    - **status**, '*live*' or '*dead*'
    - **sizeClass**, diameter category of the sorted sample; <0.5, 0.5-1, 1-2, or 2-10 mm
6. Once all masses have been recorded for a given sampling bout, QA will be performed on a subset of samples (SOP C.3), or return dried fine roots to temporary storage at ambient conditions. Samples in temporary storage can then be weighed for QA as time permits.

### C.3 Data Quality Assurance

To quantify uncertainty associated with weighing dried biomass, a portion of dried samples are re-weighed by a different technician than the person who originally weighed the biomass.

1. For each once in 5 years sampling event at a given site, select 10% of dried, previously weighed samples for re-weighing.
    - If QA weighing does not occur within several hours of the initial weighing, return the selected samples to the drying oven for 24 h prior to QA weighing. In humid environments, samples will pick up moisture from the atmosphere (especially bryophytes).
2. Record QA weight data to the nearest 0.01 g in the **qaDryMass** field of the 'Lab Weighing' datasheet.

## C.4 Grinding Fine Root Biomass for Archive and Chemical Analysis

***Overview***

Once QA masses have been recorded, samples with **status** = 'live' must be processed for archive and chemical analysis; samples with **status** = 'dead' may be discarded once data have been successfully entered to the NEON database and have passed automated QC checks.

To create a composite root sample for grinding, roots within the same **sizeClass** are pooled across the two 'N' and 'S' cores that originate from the same **clipCellNumber**. This means a maximum of 4 pooled root samples are ground per unique **clipCellNumber** (one pooled, ground sample for each **sizeClass**).

*Procedural steps:*

1. Assign roots with **status** = 'live' from the same **sizeClass** and **clipCellNumber** an incremented **poolID**, and record in the **poolID** field on the Lab Weighing data sheet. This step is critical and allows automatic creation of a sampleID that is used to track the pooled sample through the external analysis procedure.
    a. Begin by considering the cells cored for a given **plotID**. For a 20 m x 20 m plot, there will be one cell per **plotID**, and there will be two cells per **plotID** for a 40 m x 40 m plot.
    b. Within each **plotID**, for each unique combination of **clipCellNumber** and **sizeClass**, begin with **poolID** = 1, and assign an incremented **poolID** on the Lab Weighing data sheet (see **Table 9** for an example).

       If the total pooled mass within a given **sizeClass** is < 250 mg, enter **poolID** = 'NA'.

    c. Move to samples from the next **plotID**, and again assign **poolIDs**, beginning with **poolID** = 1.

**Table 9**. Example data sheet showing assignment of poolIDs to roots of the same sizeClass that originate from the same clipCellNumber. Some data sheet fields have been omitted for clarity.

| plotID | subplotID | clip CellNum | coreID *N, S* | status *LIVE, DEAD* | sizeClass *<0.5, 0.5-1, etc.* | dryMass *0.01 g* | subSample Type(s) *C, A* | poolID *1, 2, etc.* |
|---|---|---|---|---|---|---|---|---|
| SITE_042 | 21 | 167 | N | live | 2-10 | 2.56 | C, A | 1 |
| SITE_042 | 21 | 167 | N | dead | 2-10 | 0.52 | NA | NA |
| SITE_042 | 21 | 167 | N | live | 1-2 | 1.47 | C, A | 2 |
| SITE_042 | 21 | 167 | N | live | 0.5-1 | 0.33 | C, A | 3 |
| SITE_042 | 21 | 167 | N | live | <0.5 | 0.15 | NA | NA |
| SITE_042 | 21 | 167 | S | live | 2-10 | 0.00 | C, A | 1 |
| SITE_042 | 21 | 167 | S | live | 1-2 | 0.55 | C, A | 2 |
| SITE_042 | 21 | 167 | S | dead | 1-2 | 0.15 | NA | NA |
| SITE_042 | 21 | 167 | S | live | 0.5-1 | 0.63 | C, A | 3 |
| SITE_042 | 21 | 167 | S | live | <0.5 | 0.06 | NA | NA |
| SITE_042 | 39 | 573 | N | live | 2-10 | 0.21 | C | 4 |
| SITE_042 | 39 | 573 | N | live | 1-2 | 1.66 | C, A | 5 |
| SITE_042 | 39 | 573 | N | live | 0.5-1 | 0.16 | C | 6 |
| SITE_042 | 39 | 573 | N | live | <0.5 | 0.72 | C, A | 7 |
| SITE_042 | 39 | 573 | S | live | 2-10 | 0.45 | C | 4 |
| SITE_042 | 39 | 573 | S | live | 1-2 | 0.75 | C, A | 5 |
| SITE_042 | 39 | 573 | S | live | 0.5-1 | 0.26 | C | 6 |
| SITE_042 | 39 | 573 | S | live | <0.5 | 0.35 | C, A | 7 |

2. Based on the total mass of each ***pooled*** sample, decide whether enough sample is available for chemical analysis only, or whether sufficient mass is available for both chemical analysis and archive (see **Table 9** for an example).
    - If total sample mass is < 250 mg:
        - No further processing is required. The sample will not be sent for chemical analysis or archive.
        - Record **subSampleType** = '*NA*' on the 'Lab Weighing' datasheet.
    - If total sample mass is ≥ 250 mg but < 750 mg:
        - The sample will be subsampled for chemical analysis only.
        - Record **subSampleType** = 'C' on the 'Lab Weighing' datasheet
    - If total sample mass is ≥ 750 mg:
        - The sample will be subsampled for chemical analysis and archive.
        - Record **subSampleType** = 'C, A' on the 'Lab Weighing' datasheet
3. Enter the completed 'Lab Weighing' datasheet into the Data Entry webUI.
    - The webUI will generate a .csv as part of the data entry process.
    - Review the .csv for data entry errors.
    - Save a local working copy of the .csv file.

4. Copy the **subSampleID** information automatically generated in the .csv file into a label printing template. Label 20 mL plastic or glass scint vials with pre-printed labels.
    - Do not create pre-printed labels when **subSampleType** = '*NA*'
5. Grind oven-dried biomass.
    - If total sample mass is < 750 mg, use a mortar and pestle to grind the sample; this technique mitigates mass loss that occurs during milling.
        - ******Optional***: To greatly enhance grinding efficiency, add a small amount of liquid nitrogen to the mortar, and then grind the bubbling slurry.
    - If total sample mass is > 750 mg, use a grinding mill.
6. When **subSampleType** = 'C, A', mix the ground sample thoroughly with a spatula, and use an appropriately sized splitter or microsplitter to generate two representative subsamples to fill two pre-labeled 20 mL scint vials.
    - For samples with < 750 mg total mass, split the sample with a spatula instead of a microsplitter, and use the information in step (2) to allocate material to the 'C' and (possibly) 'A' subsamples.
    - If there is >> 1 g of sample, but not enough to fill two 20 mL scint vials, split evenly between two vials.
    - When **subSampleType** = 'C', only one pre-labeled scint vial is required.

## BEST PRACTICE TIPS

- If the sub-sample is too large to fit into the vial in its entirety, continue splitting until a sub-sample of the desired size is generated.
- DO NOT create sub-samples with a scoopula or spatula unless the sample is < 750 mg (see step 6 above). These tools should typically only be used to transfer an ENTIRE sub-sample into a vial.

7. Create separate chemical analysis and archive inventory sheets, for the 'C' and 'A' subsamples respectively, for shipment to external facilities.
    - Copy and paste the list of subsampleIDs from the .csv generated during webUI data entry into the shipping inventory template.
8. Discard excess ground biomass from each sample.
9. Clean grinding tools thoroughly between samples.
    - For a grinding mill, clean with compressed air.
    - Clean mortar and pestle with a kimwipe and ethanol.

## C.5 Equipment maintenance

- Balances should be calibrated with a standard calibration weight set:
  - After initial installation.
  - Any time the balance is moved to a new surface.
  - Every 6 months.
  - If you suspect readings are inaccurate for any reason.

## SOP D Dilution Sampling for Fine Root Fragments

The dilution sampling for fine root fragments procedure begins part-way through SOP C.1, after all roots ≥ 1 cm length have been picked from the sample. SOP D can be considered an add-on to SOP C that is performed on 20 randomly selected cores each time the fine root biomass protocol is implemented. Instead of ignoring and discarding organic material and root fragments < 1 cm length – hereafter referred to as the "residual fraction," the steps below describe how to separate roots from the residual fraction, and quantify them with a relatively time-efficient technique.

1. Randomly select 20 cores for processing according to this SOP.
    - First randomly select 20 clipIDs for dilution sampling, then randomly select either the 'N' or 'S' coreID from the selected clipIDs.
    - It is not possible to provide this list ahead of time because coring success in a given clip cell is not guaranteed.
    - If you are not familiar with how to generate a random list, create a JIRA ticket.
2. Take the residual fraction still in the 250 µm sieve from SOP C, and carefully wash with the root washer nozzle. The residual fraction should be free from mineral soil particles at this point.
3. Transfer the consolidated residual fraction – i.e. all roots < 1 cm length from a given clip strip – to a beaker so the sample may be randomly dispersed in water:
    a. Based on the size of the residual fraction, choose either a 1 L or 2 L beaker.

The goal is to sufficiently dilute the residual fraction so that not too many roots need to be picked and sorted, but not dilute so much that there are too few roots to weigh accurately once they are dry. If in doubt, use the 1 L beaker.

    b. With the root washer nozzle *on a low flow rate*, use ≤ 500 mL of water to transfer the residual fraction from the 250 µm sieve to the beaker.
        i. Note: using high pressure water may further disintegrate root fragments
    c. Carefully fill the beaker to ¾ full (i.e. 750 mL or 1.5 L). Be as accurate as possible, as this volume will be used to estimate the total mass of root fragments < 1 cm length.
    d. Record required metadata in the 'Lab Dilution' datasheet. Values will apply to all of the 10 pairs of aluminum weighing tins in the next step.
        - **coreDate**, date fine roots were sampled in the field
        - **plotID**, unique ID of the sampled plot
        - **subplotID**, unique ID of the sampled subplot
        - **clipCellNum**, the last three digits of the sampled clipID
        - **coreID**, either '*N*' or '*S*'
        - **residVolume**, volume of water used to suspend residual fraction in beaker; nearest 10 mL

4. Label 10 pairs (n=20 total) of aluminum weighing tins to hold 10 sub-samples of the aqueous residual fraction suspension.
    - For each pair of tins, one is for root fragments, and the other is for organic material.
    - Tins should be pre-numbered with a unique tinID (e.g. 1, 2, 3,…, 20, etc.). The tinID is tracked on the datasheet, rather than labeling each tin with the clipCellNumber.
    - Pre-weigh each tin with a microbalance, and record in the 'Lab Dilution' datasheet:
        - **subSampleID**, technician assigned number from 1 to 10
        - **tinID**, the unique number assigned to the tin
        - **tinEmptyMass**, the mass of the clean, dry, empty tin; nearest 0.1 mg
5. Work in pairs to generate 10 sub-samples from the aqueous suspended residual fraction in the beaker. Consult the training video for a visual demonstration of the following steps:
    a. [Person1] Turn the plate mixer on high, and vortex the aqueous suspended residual fraction thoroughly (approx. 10 s).

    b. [Person1] Turn off the mixer, and quickly plunge the suspension to stop the vortex and randomize the sample in the water.

    c. [Person2] Take a 20 mL sub-sample from the middle of the water volume in the beaker using the customized syringe, and transfer to one of the 'OM' tins.

    d. [Person2] Back off the plunger in the syringe to the 5 mL mark. Rinse the interior of the syringe with the squirt bottle, and transfer the rinse to the same tin.

    e. Record in the 'Lab Dilution' Datasheet:
        - **ssVolume**, the volume of the sub-sample taken from the beaker; nearest 1 mL (this volume will be the same number for both tinIDs in a pair)
            - The volume of water from the squirt bottle should not be added to this number.
        - **ssType**, the type of material the tinID will hold after picking and sorting is complete; the tin initially receiving the mixed sub-sample should be **ssType** = '*OM*', and the tin into which roots are sorted should be **ssType** = '*ROOT*.'

    f. Repeat step 5 (a) – (e) until 10 sub-samples have been transferred to 10 '*OM*' tins (**Figure 6**).
6. For each of the 10 sub-samples, carefully pick and sort root fragments from organic material, and transfer the roots to the '*ROOT*' tin of the pair (**Figure 6**).
    - A small amount of water in the *'ROOT'* tin aids in transferring root material.

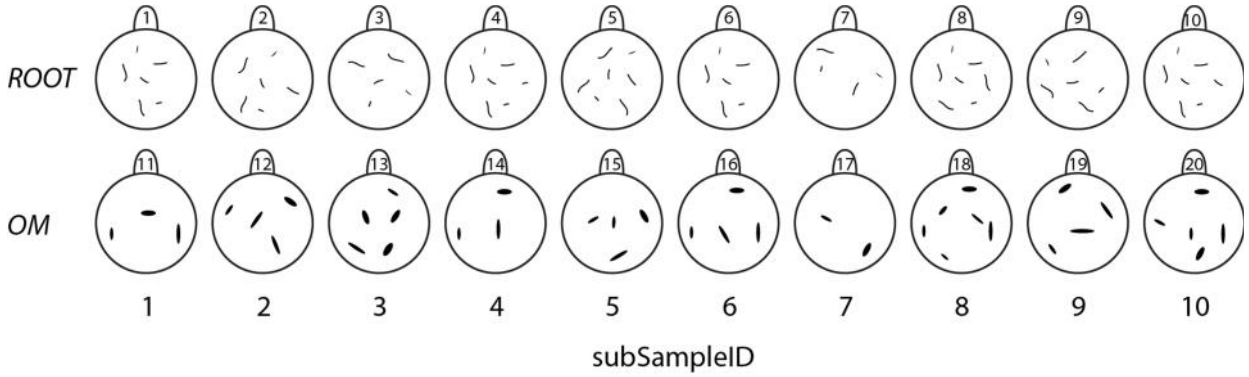


**Figure 6.** Pairs of labeled aluminum weighing tins for separating roots from OM in residual fraction sub-samples. Mixed sub-samples are initially transferred to the *OM* tins, and roots are then sorted into the *ROOT* tins.

7. Carefully transfer tins to a 65 ˚C drying oven for 48 h.
    - Use a tray to move batches of tins in the laboratory.
    - If the tray is metal, do not leave tins on the tray while drying; metal trays occasionally twist when heated, which will cause samples to spill.
8. Repeat steps (2) – (7) for additional cores.
9. Once tins are dry, weigh the total mass of each 'tin+ROOT' or 'tin+OM' with a microbalance. Record in the 'Lab Dilution' datasheet:
    - **tinSampleMass**, the mass of the dry 'tin+ROOT' or 'tin+OM' material; nearest 0.1 mg

## SOP E Data Entry and Verification

The importance of thorough, accurate data transcription cannot be overstated; the value of the efforts in the field is only manifested once the data are properly entered for delivery to NEON's end user community.

As a best practice, field data collected on paper datasheets should be digitally transcribed within 7 days of collection or the end of a sampling bout (where applicable). However, given logistical constraints, the maximum timeline for entering data is within 14 days of collection or the end of a sampling bout (where applicable).

Before entering data, all personnel ***MUST*** read RD[04] for complete instructions regarding manual data transcription. Prior to entering data via a web user interface (webUI), each technician shall enter a plot (or subplot) of data from one bout into the protocol-specific webUI housed on the Training portal, as described in RD[04].

Protocol-specific instructions and the associated data ingest workbook (RD[06]) for entering Belowground Biomass Core data can be found on the "TOS" intranet page hosted on the NEON FOPS intranet site. Be sure to enter data for all plots within a bout. If an entire bout was missed, no data need be entered.

Currently, there is a change-controlled spreadsheet version of the ingest document (RD[06]), as well as a working database version of the document. The data entry tables are the same in both the spreadsheet and database versions, but the change-controlled spreadsheet version contains summary information about each field. There are three data ingest forms that must be completed:

- **perbout**: Metadata describing individual field sampling events on a per coreID basis.
- **massData**: Oven-dried fine root biomass data for each sizeClass by status combination per coreID, as well as weighing QA data, and a record of whether subsamples were produced for archive and chemical analysis.
- **dilutionData**: Oven-dried fine root fragment and organic matter mass data per coreID.

### E.1 Field Datasheets

1. Transcribe data from the Core Sampling for Belowground Biomass Field Datasheets (RD[05]) to the 'perbout' ingest form.
    - Consult the Belowground Biomass Soil Core ingest document (RD[06]) to determine appropriate values and formats for each field in the ingest table.
2. If a representative clip cell did not support belowground biomass core sampling, noted as `coringPossible = N' in the **remarks** field of the Field Datasheet, enter in the 'perbout' ingest form:

- **coringPossible** = 'N'

3. Update permanent digital versions of the Clip Lists with **date** and **status** = '5' data recorded in the field.

### E.2 Lab Datasheets

- Transcribe data from the 'Lab Weighing' datasheet into the 'massData' ingest form.
    - Consult the Belowground Biomass Soil Core ingest document (RD[06]) to determine appropriate values and formats for each field in the ingest table.
    - If a core sample contained no fine root biomass within a given **sizeClass**, enter '0' in the **dryMass** field.
- Transcribe data from the 'Lab Dilution' datasheet into the 'dilutionData' ingest form.

## SOP F Sample Shipment

Information included in this SOP conveys science-based packaging, shipping, and handling requirements for these samples, not lab-specific or logistical demands. For lab-specific shipping information, reference the "Shipping Information for External Facilities" document on CLA's NEON intranet site.

### F.1 Timelines

Dried, ground samples may be stored indefinitely before shipping.

### F.2 Storage / Shipping Conditions

Dried, ground samples sealed in 20 mL plastic or glass vials may be shipped at ambient temperature without preservatives.

### F.3 Grouping / Splitting Samples

Samples originating from the same clip cell should be grouped together if possible.

### F.4 Procedure

1. Take scintillation vial box containing processed samples out of temporary storage for shipment.
2. Wrap the box in bubble wrap and tape securely, then place in a FedEx box for shipment.
3. Include a copy of the USDA letter pertaining to shipment of dried plant sample material in the box and affix any labels required by the permit, if necessary.
    - See the "USDA Plant Shipping Letter" on CLA's NEON intranet site.
4. Include cover letter explaining shipment, and spreadsheet detailing sample inventory. At the current time, only samples shipped to the University of Wyoming for analysis require a cover letter. To create a sample inventory that accompanies the shipment:
    a. Copy and paste the list of subsampleIDs from the .csv generated during webUI data entry into the shipping inventory template.
    b. The list will need to be sorted and filtered by 'C' and 'A' subsample types as necessary to accompany the corresponding chemical analysis and archive shipments.
5. Address shipping label appropriately and ship ground.

### F.5 Laboratory Contact Information and Shipping / Receipt Days

See the "Shipping Information for External Facilities" and "External Facilities Closure Dates" documents on CLA's NEON intranet site.

## 8 REFERENCES

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## APPENDIX A DATASHEETS

The following datasheets are associated with this protocol:

**Table 10.** Datasheets associated with this protocol

| NEON Doc. # | Title |
|---|---|
| NEON.DOC.002135 | Datasheets for TOS Protocol and Procedure: Core Sampling for Plant Belowground Biomass |
| | |

These datasheets can be found in Agile or the NEON Document Warehouse.

## APPENDIX B QUICK REFERENCES

N/A

## APPENDIX C REMINDERS

N/A

## APPENDIX D ESTIMATED DATES FOR ONSET AND CESSATION OF SAMPLING

Belowground biomass soil core sampling is ideally timed to broadly coincide with the peak aboveground biomass clip harvest, due to the scientific utility of relatively coincident estimates of both aboveground and belowground biomass. As such, dates listed in **Table 11** below are the estimated dates after which greenness begins to decrease at each site, and in theory, after which the majority of above and belowground biomass has been produced. Dates are averages of 2001-2009 MODIS-EVI satellite phenology data. However, soil moisture also influences the timing of sampling, and as such, dates below may need to be adjusted at a given site based on soil moisture conditions within a given year. Soil core sampling should be concluded within 1 month of the actual start date.

Dates are provided in day-of-year (DOY) format. Conversions to MM-DD are provided in **Table 12**.

**Table 11.** Estimated average dates after which greenness begins to decrease for each NEON site based on MODIS-EVI phenology data. Ideally, soil core sampling and aboveground biomass clip harvests should occur on or near these dates.

| Domain | Site | Approx. Start Date (Day-Of-Year) |
|---|---|---|
| 01 | BART | 220 |
| | HARV | 220 |
| 02 | BLAN | 210 |
| | SCBI | 220 |
| | SERC | 220 |
| 03 | DSNY | 190 |
| | JERC | 220 |
| | OSBS | 190 |
| 04 | GUAN | |
| | LAJA | |
| 05 | STEI | 215 |
| | TREE | 215 |
| | UNDE | 215 |
| 06 | KONA | |
| | KONZ | 90 |
| | KUFS | |
| 07 | GRSM | 215 |
| | MLBS | 220 |
| | ORNL | 210 |
| 08 | CHOC | 200 |
| | DELA | 205 |
| | TALL | 195 |
| 09 | DCFS | 120 |
| | NOGP | 115 |
| | WOOD | 210 |
| 10 | CPER | 90 |
| | RMNP | 180 |

| Domain | Site | Approx. Start Date (Day-Of-Year) |
|---|---|---|
| | STER | 2-4 wks before crop harvest |
| 11 | CLBJ | |
| | KLEM | 75 |
| 12 | YELL | 190 |
| 13 | MOAB | 85 |
| | NIWO | 220 |
| 14 | JORN | 245 |
| | SRER | 240 |
| 15 | ONAQ | 75 |
| 16 | ABBY | |
| | WREF | 210 |
| 17 | SJER | 270 |
| | SOAP | 185 |
| | TEAK | 205 |
| 18 | BARO | 210 |
| | TOOL | 205 |
| 19 | DEJU | 210 |
| | HEAL | 210 |
| | POKE | 205 |
| 20 | OLAA | |

**Table 12**. Day-of-year calendar for non-leap years.

| Day | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | Oct | Nov | Dec | Day |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 001 | 032 | 060 | 091 | 121 | 152 | 182 | 213 | 244 | 274 | 305 | 335 | 1 |
| 2 | 002 | 033 | 061 | 092 | 122 | 153 | 183 | 214 | 245 | 275 | 306 | 336 | 2 |
| 3 | 003 | 034 | 062 | 093 | 123 | 154 | 184 | 215 | 246 | 276 | 307 | 337 | 3 |
| 4 | 004 | 035 | 063 | 094 | 124 | 155 | 185 | 216 | 247 | 277 | 308 | 338 | 4 |
| 5 | 005 | 036 | 064 | 095 | 125 | 156 | 186 | 217 | 248 | 278 | 309 | 339 | 5 |
| 6 | 006 | 037 | 065 | 096 | 126 | 157 | 187 | 218 | 249 | 279 | 310 | 340 | 6 |
| 7 | 007 | 038 | 066 | 097 | 127 | 158 | 188 | 219 | 250 | 280 | 311 | 341 | 7 |
| 8 | 008 | 039 | 067 | 098 | 128 | 159 | 189 | 220 | 251 | 281 | 312 | 342 | 8 |
| 9 | 009 | 040 | 068 | 099 | 129 | 160 | 190 | 221 | 252 | 282 | 313 | 343 | 9 |
| 10 | 010 | 041 | 069 | 100 | 130 | 161 | 191 | 222 | 253 | 283 | 314 | 344 | 10 |
| 11 | 011 | 042 | 070 | 101 | 131 | 162 | 192 | 223 | 254 | 284 | 315 | 345 | 11 |
| 12 | 012 | 043 | 071 | 102 | 132 | 163 | 193 | 224 | 255 | 285 | 316 | 346 | 12 |
| 13 | 013 | 044 | 072 | 103 | 133 | 164 | 194 | 225 | 256 | 286 | 317 | 347 | 13 |
| 14 | 014 | 045 | 073 | 104 | 134 | 165 | 195 | 226 | 257 | 287 | 318 | 348 | 14 |
| 15 | 015 | 046 | 074 | 105 | 135 | 166 | 196 | 227 | 258 | 288 | 319 | 349 | 15 |
| 16 | 016 | 047 | 075 | 106 | 136 | 167 | 197 | 228 | 259 | 289 | 320 | 350 | 16 |
| 17 | 017 | 048 | 076 | 107 | 137 | 168 | 198 | 229 | 260 | 290 | 321 | 351 | 17 |
| 18 | 018 | 049 | 077 | 108 | 138 | 169 | 199 | 230 | 261 | 291 | 322 | 352 | 18 |
| 19 | 019 | 050 | 078 | 109 | 139 | 170 | 200 | 231 | 262 | 292 | 323 | 353 | 19 |
| 20 | 020 | 051 | 079 | 110 | 140 | 171 | 201 | 232 | 263 | 293 | 324 | 354 | 20 |
| 21 | 021 | 052 | 080 | 111 | 141 | 172 | 202 | 233 | 264 | 294 | 325 | 355 | 21 |
| 22 | 022 | 053 | 081 | 112 | 142 | 173 | 203 | 234 | 265 | 295 | 326 | 356 | 22 |
| 23 | 023 | 054 | 082 | 113 | 143 | 174 | 204 | 235 | 266 | 296 | 327 | 357 | 23 |
| 24 | 024 | 055 | 083 | 114 | 144 | 175 | 205 | 236 | 267 | 297 | 328 | 358 | 24 |
| 25 | 025 | 056 | 084 | 115 | 145 | 176 | 206 | 237 | 268 | 298 | 329 | 359 | 25 |
| 26 | 026 | 057 | 085 | 116 | 146 | 177 | 207 | 238 | 269 | 299 | 330 | 360 | 26 |
| 27 | 027 | 058 | 086 | 117 | 147 | 178 | 208 | 239 | 270 | 300 | 331 | 361 | 27 |
| 28 | 028 | 059 | 087 | 118 | 148 | 179 | 209 | 240 | 271 | 301 | 332 | 362 | 28 |
| 29 | 029 | | 088 | 119 | 149 | 180 | 210 | 241 | 272 | 302 | 333 | 363 | 29 |
| 30 | 030 | | 089 | 120 | 150 | 181 | 211 | 242 | 273 | 303 | 334 | 364 | 30 |
| 31 | 031 | | 090 | | 151 | | 212 | 243 | | 304 | | 365 | 31 |

## APPENDIX E SOIL CORE ASSEMBLY

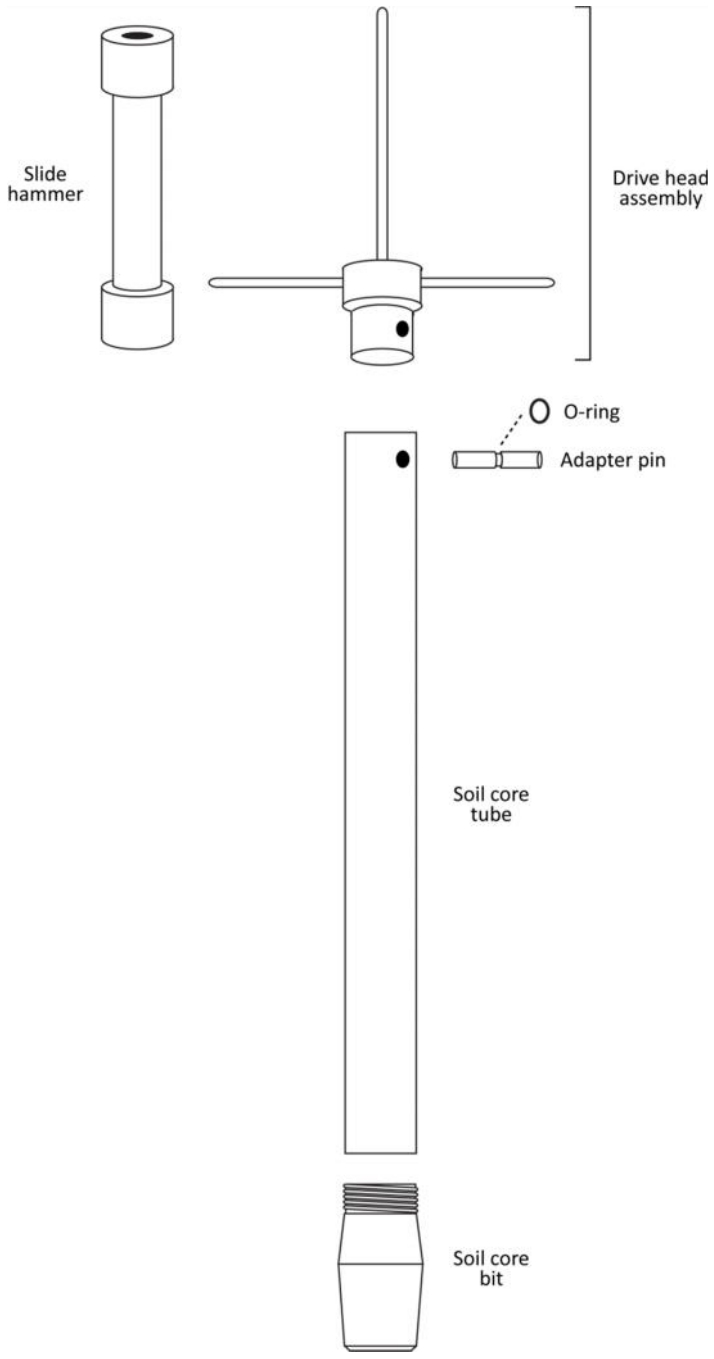


**Figure 7**. Component parts of the Giddings soil core assembly.

## APPENDIX F MANAGING EXPOSURE TO *TOXICODENDRON* SPECIES

1. **Prior to field work:**
   a. Count out coin envelopes or clasp envelopes for storing and drying root samples that will likely contain *Toxicodendron* biomass. Don't mix samples containing *Toxicodendron* biomass with any other samples.
   b. Pre-weigh (to nearest 0.01 g) and label each envelope that will be used for storing and drying cores containing *Toxicodendron* biomass. Once the weight of each empty envelope is written on the envelope, the biomass inside the bag will never have to be touched after it is initially placed in the bag.

2. **To collect soil cores containing *Toxicodendron* biomass in the field:**
   - Wear cotton gloves and dispose after single use.
   - Before collecting the core sample, use a pair of clippers dedicated solely to clipping *Toxicodendron spp* to clip and remove any aboveground *Toxicodendron* biomass that would be contacted while coring.
   - Bring a clean, new plastic bag to the field for storing and transporting contaminated gloves, soil coring equipment, and clippers after use.
   - Wear a thin outer layer of disposable PPE over clothes and shoes.
   - Upon returning to the laboratory, wear fresh latex gloves and clean clippers and soil coring equipment with Tecnu (or equivalent) after each use. Store exposed equipment separate from other laboratory equipment to prevent accidental contact.
   - After field work is complete, wash clothing according to these guidelines or similar:
     - http://laundry.about.com/od/removeoutdoorstains/a/poisonivylaundry.htm

3. **To process *Toxicodendron* biomass in the laboratory:**
   a. Use sieves and forceps dedicated to processing root samples containing *Toxicodendron* biomass. Wash sieves and forceps with Tecnu (or equivalent) following each use.
   b. Minimize potential spread of toxic oil by putting envelopes containing *Toxicodendron* roots into the same drying oven every time.
   c. When drying is complete, clean drying oven shelves used for drying *Toxicodendron* biomass with hot water and Tecnu. Wear appropriate PPE when cleaning.
   d. Record weight of bag + dried biomass to nearest 0.01 g, and also record weight of individual empty bag (to 0.01 g) on data sheets. Dried *Toxicodendron* biomass should never leave the bag.

4. After weighing, dispose of root samples containing *Toxicodendron* biomass. At this point in time, *Toxicodendron* tissue will not be ground for chemical analysis or archived.

## APPENDIX G CLIPCELLNUMBER COORDINATES AND MAPS

Belowground biomass soil core sampling and peak biomass clip harvest sampling ideally take place in the same clip cell in a given Tower plot. NEON Field Operations technicians must track the clip cell associated with coring and peak biomass clipping on the Clip Lists provided by Science Operations. When the Herbaceous Biomass clip harvest (RD[11]) precedes soil core sampling in the field, it is necessary to physically locate the clip cell in which the peak biomass clip occurred.

### G.1 Maps of clipCellNumber by subplotID

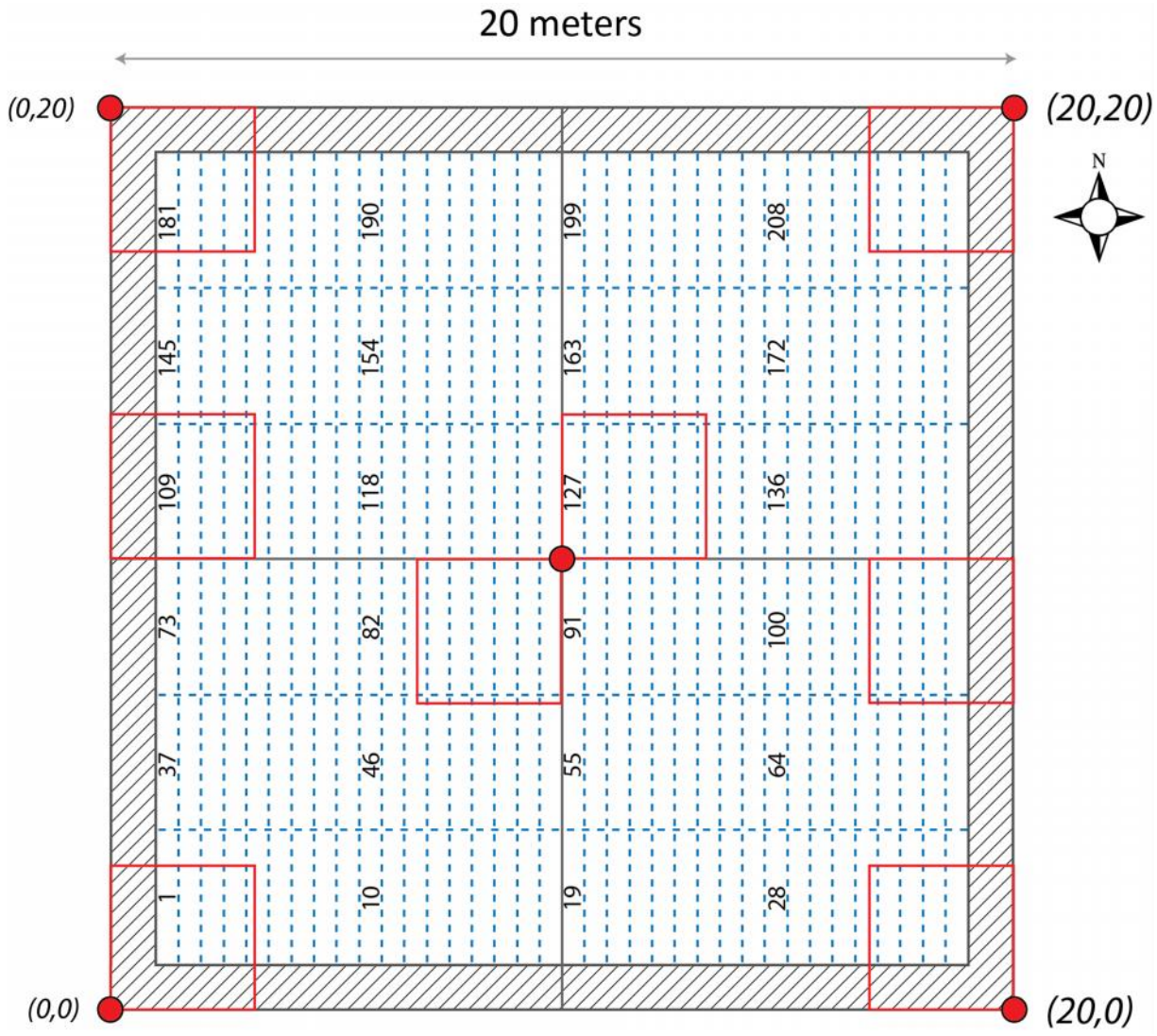


**Figure 8.** Map of clipCellNumbers in a 20m x 20m base plot (subplotID = 31 in provided Clip Lists). Red squares indicate nested subplots used for diversity sampling; clip cells that significantly overlap red squares are not used for fine root soil coring or clip sampling. However, cells with minimal overlap (e.g., 48-54, 68-72, 145-149) do support these sampling activities.

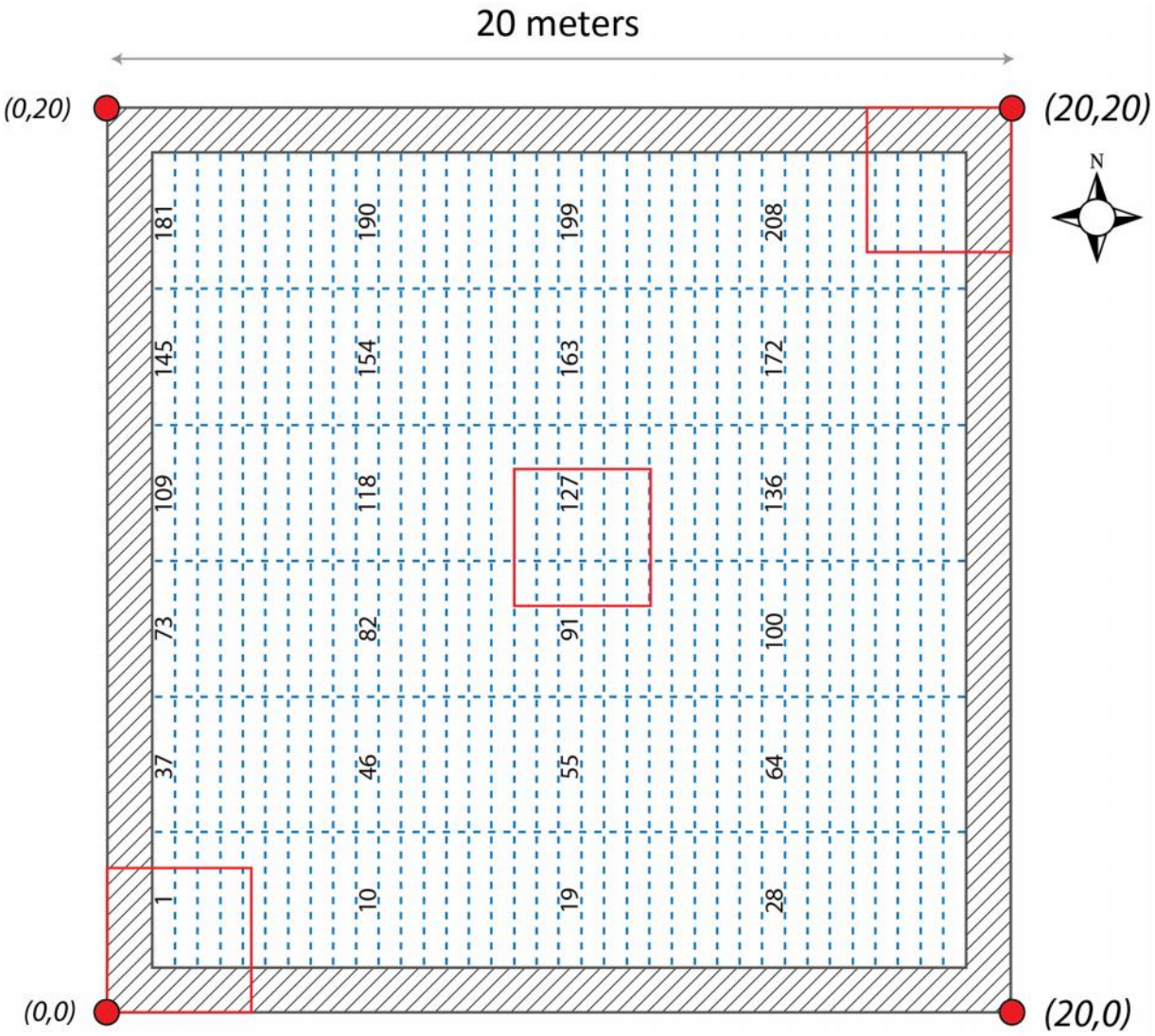


**Figure 9.** Map of clipCellNumbers for **subplotID = 21** in a 40m x 40m Tower base plot. Cells that overlap nested subplots indicated by red squares are not used for fine root soil core or clip sampling.

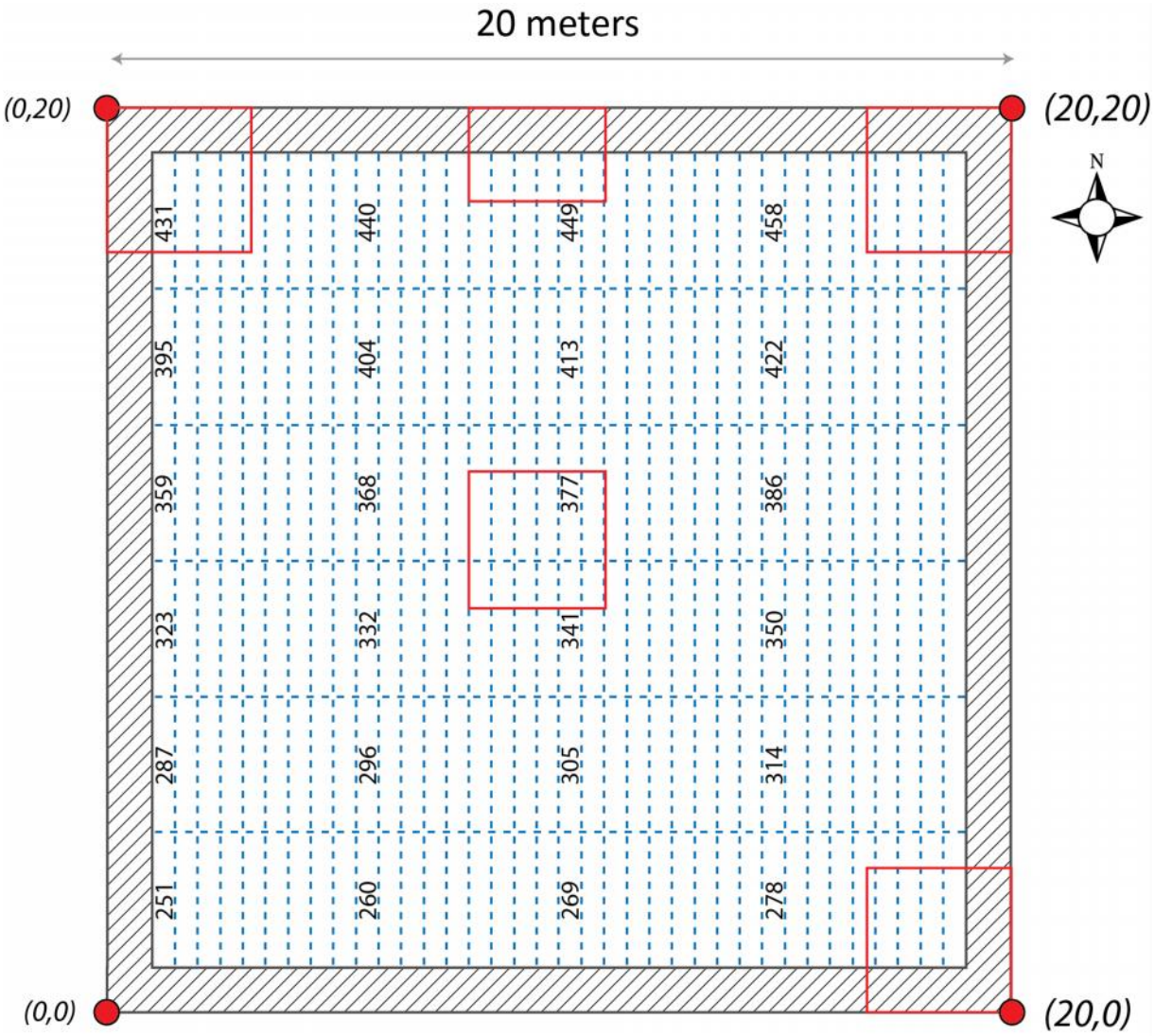


**Figure 10.** Map of clipCellNumbers for **subplotID = 23** in a 40m x 40m Tower base plot. Cells that overlap nested subplots indicated by red squares are not used for fine root soil core or clip sampling.

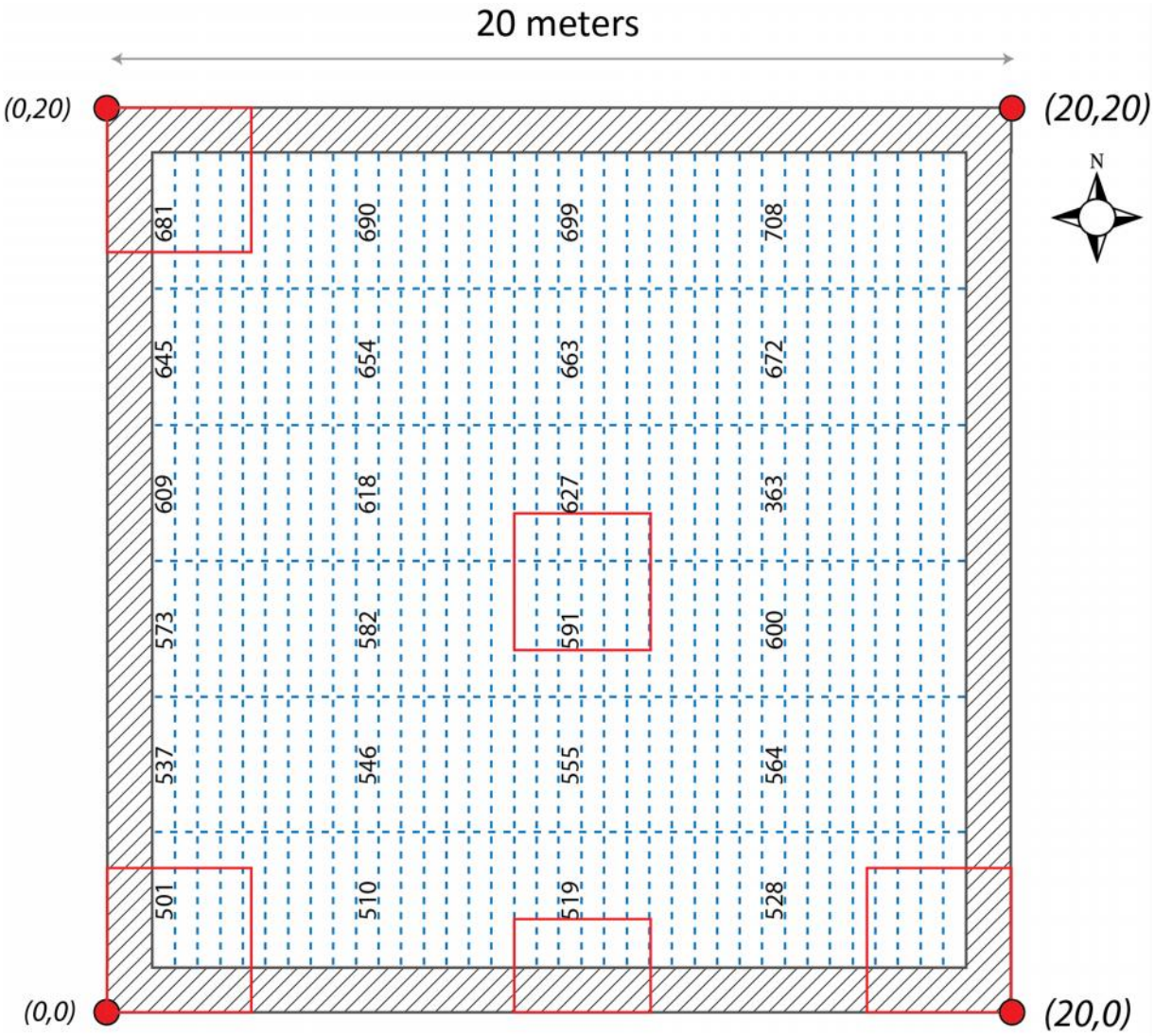


**Figure 11.** Map of clipCellNumbers for **subplotID = 39** in a 40m x 40m Tower base plot. Cells that overlap nested subplots indicated by red squares are not used for fine root soil core or clip sampling.

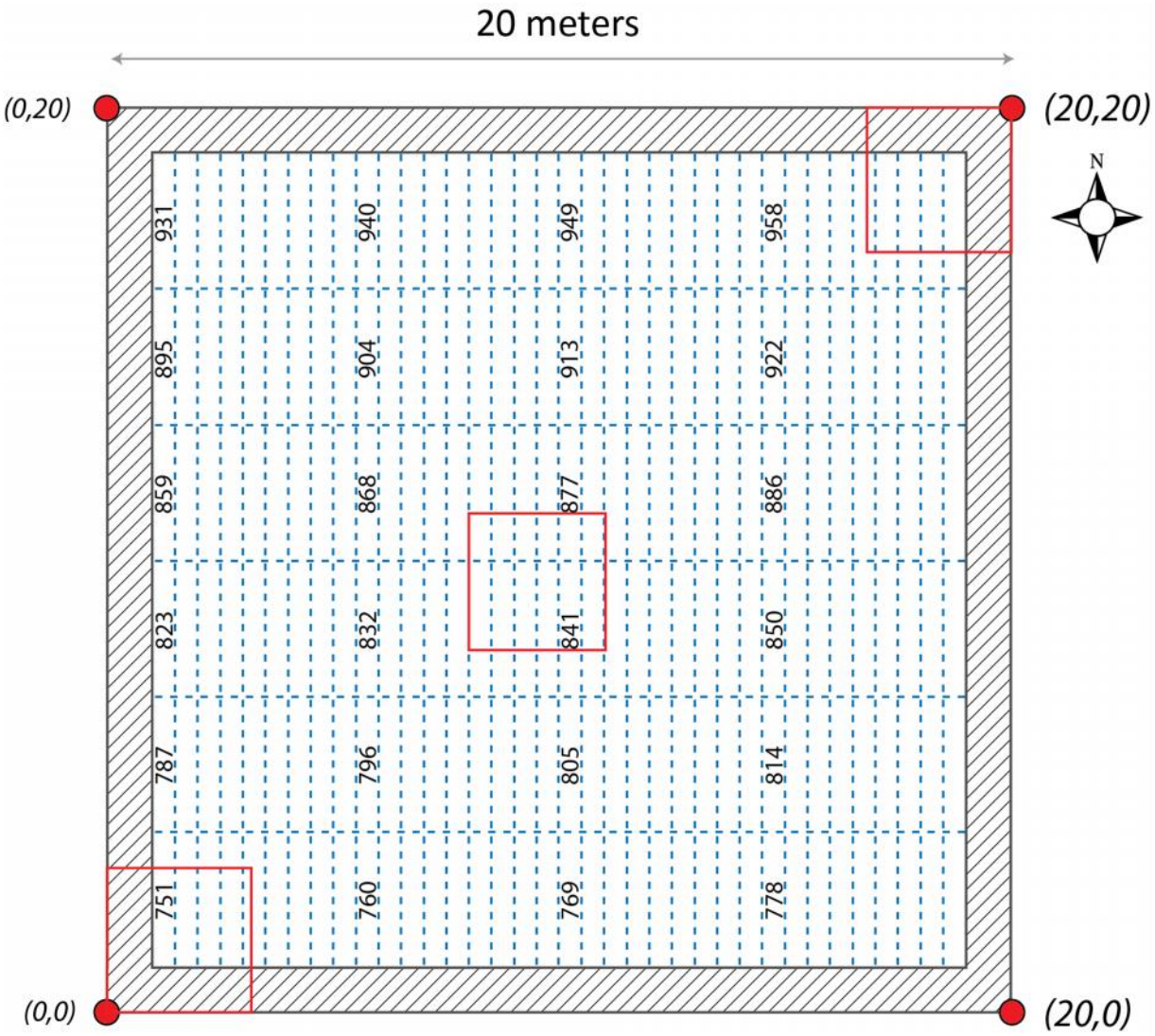


**Figure 12**. Map of clipCellNumbers for **subplotID = 41** in a 40m x 40m Tower base plot. Cells that overlap nested subplots indicated by red squares are not used for fine root soil core or clip sampling.

## G.2 Coordinates for clipCellNumbers by subplotID

**Table 13.** List of clipCellNumbers by subplotID and associated easting and northing coordinates. Coordinates correspond to the SW corner of a 0.1m x 2m Clip Strip, and indicate the distance in meters relative to the SW corner of the plot (subplotID = 31) or subplot (subplotID = 21, 23, 39, 41).

| clipCellNumber subplotID = 31 | clipCellNumber subplotID = 21 | clipCellNumber subplotID = 23 | clipCellNumber subplotID = 39 | clipCellNumber subplotID = 41 | easting offset | northing offset |
|---|---|---|---|---|---|---|
| 1 | 1 | 251 | 501 | 751 | 1.2 | 1.5 |
| 2 | 2 | 252 | 502 | 752 | 1.7 | 1.5 |
| 3 | 3 | 253 | 503 | 753 | 2.2 | 1.5 |
| 4 | 4 | 254 | 504 | 754 | 2.7 | 1.5 |
| 5 | 5 | 255 | 505 | 755 | 3.2 | 1.5 |
| 6 | 6 | 256 | 506 | 756 | 3.7 | 1.5 |
| 7 | 7 | 257 | 507 | 757 | 4.2 | 1.5 |
| 8 | 8 | 258 | 508 | 758 | 4.7 | 1.5 |
| 9 | 9 | 259 | 509 | 759 | 5.2 | 1.5 |
| 10 | 10 | 260 | 510 | 760 | 5.7 | 1.5 |
| 11 | 11 | 261 | 511 | 761 | 6.2 | 1.5 |
| 12 | 12 | 262 | 512 | 762 | 6.7 | 1.5 |
| 13 | 13 | 263 | 513 | 763 | 7.2 | 1.5 |
| 14 | 14 | 264 | 514 | 764 | 7.7 | 1.5 |
| 15 | 15 | 265 | 515 | 765 | 8.2 | 1.5 |
| 16 | 16 | 266 | 516 | 766 | 8.7 | 1.5 |
| 17 | 17 | 267 | 517 | 767 | 9.2 | 1.5 |
| 18 | 18 | 268 | 518 | 768 | 9.7 | 1.5 |
| 19 | 19 | 269 | 519 | 769 | 10.2 | 1.5 |
| 20 | 20 | 270 | 520 | 770 | 10.7 | 1.5 |
| 21 | 21 | 271 | 521 | 771 | 11.2 | 1.5 |
| 22 | 22 | 272 | 522 | 772 | 11.7 | 1.5 |
| 23 | 23 | 273 | 523 | 773 | 12.2 | 1.5 |
| 24 | 24 | 274 | 524 | 774 | 12.7 | 1.5 |
| 25 | 25 | 275 | 525 | 775 | 13.2 | 1.5 |
| 26 | 26 | 276 | 526 | 776 | 13.7 | 1.5 |
| 27 | 27 | 277 | 527 | 777 | 14.2 | 1.5 |
| 28 | 28 | 278 | 528 | 778 | 14.7 | 1.5 |
| 29 | 29 | 279 | 529 | 779 | 15.2 | 1.5 |
| 30 | 30 | 280 | 530 | 780 | 15.7 | 1.5 |
| 31 | 31 | 281 | 531 | 781 | 16.2 | 1.5 |
| 32 | 32 | 282 | 532 | 782 | 16.7 | 1.5 |
| 33 | 33 | 283 | 533 | 783 | 17.2 | 1.5 |
| 34 | 34 | 284 | 534 | 784 | 17.7 | 1.5 |
| 35 | 35 | 285 | 535 | 785 | 18.2 | 1.5 |
| 36 | 36 | 286 | 536 | 786 | 18.7 | 1.5 |
| 37 | 37 | 287 | 537 | 787 | 1.2 | 4.5 |

| clipCellNumber subplotID = 31 | clipCellNumber subplotID = 21 | clipCellNumber subplotID = 23 | clipCellNumber subplotID = 39 | clipCellNumber subplotID = 41 | easting offset | northing offset |
|---|---|---|---|---|---|---|
| 38 | 38 | 288 | 538 | 788 | 1.7 | 4.5 |
| 39 | 39 | 289 | 539 | 789 | 2.2 | 4.5 |
| 40 | 40 | 290 | 540 | 790 | 2.7 | 4.5 |
| 41 | 41 | 291 | 541 | 791 | 3.2 | 4.5 |
| 42 | 42 | 292 | 542 | 792 | 3.7 | 4.5 |
| 43 | 43 | 293 | 543 | 793 | 4.2 | 4.5 |
| 44 | 44 | 294 | 544 | 794 | 4.7 | 4.5 |
| 45 | 45 | 295 | 545 | 795 | 5.2 | 4.5 |
| 46 | 46 | 296 | 546 | 796 | 5.7 | 4.5 |
| 47 | 47 | 297 | 547 | 797 | 6.2 | 4.5 |
| 48 | 48 | 298 | 548 | 798 | 6.7 | 4.5 |
| 49 | 49 | 299 | 549 | 799 | 7.2 | 4.5 |
| 50 | 50 | 300 | 550 | 800 | 7.7 | 4.5 |
| 51 | 51 | 301 | 551 | 801 | 8.2 | 4.5 |
| 52 | 52 | 302 | 552 | 802 | 8.7 | 4.5 |
| 53 | 53 | 303 | 553 | 803 | 9.2 | 4.5 |
| 54 | 54 | 304 | 554 | 804 | 9.7 | 4.5 |
| 55 | 55 | 305 | 555 | 805 | 10.2 | 4.5 |
| 56 | 56 | 306 | 556 | 806 | 10.7 | 4.5 |
| 57 | 57 | 307 | 557 | 807 | 11.2 | 4.5 |
| 58 | 58 | 308 | 558 | 808 | 11.7 | 4.5 |
| 59 | 59 | 309 | 559 | 809 | 12.2 | 4.5 |
| 60 | 60 | 310 | 560 | 810 | 12.7 | 4.5 |
| 61 | 61 | 311 | 561 | 811 | 13.2 | 4.5 |
| 62 | 62 | 312 | 562 | 812 | 13.7 | 4.5 |
| 63 | 63 | 313 | 563 | 813 | 14.2 | 4.5 |
| 64 | 64 | 314 | 564 | 814 | 14.7 | 4.5 |
| 65 | 65 | 315 | 565 | 815 | 15.2 | 4.5 |
| 66 | 66 | 316 | 566 | 816 | 15.7 | 4.5 |
| 67 | 67 | 317 | 567 | 817 | 16.2 | 4.5 |
| 68 | 68 | 318 | 568 | 818 | 16.7 | 4.5 |
| 69 | 69 | 319 | 569 | 819 | 17.2 | 4.5 |
| 70 | 70 | 320 | 570 | 820 | 17.7 | 4.5 |
| 71 | 71 | 321 | 571 | 821 | 18.2 | 4.5 |
| 72 | 72 | 322 | 572 | 822 | 18.7 | 4.5 |
| 73 | 73 | 323 | 573 | 823 | 1.2 | 7.5 |
| 74 | 74 | 324 | 574 | 824 | 1.7 | 7.5 |
| 75 | 75 | 325 | 575 | 825 | 2.2 | 7.5 |
| 76 | 76 | 326 | 576 | 826 | 2.7 | 7.5 |
| 77 | 77 | 327 | 577 | 827 | 3.2 | 7.5 |
| 78 | 78 | 328 | 578 | 828 | 3.7 | 7.5 |
| 79 | 79 | 329 | 579 | 829 | 4.2 | 7.5 |

| clipCellNumber subplotID = 31 | clipCellNumber subplotID = 21 | clipCellNumber subplotID = 23 | clipCellNumber subplotID = 39 | clipCellNumber subplotID = 41 | easting offset | northing offset |
|---|---|---|---|---|---|---|
| 80 | 80 | 330 | 580 | 830 | 4.7 | 7.5 |
| 81 | 81 | 331 | 581 | 831 | 5.2 | 7.5 |
| 82 | 82 | 332 | 582 | 832 | 5.7 | 7.5 |
| 83 | 83 | 333 | 583 | 833 | 6.2 | 7.5 |
| 84 | 84 | 334 | 584 | 834 | 6.7 | 7.5 |
| 85 | 85 | 335 | 585 | 835 | 7.2 | 7.5 |
| 86 | 86 | 336 | 586 | 836 | 7.7 | 7.5 |
| 87 | 87 | 337 | 587 | 837 | 8.2 | 7.5 |
| 88 | 88 | 338 | 588 | 838 | 8.7 | 7.5 |
| 89 | 89 | 339 | 589 | 839 | 9.2 | 7.5 |
| 90 | 90 | 340 | 590 | 840 | 9.7 | 7.5 |
| 91 | 91 | 341 | 591 | 841 | 10.2 | 7.5 |
| 92 | 92 | 342 | 592 | 842 | 10.7 | 7.5 |
| 93 | 93 | 343 | 593 | 843 | 11.2 | 7.5 |
| 94 | 94 | 344 | 594 | 844 | 11.7 | 7.5 |
| 95 | 95 | 345 | 595 | 845 | 12.2 | 7.5 |
| 96 | 96 | 346 | 596 | 846 | 12.7 | 7.5 |
| 97 | 97 | 347 | 597 | 847 | 13.2 | 7.5 |
| 98 | 98 | 348 | 598 | 848 | 13.7 | 7.5 |
| 99 | 99 | 349 | 599 | 849 | 14.2 | 7.5 |
| 100 | 100 | 350 | 600 | 850 | 14.7 | 7.5 |
| 101 | 101 | 351 | 601 | 851 | 15.2 | 7.5 |
| 102 | 102 | 352 | 602 | 852 | 15.7 | 7.5 |
| 103 | 103 | 353 | 603 | 853 | 16.2 | 7.5 |
| 104 | 104 | 354 | 604 | 854 | 16.7 | 7.5 |
| 105 | 105 | 355 | 605 | 855 | 17.2 | 7.5 |
| 106 | 106 | 356 | 606 | 856 | 17.7 | 7.5 |
| 107 | 107 | 357 | 607 | 857 | 18.2 | 7.5 |
| 108 | 108 | 358 | 608 | 858 | 18.7 | 7.5 |
| 109 | 109 | 359 | 609 | 859 | 1.2 | 10.5 |
| 110 | 110 | 360 | 610 | 860 | 1.7 | 10.5 |
| 111 | 111 | 361 | 611 | 861 | 2.2 | 10.5 |
| 112 | 112 | 362 | 612 | 862 | 2.7 | 10.5 |
| 113 | 113 | 363 | 613 | 863 | 3.2 | 10.5 |
| 114 | 114 | 364 | 614 | 864 | 3.7 | 10.5 |
| 115 | 115 | 365 | 615 | 865 | 4.2 | 10.5 |
| 116 | 116 | 366 | 616 | 866 | 4.7 | 10.5 |
| 117 | 117 | 367 | 617 | 867 | 5.2 | 10.5 |
| 118 | 118 | 368 | 618 | 868 | 5.7 | 10.5 |
| 119 | 119 | 369 | 619 | 869 | 6.2 | 10.5 |
| 120 | 120 | 370 | 620 | 870 | 6.7 | 10.5 |
| 121 | 121 | 371 | 621 | 871 | 7.2 | 10.5 |

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|---|---|---|---|---|---|---|
| 122 | 122 | 372 | 622 | 872 | 7.7 | 10.5 |
| 123 | 123 | 373 | 623 | 873 | 8.2 | 10.5 |
| 124 | 124 | 374 | 624 | 874 | 8.7 | 10.5 |
| 125 | 125 | 375 | 625 | 875 | 9.2 | 10.5 |
| 126 | 126 | 376 | 626 | 876 | 9.7 | 10.5 |
| 127 | 127 | 377 | 627 | 877 | 10.2 | 10.5 |
| 128 | 128 | 378 | 628 | 878 | 10.7 | 10.5 |
| 129 | 129 | 379 | 629 | 879 | 11.2 | 10.5 |
| 130 | 130 | 380 | 630 | 880 | 11.7 | 10.5 |
| 131 | 131 | 381 | 631 | 881 | 12.2 | 10.5 |
| 132 | 132 | 382 | 632 | 882 | 12.7 | 10.5 |
| 133 | 133 | 383 | 633 | 883 | 13.2 | 10.5 |
| 134 | 134 | 384 | 634 | 884 | 13.7 | 10.5 |
| 135 | 135 | 385 | 635 | 885 | 14.2 | 10.5 |
| 136 | 136 | 386 | 636 | 886 | 14.7 | 10.5 |
| 137 | 137 | 387 | 637 | 887 | 15.2 | 10.5 |
| 138 | 138 | 388 | 638 | 888 | 15.7 | 10.5 |
| 139 | 139 | 389 | 639 | 889 | 16.2 | 10.5 |
| 140 | 140 | 390 | 640 | 890 | 16.7 | 10.5 |
| 141 | 141 | 391 | 641 | 891 | 17.2 | 10.5 |
| 142 | 142 | 392 | 642 | 892 | 17.7 | 10.5 |
| 143 | 143 | 393 | 643 | 893 | 18.2 | 10.5 |
| 144 | 144 | 394 | 644 | 894 | 18.7 | 10.5 |
| 145 | 145 | 395 | 645 | 895 | 1.2 | 13.5 |
| 146 | 146 | 396 | 646 | 896 | 1.7 | 13.5 |
| 147 | 147 | 397 | 647 | 897 | 2.2 | 13.5 |
| 148 | 148 | 398 | 648 | 898 | 2.7 | 13.5 |
| 149 | 149 | 399 | 649 | 899 | 3.2 | 13.5 |
| 150 | 150 | 400 | 650 | 900 | 3.7 | 13.5 |
| 151 | 151 | 401 | 651 | 901 | 4.2 | 13.5 |
| 152 | 152 | 402 | 652 | 902 | 4.7 | 13.5 |
| 153 | 153 | 403 | 653 | 903 | 5.2 | 13.5 |
| 154 | 154 | 404 | 654 | 904 | 5.7 | 13.5 |
| 155 | 155 | 405 | 655 | 905 | 6.2 | 13.5 |
| 156 | 156 | 406 | 656 | 906 | 6.7 | 13.5 |
| 157 | 157 | 407 | 657 | 907 | 7.2 | 13.5 |
| 158 | 158 | 408 | 658 | 908 | 7.7 | 13.5 |
| 159 | 159 | 409 | 659 | 909 | 8.2 | 13.5 |
| 160 | 160 | 410 | 660 | 910 | 8.7 | 13.5 |
| 161 | 161 | 411 | 661 | 911 | 9.2 | 13.5 |
| 162 | 162 | 412 | 662 | 912 | 9.7 | 13.5 |
| 163 | 163 | 413 | 663 | 913 | 10.2 | 13.5 |

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|---|---|---|---|---|---|---|
| 164 | 164 | 414 | 664 | 914 | 10.7 | 13.5 |
| 165 | 165 | 415 | 665 | 915 | 11.2 | 13.5 |
| 166 | 166 | 416 | 666 | 916 | 11.7 | 13.5 |
| 167 | 167 | 417 | 667 | 917 | 12.2 | 13.5 |
| 168 | 168 | 418 | 668 | 918 | 12.7 | 13.5 |
| 169 | 169 | 419 | 669 | 919 | 13.2 | 13.5 |
| 170 | 170 | 420 | 670 | 920 | 13.7 | 13.5 |
| 171 | 171 | 421 | 671 | 921 | 14.2 | 13.5 |
| 172 | 172 | 422 | 672 | 922 | 14.7 | 13.5 |
| 173 | 173 | 423 | 673 | 923 | 15.2 | 13.5 |
| 174 | 174 | 424 | 674 | 924 | 15.7 | 13.5 |
| 175 | 175 | 425 | 675 | 925 | 16.2 | 13.5 |
| 176 | 176 | 426 | 676 | 926 | 16.7 | 13.5 |
| 177 | 177 | 427 | 677 | 927 | 17.2 | 13.5 |
| 178 | 178 | 428 | 678 | 928 | 17.7 | 13.5 |
| 179 | 179 | 429 | 679 | 929 | 18.2 | 13.5 |
| 180 | 180 | 430 | 680 | 930 | 18.7 | 13.5 |
| 181 | 181 | 431 | 681 | 931 | 1.2 | 16.5 |
| 182 | 182 | 432 | 682 | 932 | 1.7 | 16.5 |
| 183 | 183 | 433 | 683 | 933 | 2.2 | 16.5 |
| 184 | 184 | 434 | 684 | 934 | 2.7 | 16.5 |
| 185 | 185 | 435 | 685 | 935 | 3.2 | 16.5 |
| 186 | 186 | 436 | 686 | 936 | 3.7 | 16.5 |
| 187 | 187 | 437 | 687 | 937 | 4.2 | 16.5 |
| 188 | 188 | 438 | 688 | 938 | 4.7 | 16.5 |
| 189 | 189 | 439 | 689 | 939 | 5.2 | 16.5 |
| 190 | 190 | 440 | 690 | 940 | 5.7 | 16.5 |
| 191 | 191 | 441 | 691 | 941 | 6.2 | 16.5 |
| 192 | 192 | 442 | 692 | 942 | 6.7 | 16.5 |
| 193 | 193 | 443 | 693 | 943 | 7.2 | 16.5 |
| 194 | 194 | 444 | 694 | 944 | 7.7 | 16.5 |
| 195 | 195 | 445 | 695 | 945 | 8.2 | 16.5 |
| 196 | 196 | 446 | 696 | 946 | 8.7 | 16.5 |
| 197 | 197 | 447 | 697 | 947 | 9.2 | 16.5 |
| 198 | 198 | 448 | 698 | 948 | 9.7 | 16.5 |
| 199 | 199 | 449 | 699 | 949 | 10.2 | 16.5 |
| 200 | 200 | 450 | 700 | 950 | 10.7 | 16.5 |
| 201 | 201 | 451 | 701 | 951 | 11.2 | 16.5 |
| 202 | 202 | 452 | 702 | 952 | 11.7 | 16.5 |
| 203 | 203 | 453 | 703 | 953 | 12.2 | 16.5 |
| 204 | 204 | 454 | 704 | 954 | 12.7 | 16.5 |
| 205 | 205 | 455 | 705 | 955 | 13.2 | 16.5 |

| clipCellNumber subplotID = 31 | clipCellNumber subplotID = 21 | clipCellNumber subplotID = 23 | clipCellNumber subplotID = 39 | clipCellNumber subplotID = 41 | easting offset | northing offset |
|---|---|---|---|---|---|---|
| 206 | 206 | 456 | 706 | 956 | 13.7 | 16.5 |
| 207 | 207 | 457 | 707 | 957 | 14.2 | 16.5 |
| 208 | 208 | 458 | 708 | 958 | 14.7 | 16.5 |
| 209 | 209 | 459 | 709 | 959 | 15.2 | 16.5 |
| 210 | 210 | 460 | 710 | 960 | 15.7 | 16.5 |
| 211 | 211 | 461 | 711 | 961 | 16.2 | 16.5 |
| 212 | 212 | 462 | 712 | 962 | 16.7 | 16.5 |
| 213 | 213 | 463 | 713 | 963 | 17.2 | 16.5 |
| 214 | 214 | 464 | 714 | 964 | 17.7 | 16.5 |
| 215 | 215 | 465 | 715 | 965 | 18.2 | 16.5 |
| 216 | 216 | 466 | 716 | 966 | 18.7 | 16.5 |