TOS PROTOCOL AND PROCEDURE: LTR - LITTERFALL AND FINE WOODY DEBRIS

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See configuration management system for approval history.

Change Record

REVISION	DATE	ECO #	DESCRIPTION OF CHANGE
A	09/09/2014	ECO-02136	Initial release
B	10/15/2014	ECO-02357	Migration to new protocol template
C	04/16/2015	ECO-02771	• Minor updates for clarification and to maintain consistency with other productivity protocols • Revised steps for delineating clip cells • Revised specifications for chemical analysis
D	01/02/2016	ECO-03416	• New fields added : setDate (definition changed), addDate (replaces setDate on pertrap datasheet) • Added Appendix G: clip cell coordinate maps • Added Appendix H: Safe handling of Toxicodendron • Added Appendix I: Troubleshooting • Added Appendix J: Alternative materials • Clarified relative position calculations in SOP B • Updated text in SOP G: shipping to match instructions in herbaceous clip harvest protocol. • Added dryMass QC instructions • Modified lab drying QC datasheet to accommodate multiple drying ovens • Added instruction for mass <0.01g • Added supplementalDryingTime
E	02/08/2017	ECO-04373	• Migrated to new protocol template. • Clarified use cases for mixed vs. other functional groups, added mixed option for samples require >1 hr. to sort • Removed supplementalDryingTime – no longer being used in litter data product. • Added mixing step for creating plot level chemical analysis samples. Added mass guidelines for chemistry subsampling. • General clarification throughout.
F	08/08/2018	ECO-05685	• Updated guidelines for implementing this protocol and references to new SOP • Added barcode workflow • Updated shipping instructions

			- Clarified conditions under which 8-week sampling interval is acceptable in year-round and hybrid sampling schedules - Added table of estimated sampling time for each SOP - Added specific references to mobile application - Added guidance for invertebrate bycatch - Added ground trap sampling month to Appendix D - Excluded sampling during period of high bear activity in YELL, Appendix D - Added annual re-assessment of non-qualifying plots - Changed guidance to keep weighed material until data record is ingested, then discard
G	04/24/2019	ECO-06059	- Addressing delayed collection of ground traps, Table 3 - Added warning sticker to equipment list for use with Toxicodendron spp - Added plot awareness to training requirements - Clarified requirement to label traps with the trapID - Added reference to use of ground cloths to catch litter throughfall - Added note about using multiple sizes of collection bags - Clarified yearBoutBegan - Added toxicodendronPossible field and workflow - Added guidance to sort plots according to Morton Order - Clarified that bags used to dry samples should be discarded - Changed sorting workflow, mix at 10%, prioritize leaves and needles - Clarified that samples must come to room temperature after removing from the drying oven before weighing - Added steps for grinding C/N samples to align with CFC protocol - Added details for collection in snow to Appendix H - Added processing guidelines for Toxicodendron to Appendix G - Added specific for trap construction at YELL to Appendix I
H	03/04/2020	ECO-06312	- Moved text to new template rev J - Added additional guidance to generat bagIDs - Added additional trap labeling guidance and figure - Added collection label template to ssl, clarified that labels must be populated while in the field

			- Removed guidance to cool samples prior to weighing - Clarified sorting steps - Added sampleCondition field - Created new spreadsheet to track drying progress - Added bgcArchiveMass field to SOP F - Specified that cryo labels be used for blank tags with bgc sampleID

	Title: TOS Protocol and Procedure: Litterfall and Fine Woody Debris	Date: 03/04/2020
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1 OVERVIEW

1.1 Background

Quantifying production of litterfall and fine woody debris is required to estimate annual Aboveground Net Primary Productivity (ANPP) at plot, site and continental scales, and provides essential data for understanding vegetative C fluxes over time. Litterfall and fine woody debris production is estimated within Tower plots on an annual basis, based on litter accumulation in elevated and ground traps. In ecosystems with relatively continuous canopy, sampling point selection within a plot or subplot is random; sampling points are selected from the same randomized list generated to guide clip strip locations for herbaceous clip harvest. In ecosystems where the overstory is non-continuous (i.e. patchy), litterfall and fine woody debris sampling are targeted to litter-producing areas within the plot rather than random. The selected sampling strategy is used at all plots within a site. This protocol is not implemented at sites or plots with overstory vegetation < 2 meters tall.

This protocol calls for sorting fresh litter into specified functional groups prior to drying if time permits. If it is logistically not feasible to sort fresh material before drying, litter may be sorted after drying as time allows, or be measured and reported as a mixed, unsorted, sample. However, sorting freshly collected litter is preferable as dry litter is easily fragmented and identifying small or desiccated litter fragments to functional group introduces uncertainty in sorting accuracy.

Elevated litter trap size is consistent with existing standards and traps are the same dimensions (0.5 m^2 area; 70.7 cm L x 70.7 cm W x 80 cm H) as traps used by Smithsonian Tropical Research Institute Center for Tropical Forest Studies (Muller-Landau and Wright, 2010). To minimize the number of clip strips dedicated to fine woody debris sampling, which are therefore unavailable for herbaceous biomass sampling, ground traps have the same dimensions as a single clip strip cell, 3 m x 0.5 m.

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

This protocol is modeled closely after the litter monitoring protocol written by Helene C. Muller-Landau and S. Joseph Wright (2010) for the CTFS Global Forest Carbon Research Initiative.

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.004104	NEON Science Data Quality Plan
AD[07]	NEON.DOC.000914	TOS Science Design for Plant Biomass, Productivity, and Leaf Area Index

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. 002132	Datasheets for TOS Protocol and Procedure: Litterfall and Fine Woody Debris
RD[06]	NEON.DOC.003282	NEON Protocol and Procedure: Site Management and Disturbance Data Collection
RD[07]	NEON.DOC.001025	TOS Protocol and Procedure: Plot Establishment
RD[08]	NEON.DOC.001711	TOS Protocol and Procedure: Coarse Downed Wood
RD[09]	NEON.DOC.001924	NEON Raw Data Ingest Workbook for TOS Litterfall and Fine Woody Debris
RD[10]	NEON.DOC.001813	TOS Elevated Litter Trap Assembly Instruction
RD[11]	NEON.DOC.001717	TOS Standard Operating Procedure: TruPulse Rangefinder Use and Calibration
RD[12]	NEON.DOC.001716	TOS Standard Operating Procedure: Toxicodendron Biomass and Handling
RD[13]	NEON.DOC.000987	TOS Protocol and Procedure: Measurement of Vegetation Structure
RD[14]	NEON.DOC.001024	TOS Protocol and Procedure: Canopy Foliage Chemistry and Leaf Mass per Area Measurements
RD[15]	NEON.DOC.014048	TOS Protocol and Procedure: Soil Biogeochemical and Microbial Sampling
RD[16]	NEON.DOC.005023	TOS Standard Operating Procedure: Survey Method for Assessing Vegetation Cover

RD[17]	NEON.DOC.014038	TOS Protocol and Procedure: Plant Belowground Biomass Sampling
RD[18]	NEON.DOC.014037	TOS Protocol and Procedure: Measurement of Herbaceous Biomass
RD[19]	NEON.DOC.005224	NEON Protocol and Procedure: Shipping Ecological Samples, Sensors, and Equipment

2.3   Acronyms

Acronym	Definition
ANPP	Aboveground Net Primary Productivity
CTFS	Center for Tropical Forest Studies
NLCD	National Land Cover Dataset
MODIS	Moderate Resolution Imaging Spectroradiometer (NASA Satellite)
SOP	Standard Operating Procedure

2.4   Definitions

Litterfall: Shed leaves and needles, reproductive parts (i.e. flowers, fruits, cones, seeds, etc.), and fine woody debris with butt-end diameter < 2 cm (modified from Clark et al. 2001, Bernier et al. 2008, **Table 1**). All material that falls in elevated litterfall traps is considered for collection, including material that may not have been produced in the canopy (e.g, nest material, seeds from herbaceous species); material growing up through the mesh or up over the side of the trap should be clipped away and ignored. Woody pieces with diameter ≥ 2 cm are considered coarse downed wood, and are sampled according to the NEON Field and Lab Protocol for Coarse Downed Wood (RD[08]).

Fulcrum: Mobile application software used for data collection.

Service Now: Incident reporting software used to communicate internally.

3   METHOD

Standard Operating Procedures (SOPs), in Section 7 of this document, 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 Service Now 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.

Quality assurance is performed on data collected via these procedures according to the NEON Science Data Quality Plan (AD[06]).

3.1 Sampling Methods

For both elevated and ground traps, only the portion of material that meets both the length and diameter criteria is sampled (Muller-Landau and Wright 2010). Litter sampled from elevated traps is sorted into functional groups following collection, using the groupings outlined in Table 1.

Table 1. Size limits for functional groups collected in Elevated and Ground litter traps

Functional Group	Elevated Traps	Ground Traps
Leaves	< 50 cm length	> 50 cm length
Needles	< 50 cm length	N/A
Twigs/branches	< 2 cm diameter AND < 50 cm length	< 2 cm diameter AND > 50 cm length
Woody material (e.g. seed cones, bark, other lignified structures)	< 50 cm length	> 50 cm length
Seeds (including fruits and other attached structures*)	All	N/A
Flowers (includes pollen cones and attached structures* e.g., pedicels, peduncles…)	All	N/A
Other (lichen, mosses, frass, unidentifiable material, etc.)	All	N/A
Mixed (unsorted litter material)	< 2 cm diameter AND < 50 cm length	N/A

* Sort unattached structures that may come from either flowers or seeds but cannot be confidently categorized based on time of year or maturity with the "Flowers" functional group.

To ensure the accuracy of annual litter production estimates, ground traps are cleared of all qualifying litter material following the annual sampling bout.

Leaf and needle litter from elevated traps from a single sampling bout is shipped to external laboratories to be analyzed for %C, %N, δ13C and, δ15N once every five years (refer to Section 4 for more details about timing).

If site management, natural disturbance events, or regeneration/succession occur such that the cover of vegetation is significantly changed (e.g. management of woody encroachment through removal of all woody vegetation in a grassland site), submit a problem ticket to Science HQ. Such activities will prompt a re-survey of vegetation in Tower plots and if total aerial cover of remaining woody vegetation > 2 m is < 10%, sampling will be discontinued at the site.

3.2 Laboratory processing

Following collection litter is transported back to the laboratory, sorted to functional group, then dried at 65˚C to constant mass for a minimum of 48 hrs. The woody portion of litter is cut to fit in the drying oven then dried at 105°C to release bound water (Williamson and Wiemann 2010). Additionally, lignified structures associated with functional groups other than 'Woody material' or 'Twigs/branches' (e.g. hickory husks, walnut shells) may also require higher temperatures and extended dry times to release bound water.

3.3 Equipment

Design of PVC elevated litter traps is adopted from the CTFS design, though other materials (galvanized conduit or wood) may be substituted for PVC to accommodate site specific conditions. Non-oxidizable metal rods (e.g. fiberglass, aluminum, galvanized steel, or equivalent) are used to anchor elevated litter traps in place. Where permitted by the land use agreement, the corners of ground traps are marked with non-oxidizable metal or plastic stakes to facilitate precise re-measurement of the selected sampling area.

3.4 Spatial Distribution of Sampling

NEON establishes one elevated litter trap and one paired ground trap in two randomly selected 400 m^2 subplots in 1600 m^2 Tower plots or one litter trap pair per 400 m^2 Tower plot (see RD[07] for description of different Tower plot sizes). The selected subplots are the same ones used for all other plant productivity sampling in Tower plots (RD[18], RD[08], RD[13]). This general design may be modified at sites where statistical analyses dictate that fewer number of elevated traps are required to calculate productivity across Tower plots (Appendix D). At these sites, elevated traps have a paired ground trap, and a subset of plots have ground traps only.

3.5 Elevated traps

An elevated mesh litterfall trap (0.5 m^2; 70.7 cm W x 70.7 cm L x 80 cm H) is placed at a random location within each accepted plot/subplot, with trap locations selected from randomized list of sampling cells (see RD[18] for more information about the randomized list). Once set, traps remain in the same location within the plot for sampling in subsequent years unless traps are removed for optimization or a selected location is no longer logistically feasible (e.g. a tree fall blocks the previously selected random sampling cell). Elevated traps reliably sample material with butt-end diameter < 2 cm *and* length < 50 cm, including shed leaves, needles, reproductive parts, and fine woody debris. Traps are sampled according to the guidelines outlined in Section 7 of this document.

Deciduous forests are sampled once in the spring, then once every two weeks during leaf senescence. Evergreen systems including coniferous, xeric, and tropical forests are sampled year-round; the ideal sampling interval is once a month, approximately every 4 weeks, but may be extended to 8 weeks if the total number of bouts is projected to exceed 12 in a sampling year, or a single bout is missed and cannot

be rescheduled. Sites with both deciduous and evergreen vegetation are sampled according to a hybrid approach: monthly sampling with increased frequency during senescence.

In mixed woodland and grassland ecosystems (e.g. Domain 15 Onaqui, Domain 17 San Joaquin), woody vegetation cover is frequently patchy. As such, randomly placed litter traps are unlikely to adequately capture litter dynamics from woody vegetation. In this case, NEON targets litter trap placement to randomly selected areas of the plot with woody cover. NEON data users can then scale litter production using data from either NEON's Airborne Observation Platform or from the Woody Vegetation Structure data product.

3.6 Ground traps

Ground traps for collecting material with butt-end diameter < 2 cm and length > 50 cm including, large leaves, fronds, and fine woody debris, are randomly located in plots at least 2 meters from elevated traps, consistent with Muller-Landau and Wright (2010). To avoid interfering with other sampling within the plot, the basic ground trap sampling unit is one randomly selected 3 m x 0.5 m sampling cell within the same plot or subplot as the elevated trap (Figure 8, AD[07]). Ground traps are cleared of all litter > 50 cm in length and ≤ 2 cm diameter one year prior to the onset of sampling so that any litter within the selected area can be assumed to be the result of annual production. Only portions of large fronds or long sections of fine woody debris that lie inside the ground traps are sampled. Ground trap sample locations do not move from year to year and are excluded from consideration as locations for any other plant productivity sampling activities that use sampling cells.

4 SAMPLING SCHEDULE

4.1 Sampling Frequency and Timing

Litterfall and fine woody debris samples are collected according to the schedule in **Table 2**. Collection frequency is dictated by trap and vegetation type.

Implementation of SOP F: Processing Litter for Chemistry and Archive is on an inter-annual basis at a given site, completed as part of a suite of synchronized TOS measurements aimed at characterizing plant and soil biogeochemical dynamics. Synchronized protocols and SOPs include:

1. TOS Protocol and Procedure: Canopy Foliage Sampling (RD[14])
2. TOS Protocol and Procedure: Soil Biogeochemical and Microbial Sampling, including N Transformations (RD[07])
3. TOS Protocol and Procedure: Plant Belowground Biomass Sampling (RD[17]).

Table 2. Sampling frequency for Litterfall and Fine Woody Debris procedures per SOP per plot type.

SOP	Plot Type	Plot Number	Bout Duration	Bouts Per Year	Bout Interval	Yearly Interval	Remarks
SOP C	Tower – Elevated Trap	All	1 wk	~12	variable	1 y	Refer to **Table 3** and Appendix C for additional details
	Tower – Ground Trap	All	1 wk	1	1 y	1 y	
	Distributed	NA	NA	NA	NA	NA	Distributed and Gradient plots are not sampled for this protocol.
SOP E	Tower – all trap types	All	1 wk	~12	Same as SOP C	1 y	One lab processing bout for mass per field collection bout
SOP F	Tower – Elevated Trap	All	1 wk	1x	5 y	5 y	Coordinated BGC bouts

Table 3. Elevated trap sample timing and frequency for by vegetation type

Climate / Ecosystem	When to sample elevated traps
Temperate Deciduous	• Once in the spring, ± 2 weeks of the calendar date spring sampling occurred in the preceding year • Every two weeks during leaf senescence period
Coniferous / Evergreen / Tropical	• Once a month*, all year
Arid shrub	• Once a month*, all year
Mixed Deciduous/Evergreen Or Deciduous - marcescent	• Once a month* • Every two weeks during leaf senescence period

* An approximate 4 week sampling interval should be scheduled a priori to ensure data quality but may be decreased to once every 8 weeks if dictated by unforeseen logistical constraints.

4.2   Criteria for Determining Onset and Cessation of Sampling

The primary objective is to generate data that enable calculation of annual or per growing season litterfall and fine woody debris production within the dominant vegetation type (i.e. within Tower plots). To that end, sampling frequency differs by vegetation type based on the seasonality of litter production. Coniferous or evergreen forests may produce and drop needles or leaves year-round and therefore are sampled year-round but at lower frequency than deciduous forests. On the other hand, forests with a

distinct dormant season target sampling activity to the growing season and sample with greater frequency to effectively process the greater volume of material produced, particularly during annual periods of senescence. Mixed forests implement a hybrid strategy (Appendix C).

Material left uncollected in the field for longer than the specified sampling interval may be subject to granivory by small mammals, herbivory by insects, redistribution by wind, or increased decomposition resulting in loss of mass. In deciduous forests, elevated traps must be checked, at minimum, every two weeks during leaf senescence, as traps may fill in relatively short periods. Collection of litter during leaf senescence may occur at intervals less than two weeks if litter volume is high and resources are sufficient to support additional sampling. Opportunistically increased sampling frequency is not prescribed by NEON Science.

Sampling Cessation – Elevated traps in sites dominated by deciduous vegetation

At deciduous sites, Tower Plot end of season sampling of elevated traps should continue at a two-week interval until plants return to a dormant state and leaves are no longer falling. Sampling may be discontinued when:

1. *Senescence is complete.*

    a. *Assuming senescence occurs between bouts, carry out next scheduled bout after senescence is reported, then cease sampling.*

2. *The stop date in Appendix D is reached.*

    a. *Stop dates are derived from satellite greenness data. Contact NEON Science if plants are routinely still dropping leaves at the provided date(s) relevant to your sites.*

Sampling Cessation – Elevated traps in sites dominated by evergreen, mixed, arid shrub or marcescent vegetation

At sites not dominated by deciduous vegetation in Tower Plots, sampling continues year-round according to site specific guidelines.

Timing for Laboratory Processing and Analysis

Store field collected samples in a refrigerator or cooler with ice at the end of the day, until sorting occurs. If sorting is expected to be >1 week from collection, move samples to the freezer.

4.3 Timing for Laboratory Processing and Analysis

Table 4. Holding times for different litter samples types.

Sample type	Activity	Holding Time
Oven-dried Chemistry samples	Subsample and ship to external labs	Within 60 days of collection
Oven-dried Biogeochemistry Archive samples	Ship to bioarchive facility	Within 90 days of collection

4.4 Sampling Timing Contingencies

Table 5. Contingency decisions for Litterfall and Fine Woody Debris.

Delay/Situation	Action	Outcome for Data Products
Hours	If delay prevents completion of litter collection from a single trap, resume collection as soon as possible.	No adverse outcome
	If delay occurs between plots, resume litter trap collection as soon as possible.	
1-7 days	If delay prevents completion of litter collection from a single trap: 1. Store already collected litter in a cooler/refrigerator (acceptable), or sort and oven-dry per protocol (best), 2. Resume collection of litter traps ASAP with new labeled bags	No adverse outcome
	If delay occurs between litter traps, resume collection of remaining litter traps as soon as possible.	
8-13 days or longer*	If all traps are not collected in a single bout, prioritize collection of litter from missed traps at the subsequent bout. If sorting is expected to be delayed by > 1 week, store samples in -20°C freezer. Dried samples may also be stored for up to 30 days in ambient room conditions prior to weighing, but must be re-dried for 24 hrs prior to weighing.	Some litter mass may be lost from traps or collected samples, increasing uncertainty in biomass and ANPP estimates.

* If delays occur on ground trap collection, attempt to collect as soon as possible, do not delay until next bout (i.e. 1 year later). It is important that material is collected in the same year it is produced.

4.5　　　Missed or Incomplete Sampling

Sampling according to the schedule is not always possible, and multiple factors may impede work in the field at one or more plots or sampling locations in a given bout. For example:

* Logistics – e.g., insufficient staff or equipment
* Environment – e.g., deep snow, flooding, inclement weather, etc.
* Management activities – e.g., controlled burns, pesticide application

Some types of missed sampling are due to events that should be recorded in the Site Management App; refer to the Site Management and Event Reporting Protocol for more detail (RD[06]).

Missed or Incomplete Sampling Terms

Terms that inform Missed or Incomplete Sampling include:

* **Protocol Sampling Dates**: Bout-specific sampling dates (Appendix C).
* **Scheduled Sampling Dates**: Bout-specific sampling dates scheduled by Field Science and approved by Science. These dates coincide with or are a subset of the Protocol Sampling Dates.
* **Missed Sampling**: Incidence of *scheduled sampling* that did not occur. Missed Sampling is recorded at the same resolution as data that are ordinarily recorded.
* **Rescheduled**: Missed Sampling is rescheduled for another time within the *protocol sampling dates*, resulting in no change to the total number of sampling events per year.

The documentation that must accompany missed sampling depends on the timing, subsequent action, and the audience appropriate for numerous scenarios (**Figure 1**).
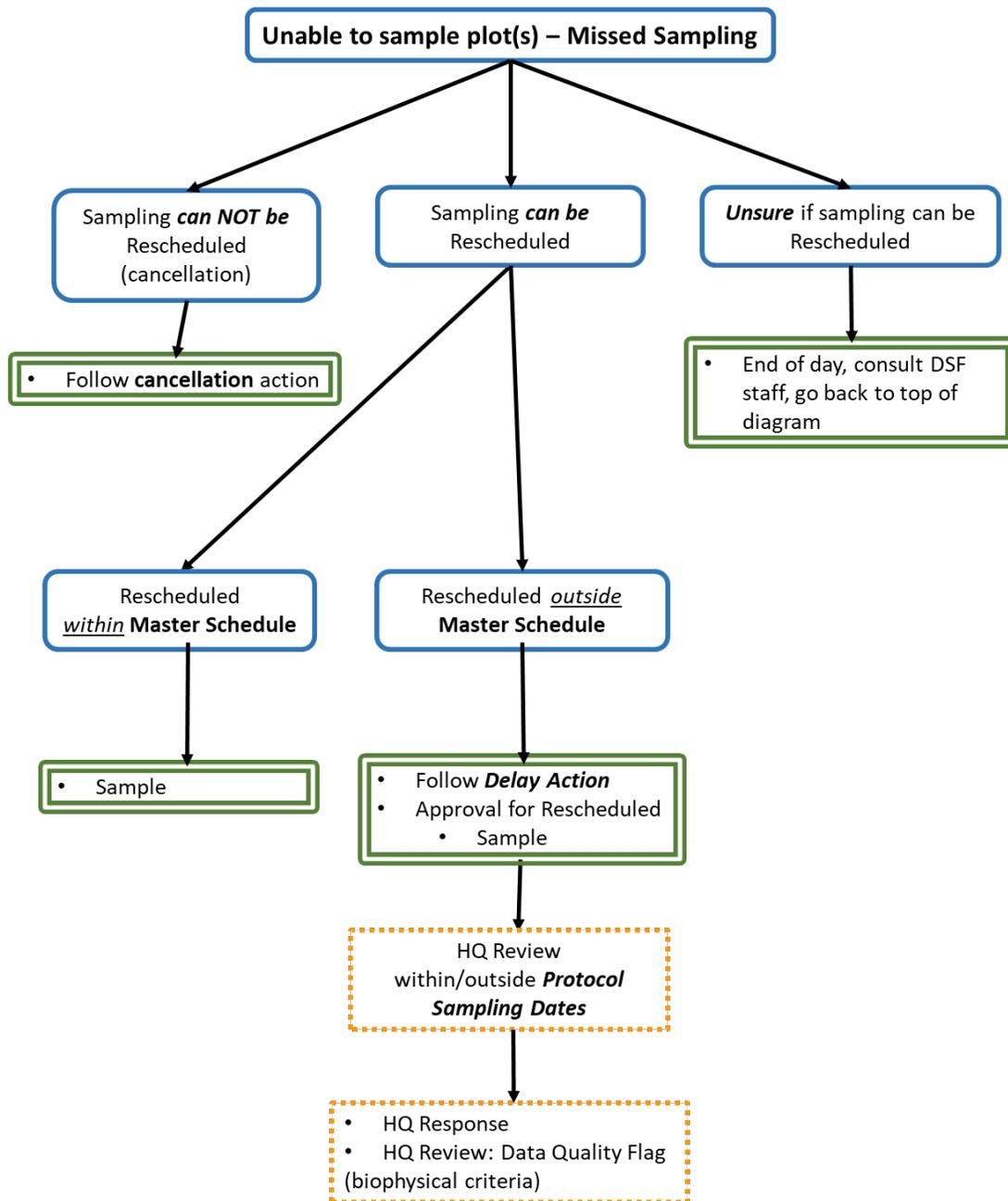
Figure 1. The documentation to account for a Missed Sampling event depends on the situation for each plot of each bout that is not sampled. Blue rounded boxes represent contingencies, green double line boxes describe the required actions, orange dotted boxes indicate HQ actions. Required delay and cancellation actions are outlined for each protocol in the 'Scheduled Field Activities – Delays and Cancellations' spreadsheet available on the SSL. Missed Sampling events may also require a Data Quality flag and/or creation of a Site Management record.

To Report Missed or Incomplete Sampling:

1. Missed or Incomplete Sampling must be communicated to Science by a Service Now Incident.

b. For Missed Sampling that is Rescheduled, there are some cases that require approval by Science and Operations (**Figure 1**).

c. The lead Field Ecologist should consult the Delayed or Cancelled Activities table to best determine when reporting is required.

d. This data product reports set date, collect date and sampling duration so missed bouts are not required to interpret the data.

e. Missed Sampling bouts do not affect boutNumber. If a bout is cancelled, the boutNumber assigned in the Master Schedule is simply skipped.

4.6 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. Please note that if sampling at particular locations requires significantly more time than expected, Science may propose to move these sampling locations.

Field collection time is expected to only take a couple of minutes for each trap. The majority of time in the field is spent travelling between plots; travel time varies by site.

Lab processing time depends heavily on the volume of material collected and number of functional groups present in a given collection. Sorting material prior to drying typically takes less than an hour per trap. Weighing dry material is also dependent on the sample volume but should not take more than a couple minutes per functional group per trap. Grinding, subsampling, filling and labeling vials may take 10-15 minutes per functional group per trap.

Table 6. Estimated staff and labor hours required for implementation of Litterfall and Fine Woody Debris.

SOP	Estimated total time	Suggested staff	Total person hours
SOP A Preparing for sampling	1 hr	2	2 hrs.
SOP B Initial Deployment of traps	1-2 hrs./plot	2	2-4 hrs./plot
SOP C Field Sampling	0.25 hrs./plot	2	0.5 hrs./plot
SOP D Laboratory Processing for Dry Mass Measurement	1.5/hrs./plot	1	1.5 hrs/plot
SOP F Processing Litter Samples for Bioarchive and Leaf Chemistry	0.5 hrs./plot	1	0.5 hrs./plot

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.

A laser rangefinder/hypsometer/compass instrument is used to locate randomly assigned trap locations. Safety considerations for this instrument include:

- Avoid staring directly at the laser beam for prolonged periods. The rangefinder is classified as eye-safe to Class 1 limits, which means that virtually no hazard is associated with directly viewing the laser output under normal conditions. As with any laser device, however, reasonable precautions should be taken in its operation. It is recommended that you avoid staring into the transmit aperture while firing the laser.
- Never attempt to view the sun through the scope. Looking at the sun through the scope may permanently damage the eyes.

Pipe glue used to attach PVC legs to the elevated trap is highly flammable and may cause skin and eye irritation. Vapors are also potentially dangerous if inhaled. Employees using glue should familiarize themselves with the hazards associated with this product (refer to the SDS), and with proper handling techniques.

Personnel assigned the task of constructing elevated traps shall complete Hand and Power Tool Safety Training and Machine Shop Safety (available on the Safety page of the NEON intranet) if cutting of PVC for construction is necessary, or if wood traps are used instead of PVC. Personnel shall be trained in the safe use, maintenance and cleaning of the Wiley® Mill or equivalent. *Toxicodendron* spp. (i.e. poison ivy, poison oak and poison sumac) are common and may cause skin rashes on susceptible individuals. The best defense is the use of clothing that covers the body with long pants and long-sleeved shirts and application of over-the-counter products for exposure to urushiol oils. Refer to NEON Operations Field Safety and Security Manual (AD[02] Section 7.1) and to Appendix G of this document and TOS Standard Operating Procedure: Toxicodendron Biomass and Handling (RD[12]) for safe handling instructions.

Heavy work gloves are recommended when collecting litter from ground traps or any time when sorting through litter where unseen hazards (e.g. spines, *Toxicodendron*, snakes, spiders) may be present.

When using the Wiley Mill to grinds samples for biogeochemistry analyses and archive, wear a dust mask if not grinding plant material in the hood.

6 PERSONNEL

6.1 Training Requirements

All technicians must complete required safety training as defined in the NEON Training Plan (AD[04]). Additionally, technicians must complete protocol-specific training for safety and implementation of this protocol as required in Field Operations Job Instruction Training Plan (AD[05]).

Field ecologists must be proficient in the use of handheld GPS units in order to successfully navigate to plots for sampling and have completed 'TOS Plots and Sampling' training (available in the FS - training center).

6.2 Specialized Skills

The lead Field Ecologist responsible for this protocol must possess the demonstrated ability to identify collected plant structures to functional group via visual inspection. Preferably, the field ecologists sorting litter are the same staff who harvested the litter in the field.

7   STANDARD OPERATING PROCEDURES

SOP Overview

This protocol is divided into six Standard Operating Procedures (SOPs). Each SOP addresses one discrete task and may be utilized as a standalone document as needed for specific field or lab tasks.

- **SOP A: Preparing for Sampling:** Includes gathering the necessary equipment and preloading the GPS with the necessary waypoints.
- **SOP B:Initial Deployment of Traps:** Describes the steps for locating sampling points and establishing litter trap pairs.
- **SOP C: Field Sampling:** Describes field collection of litterfall and fine woody debris from traps.
- **SOP D**: **Post-Field Sampling Tasks**
- **SOP E: Laboratory Processing for Dry Mass Measurement:** Covers laboratory processing including sorting, drying and weighing of samples.
- **SOP F: Processing Litter Samples for Chemistry:** Describes the steps for sub-sampling and grinding dried leaf and needle material.
- **SOP G: Data Entry and Verification:** Provides guidance for manual data transcription from paper data sheets if a mobile data recorder (MDR) is not available.
- **SOP H: Sample Shipment**: Provides science rationale for timelines and restrictions on sample handling and shipping to external facilities.
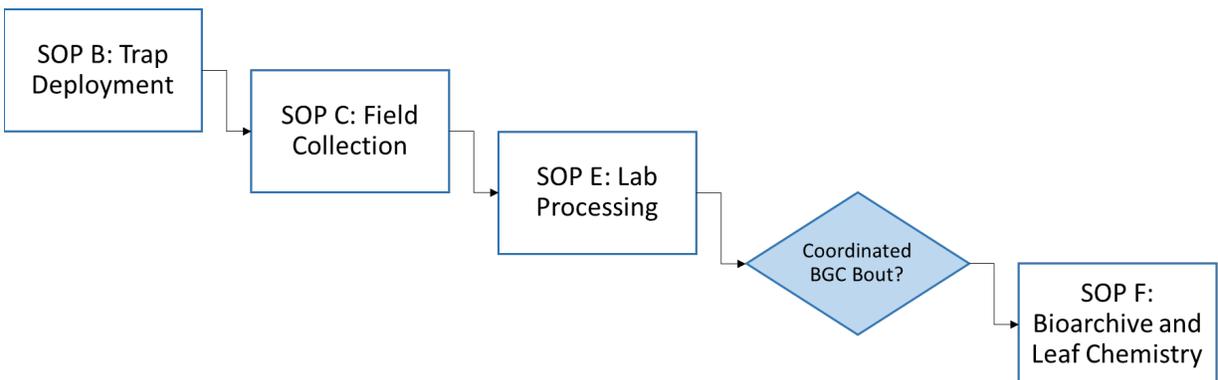
Figure 2. A high level workflow diagram that visually shows how the separate SOPs are sequentially connected.

SOP A   Preparing for Sampling

A.1   Preparing for Data Capture

Mobile applications are the preferred mechanism for data entry. Mobile devices should be fully charged at the beginning of each field day, whenever possible.

However, given the potential for mobile devices to fail under field conditions, it is imperative that paper datasheets are always available to record data. Paper datasheets should be carried along with the mobile devices to sampling locations at all times.

1. Generate randomized number lists for sites with targeted selection.
2. Print clip strip lists for the plots that will be visited. Clip lists are available on the SSL:

   - Litterfall sampling locations are selected from the plot-specific randomized lists created for herbaceous clip harvest locations (RD[18]). These lists are therefore essential for the completion of the trap deployment procedure, and must be updated to reflect the fact that two of the clipID locations are occupied by litter traps (elevated and ground). For the purpose of this protocol, trap location and clipID are used interchangeably.
   - Make sure that all fields in the clip strip lists are up to date, that clip strips that have been harvested or rejected are current and indicated on the lists.
   - These lists are utilized in the field regardless of selected trap placement strategy (i.e., random vs. targeted).

3. Gather all field equipment

   - If *Toxicodendron* is likely to be encountered, include cotton gloves and pre-weighed paper bags.
   - Include replacement mesh, pvc, zip ties, and other construction materials to repair broken traps as needed.

4. Download the 'LTR collection label template' from the SSL. Print on rite-in-the-rain paper. Cut for use during field collection.

5. Use a permanent marker to label cloth collection bags with a character string of any length, so each bag may be uniquely identified. This is the **bagID**. This string may contain numbers, letters, symbols but be aware that longer, more complicated strings may slow down data entry while in the field.

6. Prepare GPS:

   - Charge batteries
   - Load plot locations
   - Defining a route to each plot prior to going to the field will enable completion of the field collection bout in the least amount of time.

7.   Prepare laser rangefinder (if using). See RD[11] for details.

- Check battery and charge
- Clean lenses with lens cloth or lens tissue (if necessary)
- Check/set correct declination.
- Calibrate tilt sensor.
- Calibrate internal compass.

8. Prepare compass (if using).

- Check/set correct declination. Note that declination changes with time and should be looked up annually per site: http://www.ngdc.noaa.gov/geomag-web/

9. Print backup datasheets (RD[05]) on all-weather paper.

A.2   Labels and Identifiers

Barcodes are used for all samples processed for Chemistry and Archive. All barcodes need to be applied to dry containers at least 30 mins prior to use. Type I (prefix A, plus 11 numbers) are for all field samples and any non-cryo applications; they have a tolerance from 4C to 105C and still scan.

1. Prepare final sample containers by affixing one Type I adhesive barcode label to each vial used to contain a sample.
    a. If vials are used, barcode labels should be oriented such that it is possible to scan them; the scanner will not work on a curved surface. This means aligning the barcode lengthwise along a vial, *not* horizontally wrapping around a vial (**Figure 3**).
    b. Barcode labels must be associated with a unique sample and each barcode must be mapped to one sample in the database. Barcodes are unique, but are not initially associated with a particular sample, so you are encouraged to adhere barcode labels to needed containers in advance.
    c. If processing samples for bioarchive and leaf chemistry, pre-label scintillation vials with barcode labels and blank cryo-labels for writing the bgc sampleID.
        a. Affix the barcode to each vial to be filled with a unique sample.
        b. Affix the blank tag such that it does not interfere with the barcode label

Figure 3. An example of a Type I barcode. These large-size, field-tolerant barcodes have a prefix of 'A' followed by 11 numbers.

Figure 4. Annotated sampleID example. All sampleIDs are auto-generated in mobile application based on user inputs to metadata fields.

2. *About barcode uses and placement*

   This protocol generates mixed samples from the field, sorts those samples into functional groups for dry mass measurements, then pools samples from similar functional groups across the plot for grinding and chemical analyses.

3. Although it is always acceptable to use barcodes, in some cases barcodes are absolutely required. **Table 7** provides a quick reference to the types of sample that require barcodes. The rule of thumb is that the primary field sample will ALWAYS need a barcode due to its importance in generating future samples. Likewise, the final disposition of all vialed samples must have a barcode affixed to assist in the shipping and receipt of samples destined for the Biorepository or an external laboratory.

Table 7. Sample types and barcodes used.

Sample Type	Description	Example Identifier	Fulcrum App	Container Type	Barcode Used	Barcode Required?	Bar code Qty
Field samples	Unsorted samples from elevated or ground traps	NEON.ltr.SJER045008.20190618 *(NEON.ltr.plotID.trapID .collectDate)*	LTR: Field Sampling	Cloth bag	Type I	Optional, affix to paper tag inside bag	1 per trap per bout
Mass samples	Litter field samples sorted to functional group	NEON.ltr.SJER045008.20190618.lvs *(NEON.ltr.plotID.trapID .collectDate.functional Group)*	LTR: Lab Mass Data	Paper bag	Type I	Optional	1 per sub sample
BGC samples	Leaf and needle samples pooled by plot	NEON.ltr.SJER045.20190618.lvs.lig *(NEON.ltr.plotID.collect Date.functionalGroup. bgcSampleType)*	LTR: BGC Sub-sampling	20 mL polypropyl ene scint vials	Type I	Required	1 per sample

SOP B   Initial Deployment of Traps
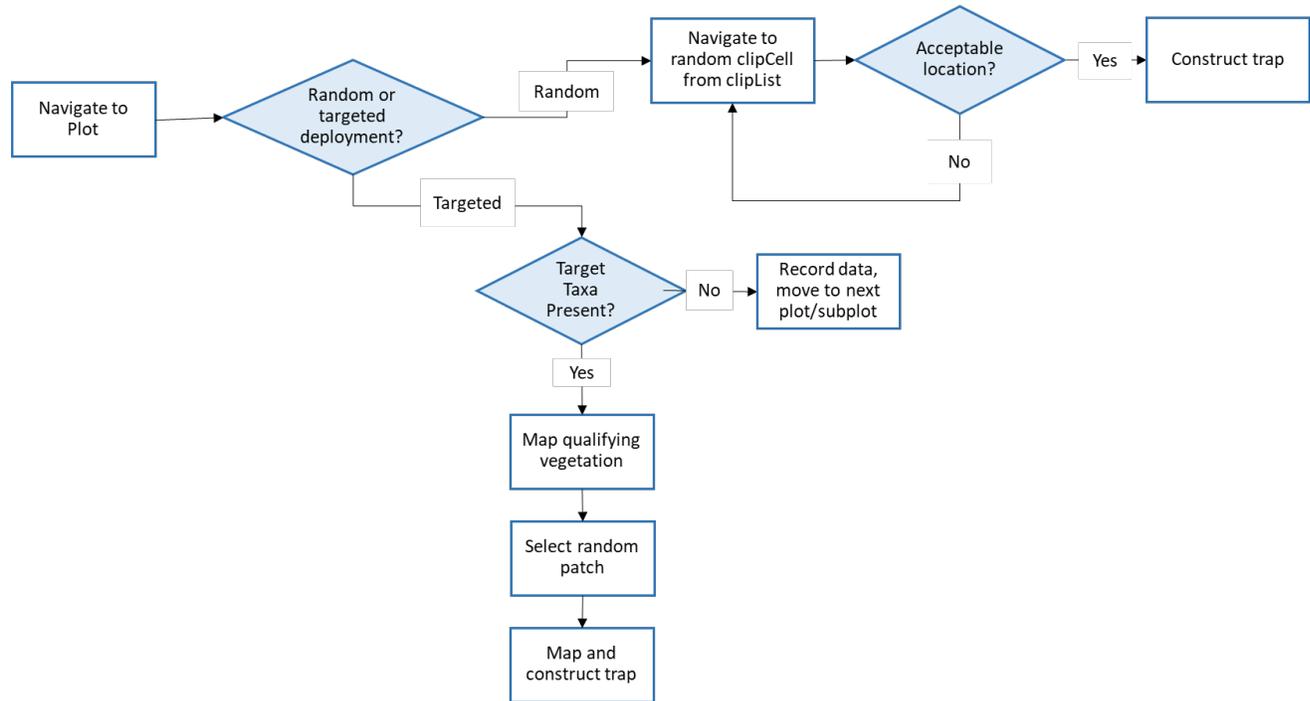
Figure 5. Workflow diagram for litterfall trap deployment.

Data entry guidelines and instructions for using the 'LTR: Trap Deployment [PROD]' fulcrum application are available in the Fulcrum Manual for this protocol, available on the SSL.

B.1   Selecting litter trap location strategy

In order to enable scaling of litter production across the site, the strategy for trap placement (i.e. Targeted or Random) is consistently applied across *all plots at a site* rather than based on plot specific conditions.

- **Targeted selection** is utilized for patchy vegetation, where overstory species ≥ 2 m height constitute < 50% canopy cover of the Tower airshed sampling area.
- **Random selection** is employed in forested sites with ≥ 50% canopy coverage of individuals ≥ 2 m tall

Refer to Appendix C for the recommended strategy by site; these recommendations are based on a combination of NLCD vegetation classification, satellite imagery and site characterization data. If the selected strategy/recommendation seems inappropriate for a particular site given the conditions on the ground (based on the criteria listed above), use Service Now to iterate with Science about the trap placement strategy.

Litter traps are typically deployed in pairs, one elevated and one ground trap per pair. One elevated trap and one ground trap is deployed in each of two randomly selected 400 m^2 subplots within a 1600 m^2 Tower plots. In smaller, 400 m^2 Tower plots, only one litter trap pair is deployed. Trap placement utilizes the clip cell grid developed for the herbaceous clip harvest protocol (RD[18]), and the random subplot selection list provided by NEON Science. Review and print Elevated Trap Assembly Instructions (RD[10]) for use in the field.

---

targetTaxaPresent = No

Any site dominated by herbaceous species, where woody vegetation is infrequent and too short to be reliably sampled by elevated litter traps (i.e., < 2 m), is excluded from consideration for implementation of this protocol. At sites where litter sampling does occur, all Tower Plots must be considered for deployment of litter traps.

Plots and sites with targetTaxaPresent = No will be re-surveyed every 5 years to determine whether qualifying vegetation is present and litter protocol should be implemented.

- If a random sampling strategy is employed and a given plot does not contain any vegetation > 2 m height
    - Record **targetTaxaPresent** = No
    - If **targetTaxaPresent** = Yes for the plot, but no qualifying vegetation is present in a given subplot, traps must still be deployed and sampled; **targetTaxaPresent** is a plot-level assessment.
- If a targeted sampling strategy is employed, a subplot or plot may be rejected if there is not sufficient woody vegetation >2 meters tall to allow for placement of both elevated and ground traps OR if all sampling locations beneath qualifying patches are within excluded sampling areas (i.e. 1 m buffer around plot edge and 1 m and 10 m diversity sampling areas).
    - Excluded clipCells are NOT available on the provided clipLists
    - Record **targetTaxaPresent = No** on datasheet or mobile app and continue to the next plot/subplot

---

B.2   Litter trap coordinates

Appendix G provides x, y-coordinates specific to litter trap placement but note that the clipLists posted on the NEON intranet only include coordinates for the SW corner of *clip strips* used to sample herbaceous biomass/productivity (RD[18]). Appendix G is necessary, along with the plot specific clipList to determine trap locations. **Figure 6** provides an overview of the relationship between clipList coordinates and litter trap locations.
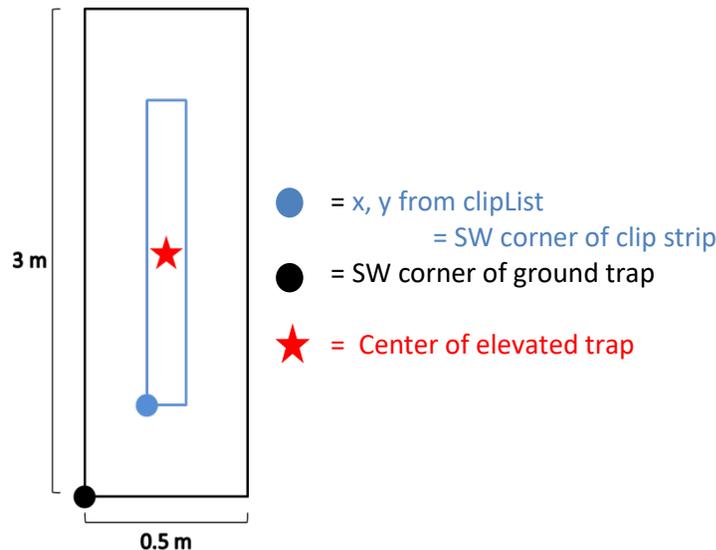
Figure 6. x y-coordinates for litter trap placement (red star, black circle, see Appendix G). These coordinates differ from those provided in the clipLists for use with the herbaceous clip protocol (blue circle) RD[18].

B.3   Targeted Sampling: Litter trap placement

Sites with patchy vegetation, where overstory species ≥ 2 m height constitute < 50% canopy cover of the Tower airshed sampling area, implement targeted trap placement in all Tower plots (see Appendix C).

1. Navigate to the desired plot and, if sampling in a 40 m x 40 m plot, the randomly selected subplot.

2. Assess location of patches of qualifying vegetation (≥ 2 m tall, outside of 1 m^2 and 10 m^2 diversity plots) within the plot or subplot (depending on plot size). If no qualifying patches are present, record **targetTaxaPresent** = No for the plot or subplot. Give each patch a numeric value. Assign values sequentially, left to right, bottom to top, beginning in the SW corner (Figure 7)

    a. If only a single qualifying patch is available, elevated and ground traps may be placed on opposite sides of the patch (even if < 2 m apart), provided the elevated trap is not situated such that it could potentially affect large particle distribution to the ground trap.

3. Use either a random number list, a series of coin flips, or other unbiased method of selection to select a patch to target for litterfall and fine woody debris sampling.

4. Once a patch is selected, select a location under the canopy, central to the patch to place an elevated litter trap.

    a. Avoid the 1-meter buffer around the plot edge, and the 1 m^2 and 10 m^2 nested subplots used for diversity sampling. Clip cell coordinates are not generated for those locations.

    b. If excluding 1 m^2 and 10 m^2 nested subplots removes all qualifying patches of vegetation from consideration, record **targetTaxaPresent** = No and move to the next plot/subplot

    c. If the selected patch is composed of dense, impenetrable vegetation such that traps cannot be placed centrally within the patch, traps may be placed along the patch edge according to one of the following strategies, listed in order of priority:

       1) Place trap along the patch edge where there is overhanging vegetation

       2) If there is a dominant wind direction, place trap along the leeward side of the patch so that the wind will carry litter from the vegetation toward the trap

       3) Use a random selection routine to select a cardinal direction

5. Place the elevated trap over the centroid of the clip cell that is nearest to the center of the target patch.

    a. Use the range finder to measure the distance to plot/subplot edges.

    b. From the selected location, measure distance to the nearest N-S plot boundary to determine the x-coordinate of this point

    c. Measure the distance to the nearest E-W plot boundary to determine the y-coordinate

    d. Use the clip cell map to identify the clip cell located closest to the selected point (Appendix G)

    e. Navigate to the centroid of that cell (Appendix G).

6. If practical, center trap over that point, this minimizes the number of clips that are removed from consideration for herbaceous clip harvest.

    a. In the example provided in Figure 7, the coordinates associated with the nearest clip strip centroid from the center of patch 4 are: x = 3.7, y = 11.5.

    b. Not centering the trap over a centroid is acceptable but not ideal as adjacent cells will also be excluded from consideration for herbaceous clip harvest.

7. Place a pin flag at the selected trap location.
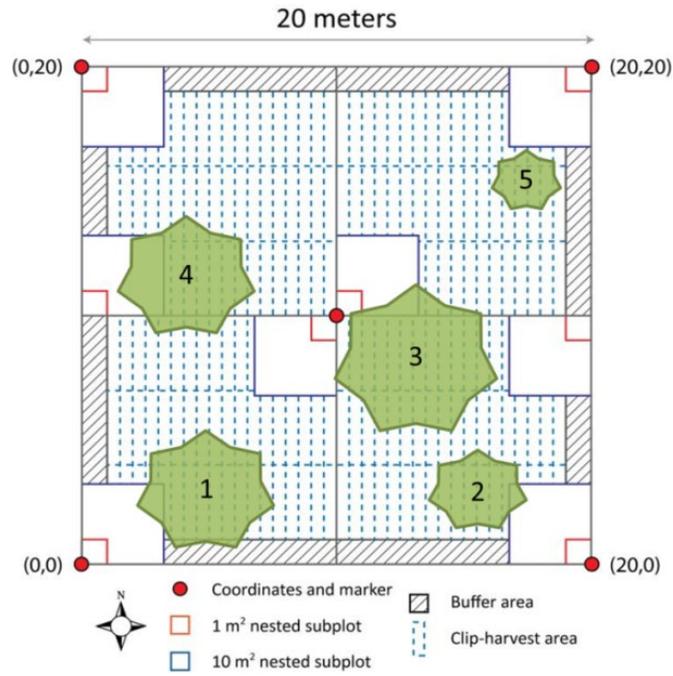
Figure 7. Example of numbering system for qualifying patches of vegetation within a plot.

B.4   Random Sampling: Litter trap placement

Sites where overstory species ≥ 2 m height constitute ≥ 50% canopy cover of the Tower airshed sampling area will implement random trap placement in all Tower plots (see **Table 14**).

Use the Site Specific Clip List (SITE_clipList.xlsx) to identify the first potential clip-strip location that has not already been sampled or rejected. Where relevant, subplot number is included in the file name and is also provided as a field in the spreadsheet.

1. Navigate to the SW corner of the clip strip of the first available sampling cell from the randomized list:
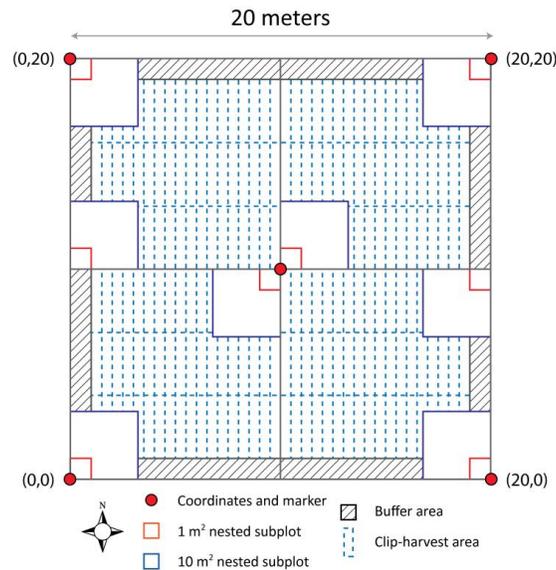
Figure 8. A 20 m x 20 m NEON plot showing the locations of 0.5m x 3m sampling "cells" (dashed blue lines). Larger plots have different nested subplots, but the coordinate numbering system for the 20 m subplot within these plots follow the same conventions as shown above. 40m x 40m plot schematic available in Appendix E.

If the Y-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 8), 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 at the X-coordinate, use the laser rangefinder in **HD** mode with a reflective surface to locate the Y-coordinate.

   • Make sure the azimuth is 0˚ (True North) when shooting the laser rangefinder to find the Y-coordinate (see RD[11]).

d. Place a pin flag at the clip-strip (X,Y) location.

If the Y-coordinate is > 10:

a. Run a tape East/West from the plot/subplot centroid (10,10) to either the (0,10) position or the (20,10) position (Figure 8) *Note: in 40 m x 40 m plots, subplot centroids may not permanently marked*:

X-Coordinate	Tape Layout[1]
1 < X < 10	From (10,10) to (0,10)[1]
10 < X < 20	From (10,10) to (20,10)[1]

[1] Use the laser rangefinder in **AZ** mode to guide the tape along the correct azimuth

b. Place a pin flag at the desired relative X-coordinate.

c. Standing directly over the pin flag that was just placed at the X-coordinate, use the laser rangefinder in HD mode with a reflective surface to locate the Y-coordinate.

d. Make sure the azimuth is 0˚ (True North) when shooting the laser rangefinder to find the Y-coordinate (see RD[11]).

e. Place a pin flag at the clip-strip (X,Y) location.

Note: If laser rangefinder is not available, the same routine described here may be completed using a handheld compass to verify azimuth and a tape measure for distance.

BEST PRACTICES

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

2. Assess the suitability of the sampling cell for an elevated litter trap:

- Accept the cell if no obstacles are present that prevent trap placement and anchoring (e.g. large shallow rock covering a majority of the cell, large boulders, impermeable vegetation, or low lying fallen trees that divert litter away from the trap location).
- Reject trap location if the selected cell is within 2 meters of an LAI sampling point and other sampling equipment located within the plot (e.g. grazing exclosure).
- If the sampling cell is not acceptable for placement of an elevated litter trap, move to the next one on the list but do NOT record the cell status as 'rejected' for herbaceous biomass sampling.

3. Navigate to the center of the cell and place a pin flag (**Figure 6**). Elevated traps are centered over this point.

   - If the trap cannot be anchored over the center of the cell, the trap may be shifted up to 1 meter North or South.
   - Record a '1' in the **Status** column of the Clip List sheet for clip-strip selected, Record the litter trap deployment date in the **Date** field, and add a note that the cell was used for litter collection.

B.5 Ground Trap placement

Ground traps are established to occupy one entire sampling cell and may not be placed such that more than one cell per 400 m² is occupied by a ground trap.

- **Targeted selection** – repeat the process described in B.4 for randomly selecting a patch in which to locate the ground trap.
  - a. Do not exclude the patch selected for the elevated trap from consideration.
  - b. If the same patch selected for elevated trap placement is randomly selected, place ground trap on the opposite side of the selected patch or > 2 meters from the elevated trap such that the elevated trap cannot re-direct litter particles toward or away from the ground trap.
- **Random selection** – continue using the randomized sampling cell locations in sequential order as described in B.4
  - a. Assess the suitability of the next potential sampling cell that has not previously been sampled or rejected.
  - b. Reject the trap location if the selected strip is < 2 meters from the elevated trap or if conditions prevent placement of stakes in all four corners of the selected cell.

1. Navigate to the SW corner of the selected cell and place a pin flag (Figure 6, Appendix G).

2. Delineate the 3 m x 0.5 m sampling cell that will be used for the ground trap using meter tape and compass or laser rangefinder to ensure that the trap is oriented to the cardinal directions.

3. Hammer in brightly colored or aluminum stakes in each of the four corners leaving ~20cm visible above ground.

   - a. At sites/plots with shallow soil or high presence of rocks that preclude placement of stakes, mark the sampling cell in an alternative appropriate method that is acceptable to the site host. Plots cannot be rejected from ground trap placement due to the presence of rocks.

4. Remove all large leaves, large fronds, and all fine woody debris > 50 cm length and < 2 cm diameter from within the ground trap area.

   - a. It is not necessary to remove small leaves, fronds, etc. that are normally sampled with the elevated litter traps.

B.6 Elevated trap construction and installation

1. Center square trap frame over pin flag placed in the center of the selected sampling cell.

2. Mark trap corners with pin flags

   a. The trap frame is 70.7 cm wide. Since a clip strip cell is 50 cm wide, trap legs will be anchored 10 cm into the adjoining cells on either side of the selected cell.

3. Hammer non-oxidizable metal stakes into ground at the pin flag locations to anchor trap legs, leaving 50 cm above ground

4. Attach trap legs to square frame

   a. Legs may optionally be glued in place.

5. Cut the trap legs so that, once installed, the square frame is level (use bubble level to check), approximately 0.8 m above the ground.

   a. Do not reject trap location if woody vegetation will be located beneath the trap, provided vegetation does not affect the shape/sag of the litter trap mesh.

   b. If possible, do not manipulate existing vegetation, though some clipping of branches is allowed at sites with continuous mid-level vegetation where a suitable location would otherwise not be available.

6. Slide trap legs over stakes.

7. Use permanent marker and meter stick to draw 50 cm line along one side of the trap frame. This will be used during collection bouts to assess qualifying material.

8. Attach screen to square frame with the provided zip ties ().

   a. The pre-cut screen is larger than the trap area and should not be taut across the trap, a minimum of 20 cm difference between the plane of the trap frame to the lowest point in the mesh is ideal to prevent litterfall from blowing away. >20 cm sag may be employed as necessary to accommodate high litter production sites (e.g., deciduous forest).

9. Physically label the trap with trapID and subplotID by writing the information on one leg of elevated trap with a permanent marker or by affixing a metal tag with the trap information on it, to the trap frame. (**Figure 10**)

10. If trap is ready to begin collecting litter material, record **addDate** as the **setDate** for the first collection bout.

Figure 9. Fully constructed elevated litter trap.

Figure 10. Label elevated traps with the trapID either written on a leg with permanent marker (right) or stamped into a metal tree tag (left)

B.7 Record data about trap deployment

When this protocol is implemented at a site, EVERY Tower Plot must be assessed for presence of qualifying vegetation.

1. For each plot record:
   - **addDate:** date of initial deployment
   - **plotID**, **subplotID**, **clipID**
   - **targetTaxaPresent: Yes/No.** Does the plot contain vegetation that qualifies for inclusion in litter sampling?
   - **trapType:** elevated or ground
   - **trapSize:** 0.5 if trapType = elevated ; 1.5 if trapType = ground
   - **trapPlacement:** random or targeted (this must be the same for *all Tower Plots* at a site and are pre-populated on mobile app)
   - **Remarks:** Free text entry about trap deployment

B.8 Annual Re-survey of Non-qualifying plots

At sites that qualify for litter sampling but do not have litter traps deployed in all Tower Plots, non-qualifying plots (i.e. **targetTaxaPresent** =No) must be reassessed annually and new traps set if the plot now contains:

- 1 or more individuals with stem diameter ≥ 10 cm or;
- 10 or more individuals with stem diameter ≥ 5 cm

If vegetation structure sampling was completed in a non-qualifying plot the previous year, re-assessment may be completed from the DSF.

1. Download records from 'VST: Apparent Individuals [PROD]' Fulcrum application for non-qualifying plots
2. If the criteria above are not met, create new 'LTR: Trap Deployment [PROD]' record for the plot with current date as **setDate** and **targetTaxaPresent** = 'No'
3. If the criteria above are met, flag this plot for new deployment and follow guidelines in the preceding sections of this SOP.

If vegetation structure sampling was *not* completed in the non-qualifying plot the previous year a field visit is required to assess the plot.

1. Include visit to non-qualifying plot in the spring data collection bout at sites with seasonal sampling strategy or in first bout of the calendar year at sites with hybrid or year-round sampling strategy.
2. Survey the plot for qualifying vegetation (RD[16]).

3. If the plot now qualifies, record **targetTaxaPresent** = Yes and deploy traps. If the plots still does not contain qualifying vegetation, create new Deployment record for each previously assessed plot with **targetTaxaPresent** = No, as in past sampling years.

SOP C   Field Sampling
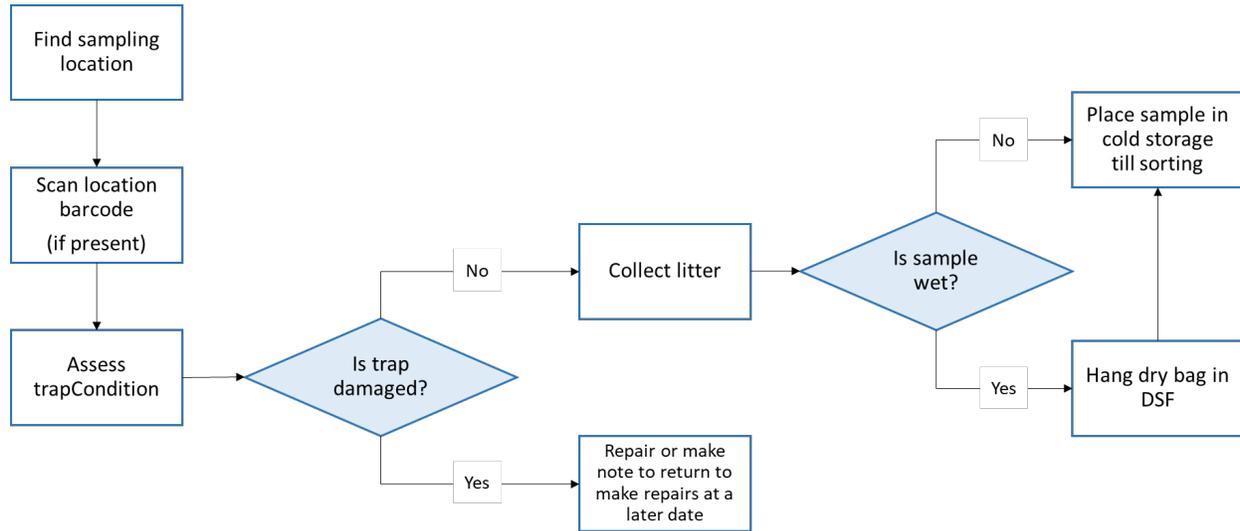
Figure 11. Workflow diagram for field collection of litter traps.

Data entry guidelines and instructions for using the 'LTR: Field Sampling [PROD]' Fulcrum application are available in the Fulcrum Manual for this protocol, available on the SSL.

C.1   Litter collection – Elevated traps

1. Navigate to plot.

2. Verify trapID from trap label, select trapID record from fulcrum drop drown to create a new field record.

3. Assess and record the **trapCondition** (Table 7).

   - *Note* - Do not use trap blocked for mounded snow. If there is too much snow in the trap to collect, skip the trap until the next collection bout, *do not create a field collection record*. The auto-calculated daysOfTrapping field will capture the extended collection period for the trap.

4. If the trap is not in good condition, discard the litter haphazardly around the trap, then make necessary repairs. Broken traps should be replaced immediately if possible.

   a. A damaged trap must be replaced or repaired within one week if repair/replacement is not possible at the time of collection. Note the date on which trap was repaired/replaced and reset; this will be the **setDate** for the next collection bout.

   b. There is no defined threshold for when litter should be discarded from traps with holes (HO). As the size and location of holes in the mesh that may allow material to be lost varies based on the dominant vegetation at a site, it is at the discretion of the technician collecting

litter to determine if the sample should be discarded due to the presence of holes. If it is likely that < 5% of mass has been lost through the holes, material may still be collected with a **trapCondition** code = 'OK'. Holes should still be repaired.

Table 8. trapCondition codes for paper datasheets

Code	Description
OK	Litter collected - Trap in good shape, no issues
TE	Litter not collected – Trap empty
HO	Litter not collected - Holes large enough for leaves to pass through. Holes near the base of the screen (the lowest hanging point) are of greater concern than holes on the side of the screen.
TB*	Litter not collected – trap blocked. Large branches or leaves (especially palm fronds) present in the trap which may have prevented trap from collecting litter or diverted falling litter away from the trap
TT	Litter not collected – trap tilted ≥ 10° (use clinometer on compass to measure)
RE	Litter not collected – trap broken
PF	Litter collected – Trap previously flooded

    c.   If the trap is in good condition (OK) continue with collection procedure.

    d.   If the plot contains needle bearing species that may be capable of passing through the mesh (Figure 12and others), spread a cloth below the trap to catch any material that falls through during collection to save as part of the sample.

5.   Discard litter > 50 cm in length, this material is not reliably collected in the elevated traps and is sampled in ground traps.

    a.   All woody material > 2 cm diameter is quantified according to the Coarse Downed Wood (CDW) protocol (RD[08]). Use calipers to measure diameter of woody branches

    b.   Discard branches > 2 cm at narrowest point

    c.   For branches that taper to ≤ 2 cm, cut off and discard the portion > 2 cm diameter; drop discarded portion of branches haphazardly (i.e. do not group or stack discarded material) beside the elevated litter trap

6. Transfer all other material, including parts hanging out of the trap, into the cloth bag designated for elevated trap litter.

Figure 12. Use of ground cloth to catch trap throughfall.

Note: Cotton bags of different size may be utilized throughout the year to accommodate variable volume of material in traps (e.g. larger bags during senescence or if snow is present in traps; smaller during periods of low production).

7. Using a pre-printed label template, handwrite in clipID, date, trap type, and technician name (Figure 13), and place inside bag prior to collecting material

    a. If material from a single trap does not fit in a single cloth collection bag, create a duplicate tag for the second bag and add "1 of 2", "2 of 2" to each tag. Record additional **bagID** in the remarks field of the mobile app and pool contents of each bag for sorting, drying and weighing.

Figure 13. Example field collection label

8. Knot cloth bag to prevent material from falling out while in transport, do not use draw strings if present on bags

9. The mobile application for field sampling is a 'flat app', there are no parent/child relationship within records. Each trap visited creates a separate record with 'trapType' prepopulated from the Deployment application based on the selected trapID. Record the following using the 'SOP C: Field Sampling' datasheet or the "LTR: Field Sampling [PROD]" mobile application:

- **measuredBy/recordedBy**
- **setDate:** the date the trap was set/reset, if previous bout trapCondition = OK then setDate=previous collectDate, else, date that damaged trap was replaced /repaired and reset.
- **boutNumber:** used for internal work tracking and construction of eventID, resets with the *calendar year in which a trap is collected, not the year it is set.*
- **yearBoutBegan:** The calendar year the collection bout began (i.e. the year associated with the **collectDate** for the first trap collected in a given bout) – typically the current year except in rare cases where a collection bout begins late December and does not conclude until early January.
- **eventID:** (auto-generated by fulcrum) use the format LTR.yearBoutBegan.siteID.boutNumber (ex. LTR.2016.TREE.06)
- **collectDate**: use *YYYYMMDD* format.
- **collectTime:** 24hr *hh:mm* format.
- **plotID:** xxxx_### - assigned by NEON Science.
- **subplotID:** see Appendix F for a plot map
- **clipID** (aka **trapID**)**:** unique identifier for trap location within the plot (format: plotID + '_' + clipCellNumber ex: OSBS_033_145)
- **trapType:** Elevated
- **trapCondition:**

- **bagID:** transcribe from cloth bag, this is a unique string, written on the individual bag. In the event that a tag is lost, metadata can be recovered by linking value to the fulcrum record for the collection event.
- **trapReset**: Yes, No
- **toxicodendronPossible:** 'Yes', 'No'. Report 'Yes' if *Toxicodendron sp.* is present anywhere in the plot, even if not present in trap. There is no need to conduct a full survey of the plot, report toxicodendron Possible=Yes if *Toxicodendron sp.* is visible in any direction from the trap.

10. If *Toxicodendron spp*. are present and *Toxicodendron* tissue may be present in the trap:

    a. Follow the guidelines established in TOS Standard Operating Procedure: Toxicodendron Biomass and Handling (RD[12]) and Appendix H to minimize exposure to toxic oils and for guidance on how to clean equipment.
    b. Collect sample in pre-weighed paper bag rather than the cloth bag.
    c. Label sample bags that may contain Toxicodendron with a sample warning label so samples will be handled with appropriate caution during downstream processing.
    d. Record remarks if necessary

C.2   Litter collection – Ground traps

1. Locate stakes marking ground trap location

2. Verify trapID from trap label, select trapID record from fulcrum drop drown.

3. Assess and record **trapCondition** (Table 9)

Table 9. Modified trapCondition codes for ground traps

Code	Description
OK	Litter collected –Trap in good shape, no issues
TE	Litter not collected – Trap empty
TB	Litter not collected – trap blocked. Large branches or tree > 10 cm diameter have fallen over trap which may have diverted falling litter away from the trap
PF	Litter collected – Trap previously flooded

If trap condition is blocked (code = TB), do not collect. If obstruction cannot be cleared, move ground trap to a new location from the clip strip list using either the random or targeted approach described in SOP B.
  o   Record new **clipID** in LTR: Trap Deployment app
  o   Clear all qualifying litter from the new clip cell.

o <u>Do not collect</u>

3. Wrap nylon cord around the four staked corners of the ground trap, delineating the trap edges.

4. Identify qualifying litter, including all litter particles (e.g. leaves, rachi twigs) which are:

    a. > 50 cm length and
    b. < 2 cm diameter (averaged between major and minor axes if elliptical) and
    c. < 2 m from soil surface (suspended litter, caught in overhanging vegetation, if within the 0.5 m x 3 m sampling cell, qualifies)

5. Material found in ground traps clearly originating from herbaceous plants present in the ground traps, that would otherwise qualify for collection in the herbaceous clip harvest protocol (RD[18]) may be excluded from ground trap collections.

6. Cut off and discard portions of qualifying litter which extend beyond trap edges, retaining only the portion which lies within trap perimeter, even if the retained portion is < 50 cm in length (**Figure 14**).
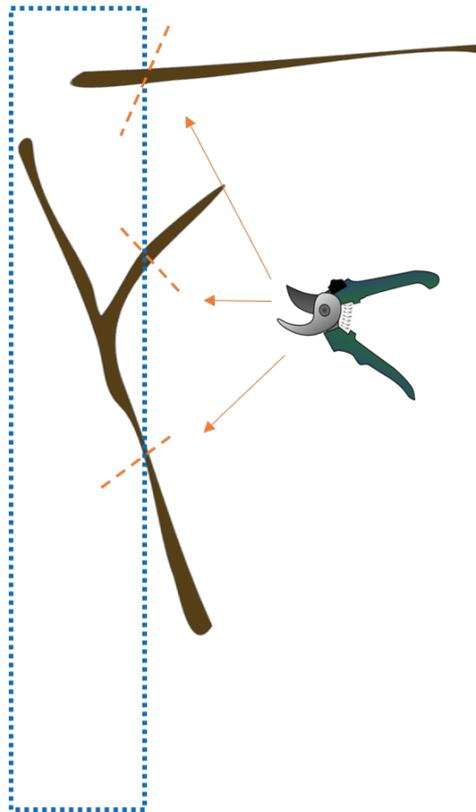
Figure 14. All particles >50 cm length qualify for collection (both particles here are > 50 cm), but only the portions located within ground trap boundaries (blue dotted line) are collected.

7. Cut off and discard portions of woody branches > 2 cm diameter (**Figure 15**).
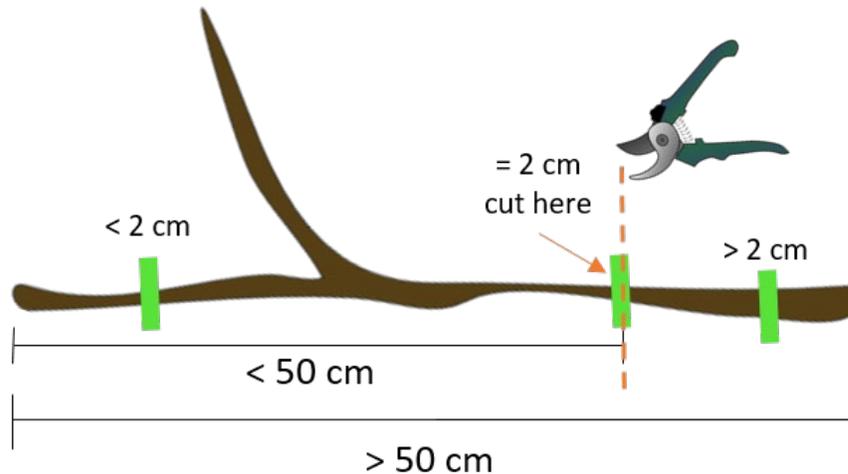
Figure 15. Discard portions of a qualifying particle that are > 2 cm diameter even if the resulting particle is < 50 cm in length.

8. Collect all remaining qualifying litter from within the ground trap, transfer material to a uniquely numbered cloth bag

   a. Pieces may be cut to smaller lengths if they are too long to fit in the cloth collection bags

9. Move remaining litter that is too large to qualify for collection outside of the ground trap boundaries to avoid re-assessing in future collection bouts

10. Create a label with clipID, collectDate, trapType, technician name (Figure 13), and attach to bag.
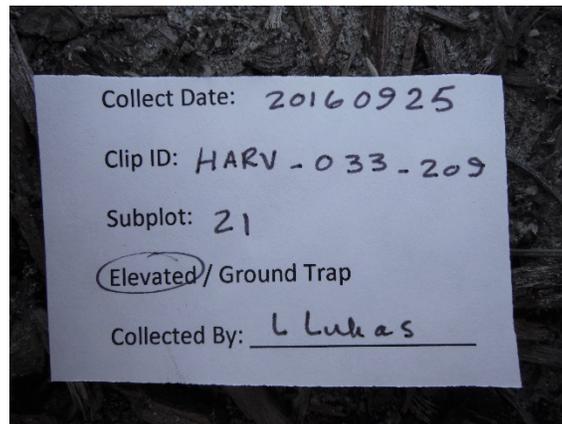
11. Knot cloth bag to prevent material from falling out while in transport, do not use draw strings if present on bags

12. The mobile application for field sampling is a 'flat app', there are no parent/child relationship within records. Each trap visited creates a separate record with 'trapType' prepopulated from the Deployment application based on the selected trapID. Using the 'SOP C: Field Sampling' datasheet, or "LTR: Field Sampling [PROD]" mobile application record:

- **measuredBy/recordedBy**
- **setDate**
- **collectDate**
- **yearBoutBegan**: calendar year of the collectDate
- **plotID**: xxxx_### - assigned by NEON Science
- **subplotID:** see Appendix F for a plot map

- **trapID:** unique identifier for trap location within the plot, designated by the clipID
- **trapType:** Ground
- **trapSize:** 1.5 if trapType = ground
- **trapCondition:** Table 9
- **bagID:** transcribe from cloth bag, this is a unique string, written on the individual bag. In the event that a tag is lost, metadata can be recovered by linking value to the fulcrum record for the collection event.
- **trapMoved**: Yes, No (if 'yes', enter new location on 'trap deployment' datasheet or pertrap mobile app)
- **toxicodendronPossible:** 'Yes', 'No' Report 'Yes' if *Toxicodendron sp. is* present anywhere in the plot, even if not present in trap. There is no need to conduct a full survey of the plot, report toxicodendronPossible=Yes if *Toxicodendron sp.* is visible in any direction from the trap.

If a *Toxicodendron sp* is present and *Toxicodendron tissues may* be sampled:

- Follow the guidelines established in TOS Standard Operating Procedure: *Toxicodendron* Biomass and Handling (RD[12]) and Appendix H to minimize exposure to toxic oils and for guidance on how to clean equipment.
- Collect sample in paper bag rather than the cloth bag.
- Label sample bags that may contain *Toxicodendron* with a sample warning label so samples will be handled with appropriate caution during downstream processing.

13. Record **remarks** if necessary.

SOP D   Post-Field Sampling Tasks

D.1   Document Incomplete Sampling Within a Site

Litterfall and Fine Woody Debris sampling is scheduled to occur at all prescribed sampling locations according to the frequency and timing described in Section 4 and Appendix C. Ideally, sampling will occur at these sampling locations for the lifetime of the Observatory (core sites) or the duration of the site's affiliation with the NEON project (relocatable sites). However, sampling may be shifted from one location to another when sampling is compromised. In general, a sampling location is compromised when sampling becomes so limited that data quality is significantly reduced.

There are two main pathways by which sampling can be compromised. First, sampling locations can become inappropriately suited to answer meaningful biological questions – e.g., a terrestrial sampling plot is compromised after road-building activities). Second, sampling locations may be located in areas that are logistically impossible to sample on a schedule that that is biologically meaningful.

A given plot must be sampled at least 50% of the bouts expected over a two-year period (i.e., a minimum of 6 bouts per year, covering multiple seasons). Plots that cannot be sampled on this schedule should be considered compromised.

After a field sampling bout is complete, review all records created during that bout to verify a record exists for *every trap at the site*. The **trapCondition** field indicates whether or not a plot was successfully sampled; for this reason, there is no need to create a problem ticket for every time a plot/trap cannot be sampled. Staff Scientists will periodically review field data to determine if a plot has become compromised. Field Science may request a review of whether a plot is compromised by submitting a problem ticket.

If site management, natural disturbance events, or regeneration/succession occur such that the cover of vegetation is significantly changed (e.g. management of woody encroachment through removal of all woody vegetation in a grassland site), submit a problem ticket to Science HQ. Such activities will prompt a re-survey of vegetation in Tower plots and if total aerial cover of remaining woody vegetation > 2 m is < 10%, sampling will be discontinued at the site.

SOP E   Laboratory Processing for Dry Mass Measurement
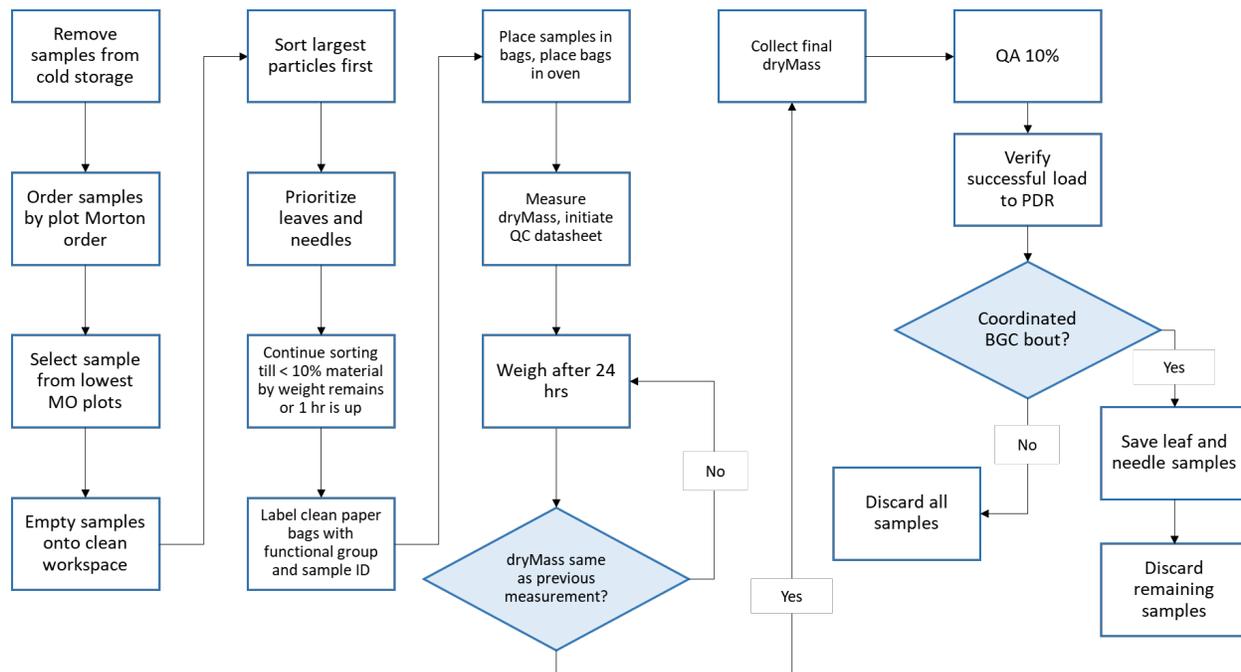
Figure 16. Workflow diagram of laboratory processing of litter trap collections, *Note, samples are discarded only after successfully loading to the NEON database (PDR)*.

E.1   Preparing for Data Capture

Mobile applications are the preferred mechanism for data entry.

Data on dry mass values from litter samples are entered in the 'LTR: Lab Mass Data [PROD]' Fulcrum application. Data entry guidelines and instructions for using this app are available in the Fulcrum Manual for this protocol, available on the SSL.

Though mobile devices are not subject to field conditions while in the lab it is, nonetheless, recommended that paper datasheets are always available to record data.

Verify that the Field Sampling application has been synched as the Lab Mass Data entry application references that application and provides a constrained list of field samples based on data uploaded from the Field Sampling effort.

E.2   Sorting, drying and weighing litter samples

- If litter and bags are very wet (i.e. dripping) or filled with snow, hang bags to melt and air dry before further processing.
- If transfer of arthropods or gastropods between sites is a concern, freeze collection bags for a minimum of 24 hours prior to sorting

- If sorting immediately following collection is not possible, store samples in refrigerator to slow decomposition
- If sorting is expected to be delayed by > 1 week, store samples in -20°C freezer.

1. Order samples according to Morton Order, lowest to highest to ensure that the lowest Morton Order plots are always sorted even if sorting of all samples is not completed.

2. Assess sampleCondition

3. The **sampleCondition** field reports the status of a field collected sample when processed in the lab. For majority of records **sampleCondition** = 'OK'; this is the default value for this field in the mobile application. However if storage requirements are not met or a field collected sample is compromised or lost after collection such that it cannot be processed for dry mass, create records for each **fieldSampleID** for which no mass samples will be generated.

Table 10. Litterfall sampleCondition values.

Code	Description	Sample handling	Field sample fate
OK	OK	No changes	Processed
Cold chain broken	Cooling requirements not met	Add remark describing storage conditions	Processed
Compromised	Sample integrity compromised during collection, drying or weighing	Discard	Discarded
Lost	Sample lost after collection and before measurement of dryMass	None, sample lost	Lost

4. Sort litter from each trap to litter functional group.

   a. If the bout will be processed for bioarchive and leaf chemistry analyses, or there is a chance it may be used for bioarchive and leaf chemistry, wear nitrile or latex-free gloves while sorting. This will prevent contamination of leaves from sweat and oils. Gloves may be re-used between samples.
   b. Clear adequate bench space in the laboratory.
   c. Empty the cloth bag filled with litter onto the bench or sorting tray (material is easier to see against a light colored surface)
   d. Remove invertebrate bycatch. Place living individuals in freezer to euthanize. Do not release invertebrates locally.
   e. Sort litter pieces to the functional groups in **Table 11** (Elevated trap collection bags) or Table 12 (Ground trap collection bags).

   Note: it may be useful to create a domain specific litter sorting guide to help streamline identification of litter material and overall sorting time.

   f. Clean off any dirt attached to litter

g. Cut or break any large seeds (i.e. dime size or greater) into smaller sections. The primary goal is to break the seed coat to allow water to escape in the drying process; if seeds cannot be cut all the way through, partial cuts are acceptable for this purpose.

Table 11. Elevated trap litter functional group codes (for use on paper data sheets, data entry has the full functional group name)

Code	functionalGroup – Description
ELVS	Leaves (including petioles, rachis, non-woody tendrils, and herbaceous stems)
ENDL	Needles/scales/awls from coniferous species
ETWI	Twigs/branches < 2 cm diameter *and* < 50 cm length
EWDY	Woody material (e.g. bark, seed cones, etc.)
ESDS	Seeds (including fruit and associated structures)
EFLR	Flowers (including associated pedicels and pollen cones)
EOTR*	Other (cactus spines, bud scales, lichen, mosses, frass, unidentifiable material, etc.)
EMXT*	Mixed, unsorted, all litter functional groups included

* Only use EOTR for material that cannot be identified or does not qualify for another category. EMXT should be used for unsorted material in which individual particles may be identifiable but sorting did not occur; use only if 1 hour sorting limit is reached, if only tiny fragments left to sort and material < 10% of total mass, or if directed by Science.

Table 12. Ground trap litter functional group codes (for use on paper data sheets, data entry application has the full functional group name)

Code	functionalGroup – Description
GLVS	Leaves and needles > 50 cm length (including petioles, rachis and non-woody tendrils)
GWDY	Woody material (e.g. bark, seed cones, etc.) > 50 cm length.
GTWI	Twigs/branches < 2 cm diameter *and* > 50 cm length
GOTR	Other (non-qualifying material previously attached to qualifying particle)
GMXT*	Mixed, unsorted, all litter functional groups included

* Use only if 1 hour sorting limit is reached, or if directed by Science. For woody particles from ground traps, still attached leaves, needles and lichen do not require special sorting or detaching.

h. Label clean, unused, paper bags to hold sorted litter functional groups from each trap. Clean undamaged, coin envelopes may be reused. Include sampling information from tag on cloth bag, as well as the appropriate **litterCode.** Choose either 8# or 25# kraft bags, or smaller, or manila coin envelope depending on the quantity of litter.

i. Tips for efficient sorting:
   1) Quickly remove large and woody particles that are fast and easy to sort (~3 mins)

2) Weigh remaining material
3) Begin sorting, prioritizing leaves and needles, then sort remainder beginning with largest particles and working down in size
4) Periodically re-weigh
5) When the weight of unsorted material is < 10% of initial weight (total collection, minus large and woody particles), stop sorting. Categorize remaining material as Mixed, unsorted.
6) Continue to next sample even if 1 hour maximum sorting time has not been hit.

5. Label the **ovenStartTime** (24 hr time, e.g. 1645 for 4:45 pm) and **ovenStartDate** (YYYYMMDD) that bags are placed in the drying oven on the paper bag.

   a. Place all bags from a given clipID or collectDate in the drying oven at the same time.
   b. *Critical step*: Labeling bags allows assessment of how long different batches of bags have been in the oven, especially when litter collections from multiple days occupy the same oven. Additionally, organizing the oven by grouping samples from a given day in the same area will streamline the re-measurement process; samples may be located and removed for weighing without requiring a complete unloading of the contents of the oven.

   A custom stamp with blank fields for all required information may facilitate consistent labeling of bags and organization of samples in drying ovens.

6. Record the number of bags and the specific litterCodes present for each clipID on the "Sorting QC Datasheet".

7. Place bags of litter (excluding ETWI, EWDY, GWDY, and GTWI) in a drying oven set to 65°C for a minimum of 48h (2d), until constant mass is attained.

8. Check the drying progress of litter bags using the generalized "Multi-Protocol Drying Datasheet" available on the NEON intranet.

   a. Check the weight of the same subset of n=10 bags every 24 hours after day 1, 2, 3, etc.

   b. Use the drying tracking spreadsheet on the SSL to determine when the drying threshold has been met. (Mean delta between t1 and t2 across all n=10 bags = 0 ± 0.05g or 0 ±1% of the t1 mass, whichever is greater)

   c. Once constant mass is achieved, remove bags from oven, label bags with the **ovenOutDate** and **ovenOutTime**.

9. Place bags of ETWI, EWDY, lignified ESDS, GWDY, and GTWI litter in a drying oven set to 105° C for a minimum of 48 hours, until constant mass is attained. If multiple drying ovens are available,

steps 4-5 and 6-7 may be occur simultaneously, otherwise, complete drying of litter material at 65° C before increasing the temperature to 105° C to dry lignified tissue. Woody material requires higher drying temperatures to release wood-bound water.

10. Check the drying progress of litter bags using the generalized "Multi-Protocol Drying Datasheet" available on the SSL.

    a. Check the weight of the same subset of n=10 bags per **collectDate** after day 1, 2, 3, etc.
    b. Calculate the difference in weight between the latest two time points for each bag.
    c. Samples are dry when the average weight difference between the latest two timepoints = 0 (Mean delta between t1 and t2 across all n=10 bags = 0 ± 0.05g or 0 ±1% of the t1 mass, whichever is greater)
    d. Once constant mass is achieved, remove bags from oven, label bags with the **ovenOutDate** and **ovenOutTime**.

11. Remove samples one at a time, weigh material from each functional group (i.e**. litterCode**) with a mass balance (0.01 g minimum measurement resolution).

    a. Prolonged exposure to ambient humidity prior to weighing may affect the measurement. If material cannot be weighed immediately, store sorted material in labeled paper bags (8# or 25# kraft bags, or similar), inside a larger, sealed, plastic bag (e.g. a black plastic garbage bag or equivalent).

    b. If necessary, 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.
    c. Only trap + date combinations from which samples were collected (i.e. **trapCondition** = OK or PF) will be available for **dryMass** data entry.
    d. Record **dryMass =** '0' for all functional groups not present in the sample (except Mixed). The mixed functional group only requires a value if this category was used.
        • If there is no parent sample (i.e. if a trap was not collected), no entry should occur for **dryMass** for any **functionalGroup**. Do not enter '0' for traps for which samples were not collected.

    e. Record the **litterMass** to the nearest 0.01 g. If material weighs <0.01g, record actual value from balance; if balance does not register material, record value as 0.005g. For large volumes of biomass that do not readily fit into a large weigh boat, use any of the following strategies:

        Crush or chop the biomass to reduce volume so it will fit into a weigh boat.
        Use an HDPE tray, 'larval tray' plastic box lid (or equivalent) instead of a weigh boat.
        Avoid splitting biomass into subgroups for weighing as this will increase the total amount of error introduced by the weighing process.

Note: paper bags or a large piece of cardboard may absorb atmospheric moisture resulting in skewed mass measurements, if using a paper or cardboard container as a weigh boat, balance must be zeroed out immediately prior to adding litter mass for each measured sample, or such containers should be avoided in humid environments.

12. Re-weigh a subset of mass samples to assess uncertainty associated with the measurement process.

   a. QA measurements must be completed by a different technician than the person who originally weighed the sample. I

   b. Per bout, for each site, select 10% of dried, previously weighed samples for re-weighing.

   1) If QA weighing does not occur coincident with initial measurement, dried samples may be stored as described above then returned to the drying oven for 24 h prior to QA weighing.

   c. Record QA weight data to the nearest 0.01 g in the **qaDryMass** field

   d. Return litter samples to temporary storage until all data have been successfully ingested into the NEON database.

13. If the collection event has been selected for bioarchive and leaf chemical analyses:

   a. Return biomass from the leaves and needle functional groups to paper bags and store together in the large plastic bag. Then seal and place in temporary storage.

   b. Samples in temporary storage can then be prepared as time permits for bioarchive and leaf chemical analysis (SOP F).

   c. All other material may be discarded in a manner approved by the site host or domain office.

14. If the collection event has not been selected for bioarchive and leaf chemistry analyses, discard all litter material and paper bags used for drying after data have been successfully ingested to the NEON database. Reuse of clean, undamaged coin envelopes is acceptable, but do not reuse paper bags for subsequent bouts.

SOP F   Processing Litter Samples for Chemistry and Archive

Samples from all elevated traps per plot from one collection event every five years are ground and submitted for chemical analyses and archive. Two functional groups, leaves and needles, are targeted. Mass from all other functional groups and from ground traps is not processed for chemistry and archive.
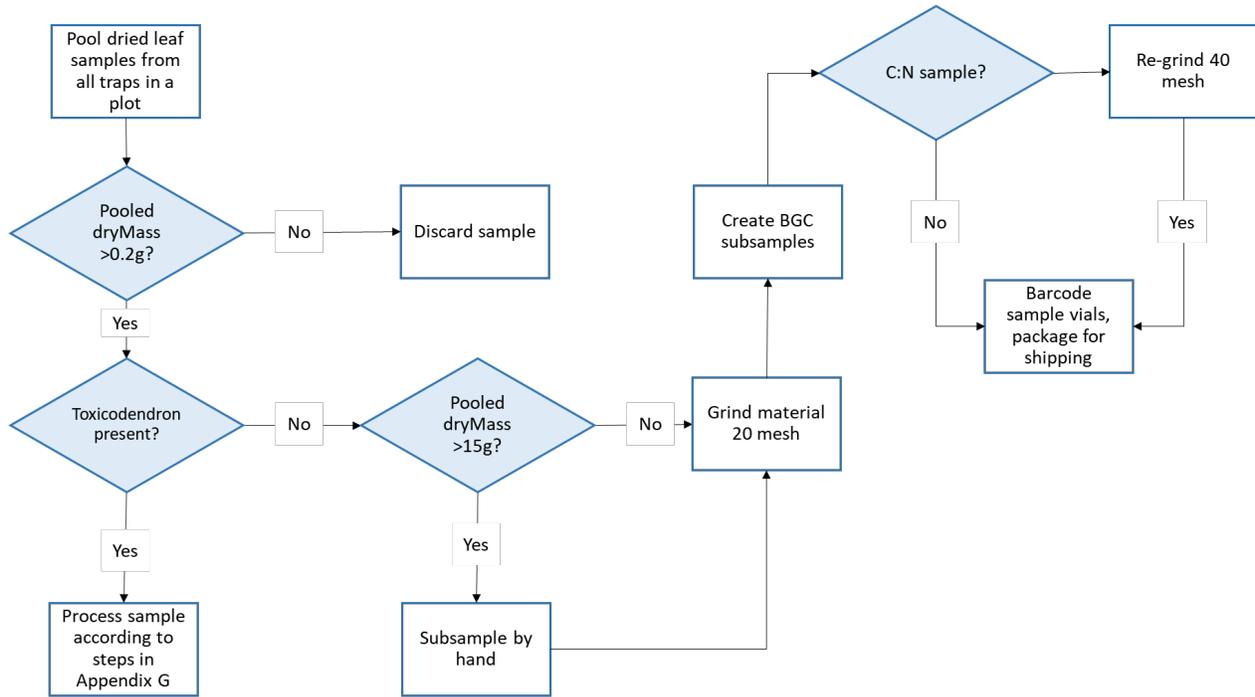
Figure 17. Workflow diagram of BGC subsampling.

Data entry guidelines and instructions for using the 'Litter: BGC Sub-Sampling [PROD]' fulcrum application are available in the Fulcrum Manual for this protocol, available on the SSL.

F.1   Timing

Conifer dominated forest: Select a collection bout for chemistry and archive from the October collection event.

Deciduous broadleaf forest: Select a collection bout from the period of peak senescence, this date may vary from site to site and from year to year.

Mixed forest: Select a collection bout from fall senescence. If needle litter production is limited, select the bout during senescence with the greatest needle mass to process for leaf chemistry.

Refer to Appendix C for suggested sampling windows, use assessment of local conditions to ultimately decide when sampling occurs.

F.2   Grinding Samples for Bioarchive and Analysis

Domains will generate a maximum of one leaf sample and one needle sample per subsample type (C:N, lignin, archive) per plot (**Figure 19**).

1. Wear a pair of Nitrile (Latex-free) gloves when handling and subsampling foliage and while handling equipment used to process material intended for chemical analyses. Gloves may be used for >1 sample but should be changed if they become visibly dirty or coated in sap or residue. Rinse with ethanol between samples.

2. In a clean container, mix dried leaves from all (one or two) elevated traps in a given plot. In another container, mix needles from all (one or two) elevated traps in the same plot. In a mixed forest system, this will generate two samples for each selected plotID, one from the 'leaves' functional group, and one from the 'needles' functional group.

   a. If *Toxicodendron* is present in the sample, process and label chemistry samples according to Appendix H.

   b. Do not save and process for chemical analysis if dry mass is < 0.2 g

   c. If the sum of dry mass from the trap(s) is > 15 g, subsample material by hand:

      i. With a nitrile gloved hand coarsely crush material into a clean container (e.g. bucket for large amounts of material, bowl for less)

      ii. Mix crushed material by hand to create an even blend

      iii. Haphazardly select one handful of crushed leaves, ~15 g to grind and process for chemical analysis

   Note: Use a balance to achieve the target mass for the first couple of samples but after this initial 'calibration,' it is okay to estimate sample quantity by eye.

3. Clean the Wiley Mill and all accessories (splitter, mesh etc.) before grinding, to remove any debris adhered to the equipment and prevent cross contamination. Use canned air or a vacuum, plus ethanol if there is evidence of resin.

4. Weigh the whole sample prior to grinding and splitting, note samples to be created based on the dry mass (**Table 13**)

5. If the combined dry mass from the selected trap(s) is >1.5 grams for a given functional group, coarsely grind material with a Wiley Mill (0.85mm, 20 mesh size).

   a. Continue grinding until all material is fully ground.

   b. Refer to **Table 13** for processing samples with dry mass <1.5 grams

Table 13. Sub-sampling guidelines for C:N, Lignin and Archive subsamples.

dryMass	Samples to create			Processing guidelines
	C:N Sample *min 0.2 g*	**Lignin Sample** *min 1 g*	**Archive sample** *min 3 g*	
< 0.2 grams	-	-	-	Do not create subsample, discard all material
0.2 – 1.5 grams	X	-	-	Do not grind, place entire sample in scintillation vial. Use gloved hand to crush if necessary to fit.
1.5 - 5 grams	X	X	-	Grind sample, distribute 1/4 sample to C:N and 3/4 sample to lignin sample
5 - 15 grams	X	X	X	Grind sample, distribute 1/4 to C:N, 1/4 to lignin, 1/2 to archive.

6. After grinding, clean material adhered to the glass cover or interior of the mill (common when grinding needles).

   a. To clean the mill, remove the glass face, loosen the screen, knock the screen into the collection jar 3-4 times, then wipe the inside of the grinding mechanism and glass plate with a paint brush to collect ground material. Flick material from paint brush bristles between samples.

   a. Use canned air or a vacuum to remove additional loose material and ethanol to remove resin.

7. Use an appropriately sized splitter or microsplitter to generate three subsamples.

   a. Split the sample once

      i. If dry mass of > 5 grams, one half will be the Archive sample

      ii. If dry mass is < 5 grams, this first half will contribute to the Lignin sample

   b. Split the remaining material in half again

      i. Half will contribute to the Lignin sample

      ii. The other half will be the C/N sample

8. Take the material from the C/N sample only and re-grind it in the Wiley Mill with the 40- mesh attachment (0.42 mm mesh). Do not re-grind lignin/elements or chemistry archive subsamples, only the C/N laboratory requires very finely ground material for analysis.

SOP F

    a. Keep grinding until no more material is observed passing through the mill, grind another 30 seconds, then stop and consider the C/N subsample complete. Do not collect leftover material that is adhered to the mill.

9. If an archive sample was generated, place a pre-labeled and barcoded scint vial on the balance and tare it, then transfer archive sample material and record **bgcArchiveMass** to nearest 0.01 g.

10. Place the remaining split sub-samples into 20 mL polypropylene scintillation vials with the barcode label already affixed and place a cryogenic label with the human readable sample ID vertically on the vial such that it does not interfere with the barcode label.

    a. SampleIDs are generated automatically by the mobile application. **Figure 19** outlines the relationship between samples and outlines how sampleIDs are assigned. SampleIDs are formatted as follows:

- **massSampleID:** (from SOP D)
  - ○ "NEON.ltr."clipID[no underscores].date.functional group code[just 'lvs' or 'ndl']
  - ○ *Example:* 'NEON.ltr.OSBS025175.20151115.lvs'
- **massSampleMixtureID:** generated in step F.2
  - ○ Remove the clip cell component from the **massSampleID**
  - ○ *Example:* 'NEON.ltr.OSBS025.20151115.lvs'
- **cnSampleID:** massSampleMixtureID + ".cn"
  - ○ *Example:* 'NEON.ltr.OSBS025.20151115.lvs.cn'
- **ligninSampleID:** massSampleMixtureID + ".lig"
  - ○ *Example:* 'NEON.ltr.OSBS025.20151115.lvs.lig'
- **archiveSampleID:** massSampleMixtureID + ".ar"
  - ○ *Example:* 'NEON.ltr.OSBS025.20151115.lvs.ar'

11. Record which samples were created.

    a. For each subsample, select the **sampleID,** scan the barcode label with the tablet (**Figure 18**).

12. Store subsamples in a cool, dry location until they can be shipped to analytical facilities or biorepository.

Figure 18. Barcode label scanning
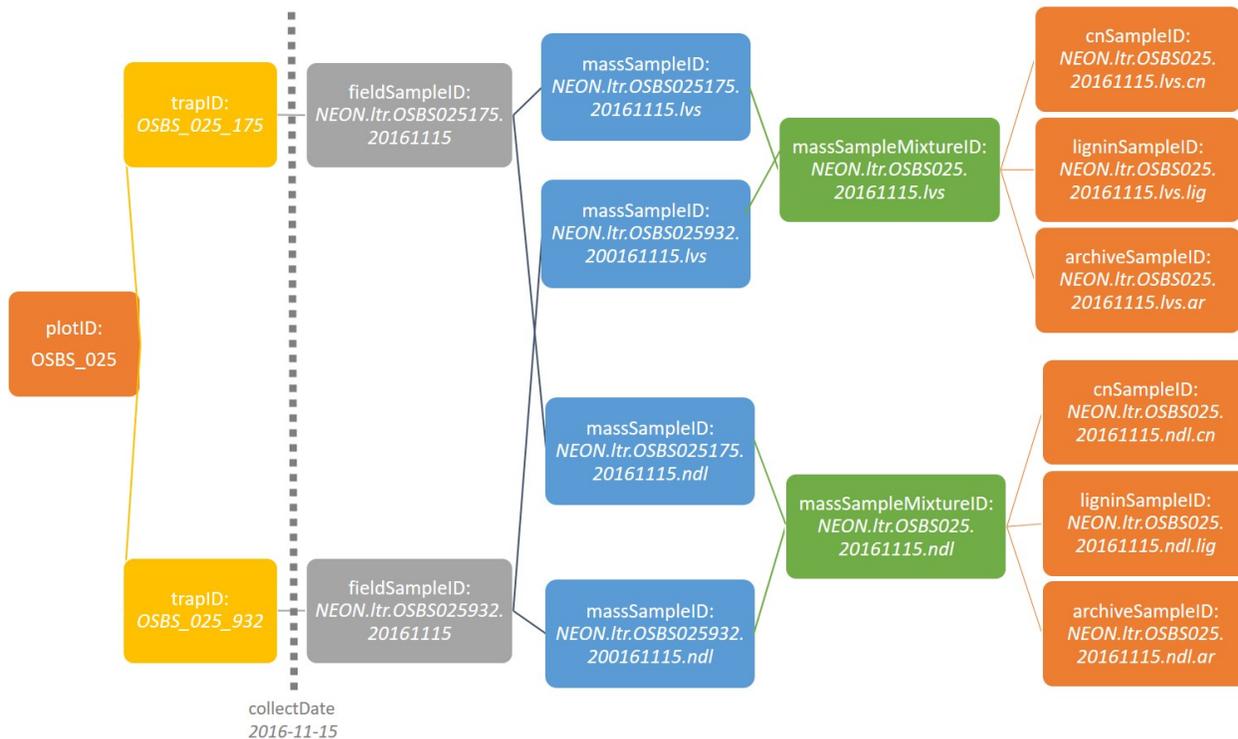
Figure 19. Anatomy of a sampleID. Note, if samples from only one trap are created, a massSampleMixtureID must still be created. Data generated will maintain traceability to original trap locations.

SOP G   Data Entry and Verification

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

Mobile applications are the preferred mechanism for data entry. Data should be entered into the protocol-specific application as they are being collected, whenever possible, to minimize data transcription and improve data quality. For detailed instructions on protocol specific data entry into mobile devices, see the NEON Internal Sampling Support Library (SSL). Mobile devices should be synced at the end of each field day, where possible. Alternatively, devices should be synced immediately upon return to the Domain Support Facility.

However, given the potential for mobile devices to fail under field conditions, it is imperative that paper datasheets are always available to record data. Paper datasheets should be carried along with the mobile devices to sampling locations at all times. 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). See RD[04] for complete instructions regarding manual data transcription.

G.1   Mobile Applications

Data are entered via the following Mobile or Desktop applications:

- **LTR: Trap Deployment**: Metadata describing trap placement
- **LTR: Field Sampling**: Metadata describing individual sampling events on a per trap per sampling date basis.
- **LTR: Lab Mass Data**: Oven-dried biomass data for each functional group per trapID per collectDate, as well as weighing QA data.
- **LTR: BGC Sub-Sampling**: Lab processing for chemistry analysis and bioarchive

G.2   Sample Labels and Identifiers

By default each sample, subsample or mixture produced by this protocol is assigned a human-readable sample identifier which contains information about the location, date, and/or functional group of the collected sample. Bioarchive and leaf chemistry samples are also associated with a scannable barcode, which does not contain information specific to sample provenance, but does reduce transcription errors associated with writing sample identifiers by hand.

If available, adhesive barcode labels should be applied to dry, room temperature containers in advance of their use in the lab (at least 30 minutes prior, but may be applied at the start of the season). Barcodes are unique, but are not initially associated with a particular sample; it is encouraged to make these up in advance. Use Type I barcode labels for litter samples. Following sample creation, apply an address label with the human readable sampleID horizonatlally on the vial.

Barcodes are scanned into the mobile application when the sample is placed into the container; only one barcode may be associated with a particular sample. Do not reuse barcodes. If a barcode is associated with multiple samples, the data ingest system will throw an error and refuse to pull in entered data. If multiple vials or containers are required to contain a sample from one trap, place the barcode on the outer container that will hold all vials associated with just that sample.

Data and sample IDs must be entered digitally and quality checked prior to shipping samples to an external lab.

G.3   Entering and uploading field data

1. For data collected on paper datasheets: Transcribe data into appropriate data entry application in accordance with data entry and data QA/QC protocols (AD[06], RD[04]).

2. Use the Data QA/QC Checklist: LTR, available on the SSL, to guide review of entered data

3. Upload data collected on mobile app to the NEON server

4. If this is the first bout at a site or a trap had to be moved to a new location and data were recorded on paper data sheets, transcribe data from the '**Trap Deployment**' Datasheets to the "**LTR: Trap Deployment**" application.

5. If **trapMoved** = 'Yes' for a given field collection record, record data for new **clipID** in the trap deployment application.

6. Update permanent digital versions of the "clip-strip coordinate" lists with **status** and **date** grid cells were used.

7. Once all data from the most recent sampling bout have been collected and transcribed, submit data for ingest to the NEON database according to the guidelines provided in RD[04].

G.4   Equipment maintenance, cleaning, and storage

1. Charge/replace laser rangefinder batteries, if necessary.
2. Charge GPS unit.
3. Clean grinding mill and splitters.
4. Rinse field collection bags as needed. Do not use soap. Water only.

SOP H   Sample Shipment

Only leaf and needle biomass samples from Litterfall collection bouts scheduled for chemistry and archive are shipped to external facilities. Before shipping samples, verify with Domain Manager and cross-check the TOS multi-year schedule on the NEON intranet to ensure that the current year is scheduled for litter chemistry and archive. Follow the instructions in NEON Protocol and Procedure: Shipping Ecological Samples, Sensors, and Equipment (RD[19]) in order to ship samples to external laboratories for chemical analysis and to the biorepository for archive. Follow the sample shipping timelines outlined in **Table 4**.

8   REFERENCES

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APPENDIX A   QUICK REFERENCES

A.1   Delineating the clip cells for litter trap placement

LOCATE AND ASSESS POTENTIAL CLIP CELL

STEP 1 – Locate southwest corner of sample plot - plot coordinate (0,0)

STEP 2 – If no woody vegetation is present in the plot, record targetTaxaPresent=No

STEP 3 – Select first available clip-strip location from Work Order list.

STEP 4 – Use Appendix F to determine x-y coordinates for the trap.

STEP 5 – Locate Y-coordinate with laser rangefinder in HD mode (azimuth 0°), place pin flag.

STEP 6 – Locate clip cell centroid (elevated trap)

STEP 6b – Locate clip cell SW corner (ground trap)

STEP 7 – Assess suitability of clip-strip. Reject if not suitable.

- ● = x, y from clipList
-     = SW corner of clip strip
- ● = SW corner of ground trap
- ★ = Center of elevated trap

3 m

0.5 m
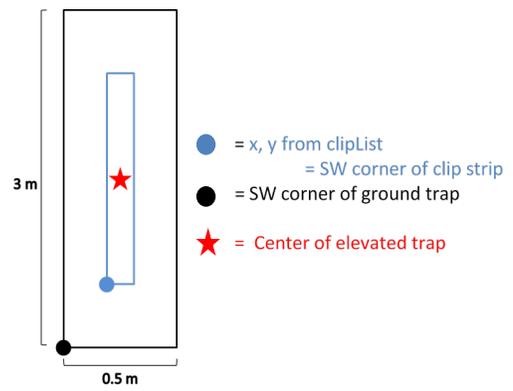
DELINEATE 0.5 M X 3 M CLIP-STRIP

STEP 1 – Place one stake in SW corner of clip-cell.

STEP 2 – Use laser range finder and tape measure to locate remaining three corners.

STEP 3 – Check distance between all four corner with ruler or tape measure. Use handheld compass to check orientation.

STEP 4 – Monument clip strip corners with aluminum stakes

A.2 Litter trap status codes

Code	Description
OK	Litter collected –Trap in good shape, no issues
TE	Litter not collected – Trap empty
HO	Litter not collected –Holes large enough for leaves to pass through. Holes near the base of the screen (the lowest hanging point) are of more concern than holes on the side of the screen.
TB	Litter not collected – trap blocked. Large branches or leaves (especially palm fronds) present in the trap which may have prevented trap from collecting litter or diverted falling litter away from the trap
TT	Litter not collected – trap tilted ≥ 10° (use clinometer on compass to measure)
RE	Litter not collected – trap broken

A.3 Example field collection label
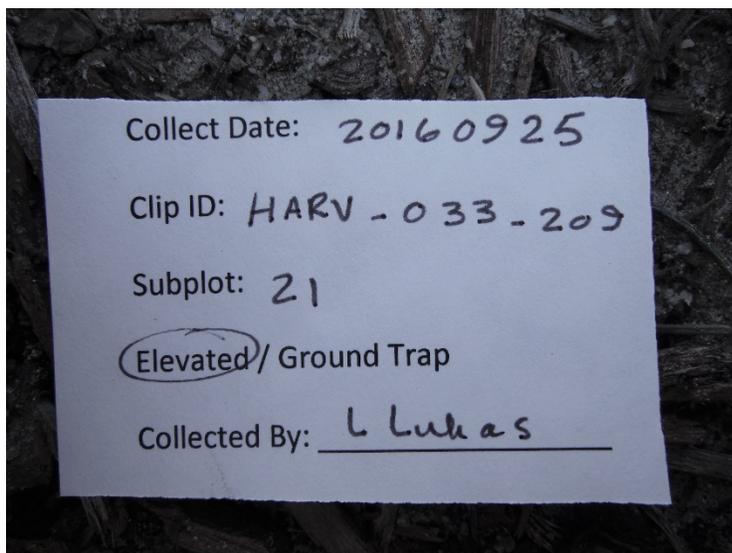
APPENDIX B   REMINDERS

Before leaving the DSF, double check that you have

- Plot map

- Trap repair supplies

- Uniquely numbered cloth bags

After completing field collection:

- Inventory all samples, make sure your count of bags matches the number of records for successfully collected traps

- Place all bags in refrigerator or hang to dry if wet

Before sorting

Pre-label several paper bags with the fieldSampleID prior to sorting. Add functional group designator once sorting is complete and functional groups are known.

APPENDIX C   ESTIMATED DATES FOR ONSET AND CESSATION OF SAMPLING

The dates in the table below are estimated from satellite MODIS-EVI phenology data averaged from 2005-2014 (Didan 2015). Dates presented here are only a guide, and are derived according to the logic presented in Section 4.2. Because individual years may vary widely from the average dates provided below, it is essential that domain staff monitor real-time conditions to determine when to start (and stop) sampling, as described in Section 4 of this protocol.

The 'average Greenness Increase' date is a proxy for the beginning of spring. At seasonal sampling sites, this indicates when collection of winter litterfall should occur and the beginning of the regular sampling season. Beginning and average end of senescence indicate the period of time during which leaf fall occurs and fall sampling interval is necessary at sites dominated by deciduous vegetation. NLCD vegetation classification listed is based on the dominant vegetation found in the tower airshed.

Sampling schedules may be modified based on local conditions, for example, if the NLCD vegetation class is identified as 'Mixed Forest' but plots are almost entirely coniferous trees, sampling may be shifted to 'Monthly, Year-round' even though the table specified 'Spring + Senescence' or 'Hybrid' sampling schedule. Dates are only listed for sites with forests where intensive sampling during fall senescence is anticipated; all other sites are sampled once a month all year or not at all.

Non-forested sites with estimated < 25% cover may be subject to additional vegetation surveys to determine mean cover of woody vegetation > 2 m across all tower plots (see RD[16]).

Table 14. Estimated sampling dates

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
01	BART	Mixed Forest	Random	Hybrid	28-Apr	9-Aug	22-Oct	May	
01	HARV	Mixed Forest	Random	Hybrid	22-Apr	7-Aug	31-Oct	April	

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
02	BLAN	Deciduous Forest/ Pasture Hay	Random	Hybrid	13-Mar	13-Jul	10-Nov	March	
02	SCBI	Deciduous Forest	Random	Hybrid	27-Mar	3-Aug	19-Nov	March	
02	SERC	Deciduous Forest	Random	Hybrid	17-Mar	9-Aug	21-Nov	March	
03	DSNY	Grassland Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
03	JERC	Mixed Forest	Random	Hybrid	23-Mar	10-Jul	23-Oct	February	
03	OSBS	Evergreen Forest	Random	Monthly, Year-round				March	
04	GUAN	Evergreen Forest	Random	Monthly, Year-round				October	
04	LAJA	Cultivated Crops		None					schedule survey for qualifying vegetation every 5 y.
05	STEI	Deciduous Forest	Random	Spring + Senescence	29-Apr	8-Aug	12-Oct	May	
05	TREE	Deciduous Forest	Random	Spring + Senescence	27-Apr	7-Aug	15-Oct	May	

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
05	UNDE	Deciduous Forest	Random	Spring + Senescence	30-Apr	8-Aug	09-Oct	May	
06	KONA	Cultivated Crops		None					
06	KONZ	Grassland Herbaceous	Targeted	Spring + Senescence	02-Apr	30-Jul	3-Nov	November	Recheck targetTaxaPresent = 'No' annually for qualifying vegetation
06	UKFS	Deciduous Forest	Random	Spring + Senescence	22-Mar	28-Jul	28-Nov	November	
07	GRSM	Deciduous Forest	Random	Spring + Senescence	2-Apr	3-Aug	6-Nov	September	
07	MLBS	Deciduous Forest	Random	Spring + Senescence	17-Apr	8-Aug	6-Nov	September	
07	ORNL	Deciduous Forest	Random	Spring + Senescence	17-Mar	24-Jul	15-Nov	April	
08	LENO	Woody Wetlands	Random	Spring + Senescence	9-Mar	17-Jul	4-Dec	July	
08	DELA	Woody Wetlands	Random	Spring + Senescence	1-Mar	17-Jul	15-Nov	August	
08	TALL	Evergreen Forest	Random	Monthly, Year-round	16-Mar	14-Jul	4-Dec	May	
09	DCFS	Grassland Herbaceous		None					schedule survey for qualifying

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
									vegetation every 5 y.
09	NOGP	Grassland Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
09	WOOD	Grassland Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
10	CPER	Grassland Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
10	RMNP	Evergreen Forest	Random	Monthly, Year-round	5-May	2-Aug	11-Oct	October	
10	STER	Cultivated Crops		None					
11	CLBJ	Grassland Herbaceous	Random	Hybrid**	27-Feb	28-Aug	11-Nov	January	
11	OAES	Grassland Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
12	YELL*	Shrub Scrub	Random	Monthly, Year-round	5-May	12-Jul	17-Sep	October	

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
13	MOAB	Shrub Scrub	-	None					schedule survey for qualifying vegetation every 5 y.
13	NIWO	Evergreen Forest	Targeted	Monthly, Year-round				September	Recheck targetTaxaPresent = 'No' annually for qualifying vegetation
14	JORN	Shrub Scrub	-	None					schedule survey for qualifying vegetation every 5 y.
14	SRER	Shrub Scrub	Targeted	Monthly, Year-round	1-Mar	7-Sep	18-Nov	April	Recheck targetTaxaPresent = 'No' annually for qualifying vegetation
15	ONAQ	Shrub Scrub	-	None					schedule survey for qualifying vegetation every 5 y.
16	ABBY§	Evergreen Forest	Random	Monthly, Year-round	18-Apr	15-Aug	15-Nov	October	
16	WREF	Evergreen Forest	Random	Monthly, Year-round	21-Apr	27-Jul	19-Oct	May	

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
17	SJER	Evergreen Forest	Targeted	Monthly, Year-round	7-Sep	6-Apr	3-Jun	November	Recheck targetTaxaPresent = 'No' annually for qualifying vegetation
17	SOAP	Evergreen Forest	Random	Monthly, Year-round	30-Mar	8-Jul	12-Oct	August	
17	TEAK	Evergreen Forest	Random	Monthly, Year-round	4-May	27-Jul	09-Oct	September	
18	BARR	Sedge Herbaceous		None					schedule survey for qualifying vegetation every 5 y.
18	TOOL	Dwarf Scrub		None					schedule survey for qualifying vegetation every 5 y.
19	BONA	Mixed Forest	Random	Hybrid	13-Mar	26-Jul	7-Sep	July	
19	DEJU	Evergreen Forest	Random	Monthly, Year-round				May	
19	HEAL	Shrub Scrub	Targeted	Monthly, Year-round	18-May	28-Jul	6-Sep	May	Recheck targetTaxaPresent = 'No' annually for qualifying vegetation

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence	Ground trap Collection	Remarks
20	PUUM	Evergreen Forest	Random	Monthly, Year-round				September	

* Litter sampling at YELL is suspended March 10 – July 1 each season due to annual closure of the Bear Management Area within the Tower Airshed

** High production period in January, 2-week sampling interval employed during this peak

§ Dates updated based of Field Ecologist request to better represent Tower plot phenology.

APPENDIX D SITE-SPECIFIC INFORMATION

D.1 Optimization

Statistical analyses were conducted using previously collected litter data to determine whether litter and fine woody debris current measurements were sufficiently spatially homogeneous to reduce sampling or processing intensity, eliminate redundant sampling effort when possible, and maintain robust site-level litter production estimates and the ability to detect temporal trends.

Elevated Traps: Based on an analysis of sites that had a minimum of three years of data per site, reduction of sampling effort is supported at a subset of sites (**Table 15**).

Ground Traps: Production of fine woody debris that qualifies for ground trap collection is spatially and temporally heterogeneous; at this time, reduction of number of traps sampled is not supported at any site.

Table 15. Elevated trap plot reduction.

Domain	Site	2019 Elevated Trap Number	Optimized Elevated Trap Number	Plots to Retain Elevated Traps
D01	BART	40	20	032, 070, 033, 034, 036, 037, 071, 039, 040, 041
D01	HARV	40	20	033, 034, 035, 036, 037, 038, 039, 040, 041, 042
D02	SCBI	40	20	067, 045, 046, 047, 048, 049, 050, 051, 052, 053
D02	SERC	40	20	043, 044, 045, 047, 048, 049, 050, 067, 051, 052
D03	JERC	40	20	047, 048, 049, 050, 051, 052, 053, 054, 055, 056
D05	TREE	40	30	039, 040, 041, 042, 043, 044, 045, 046, 025, 047, 048, 049, 050, 051, 052
D05	UNDE	40	20	065, 037, 038, 043, 044, 045, 047, 048, 049, 050
D07	GRSM	40	20	047, 048, 049, 050, 051, 052, 053, 054, 055, 056
D07	ORNL	40	20	036, 037, 043, 045, 046, 049, 050, 051, 052, 053
D08	TALL	40	20	051, 044, 054, 047, 042, 056, 050, 061, 046, 043

D.2 Prescribed Burns

At sites with prescribed burning, collect litter as if conducting a regular sampling bout even if one is not scheduled, prior to scheduled burn. Remove traps then replace as soon as possible following after. Dates

of removal and re-setting of litter traps do not need to be recorded as no litter production is expected during this period. Resume prescribed sampling schedule once traps are reset.

Burn sites include, but may not be limited to, the following:

Table 16. Burn sites

Domain	Site Code	Site Name
D03	JERC	Jones Ecological Research Center
D03	OSBS	Ordway-Swisher Biological Station
D06	KONZ	Konza Prairie Biological Station (Core)
D08	TALL	Talladega National Forest
D09	WOOD	Woodworth
D11	CLBJ	LBJ National Grassland
D17	SOAP	Soaproot Saddle

D.3   Fine Needled Species

Mesh size on elevated trap assembly kits is 1mm. Particles < 1mm diameter may be capable of passing through the standard mesh and may be underestimated in dry mass measurements though field crews will attempt to collect material that passes through the trap mesh during collection activities.

Sites with known coniferous species with needles < 1mm or at which needles have been observed to pass through elevated trap mesh during collection summarized in **Table 17**. Sites with fine needled species capable of passing through 1mm mesh

Table 17. Sites with fine needled species capable of passing through 1mm mesh

Domain	Site Code	Site Name	Species with needle width < 1mm
D01	HARV	Harvard Forest	*Larix decidua*
D01	BART	Bartlett Experimental Forest	*Picea rubrum, Tsuga Canadensis*
D05	STEI	Steigerwaldt Land Services	*Larix laricina*
D05	UNDE	UNDERC	*Larix laricina*
D05	TREE	Treehaven	*Larix laricina*
D16	ABBY	Abby Road	*Tsuga heterophylla, Pseudotuga menziesii* var. *mensiesii*
D17	WREF	Wind River Experimental Forest	*Tsuga heterophylla, Pseudotuga menziesii var. mensiesii*
D19	DEJU	Delta Junction	*Picea mariana, Larix laricina*
D19	HEAL	Healy	*Picea mariana, Larix laricina*
D19	BONA	Caribou Creek - Poker Flats Watershed	*Picea mariana, Larix laricina*

APPENDIX E   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 18. Equipment list – Initial trap deployment, SOP B

Item No.	Exact Brand Y/N	Description	Purpose	Quantity*
	N	Non-oxidizable metal rods (e.g. aluminum, galvanized stainless steel, or equivalent) ~1 m length	Anchor trap to sampling location	4 per trap
Yardandpool.com #MYLS	N	Aluminum stake	Mark corners of ground traps	4 per trap
Ben Meadows #100952 Forestry Suppliers #39167	N	Chaining pins or other suitable anchor	Anchor measuring tapes	2
	N	Coin	Randomize selection of patches at sites with targeted selection	1
Ben Meadows #213379 Forestry Suppliers #37184 #37036	N	Compass with mirror and declination adjustment	Locate X, Y coordinates of within-plot trap location; alternative to high-accuracy laser rangefinder (with less precise rangefinder)	1
#EG07670000	Y	Elevated litter trap assembly	Collect litter sample	40-50
Compass Tools #703512 Forestry Suppliers #90998	Y	Foliage filter	Allow laser rangefinder use in dense vegetation	2
Forestry Suppliers #91567	Y	Laser Rangefinder, ½ foot accuracy	Locate X, Y coordinates of within-plot trap location	1

Item No.	Exact Brand Y/N	Description	Purpose	Quantity*
B&H #SISOK12601	Y	Laser Rangefinder, 1 yard accuracy	Measure distances. May be used, in conjunction with handheld compass, as an alternative to TruPulse	1
Ben Meadows #122731 Forestry Suppliers #40108 #39943	N	Measuring tape, minimum 30 m	Locate clip-harvest strips within plots/subplots. Plot slope < 10 deg; grassland, savannah	1
Grainger #3CYN7	N	PVC pipe cutter	Cut PVC to length	1
Home Depot #EM81.9	N	Torpedo bubble level	Check the angle of the elevated trap	1
Grainger #1F017	N	White reflector or reflective tape	Reflective target for laser rangefinder; aids in measuring distance to target accurately	1
Grainger #2RUV1	N	CR123A battery	Spare battery for laser rangefinder	2
	N	PVC pipe glue	Permanently attach PVC from the elevated trap kits	1 jar
Forestry Suppliers #33790 #3JVC4	N	Survey marking flag, PVC or fiberglass stake	Delineate sampling area	4 per trap
RD[05]	Y	Datasheets for Litterfall and Fine Woody Debris	Record required data and metadata	Variable
	Y	Per plot or subplot Clip Lists	Identify random clip-strip locations	
	N	Random number list	Randomize selection of patches at sites with targeted selection	1

Table 19. Equipment list – Field sampling elevated and ground litter traps, SOP C

Item No.	Exact Brand Y/N	Description	Purpose	Quantity
Grainger #12U275 Grainger #2PRP6 Amazon #B001OK8MM8	N	Nylon rope	Delineate ground trap	1, 8 m
Yardandpool.com #MYLS	N	Aluminum stake	Replace stakes on damaged ground traps	4
Amazon #B016V82RKA	N	Cotton bags, uniquely numbered [1]	Carry fresh, potentially wet, litter samples	2 per trap pair
#EG07670000	Y	Elevated litter trap assembly	Replace damaged traps	2
	N	80 cm long, 0.5 in diameter PVC pipe	Replace damaged elevated trap leg pieces	As needed
	N	69.5 cm long, 0.5 in diameter PVC pipe	Replace damaged elevated trap frame pieces	As needed
	N	PVC right angle out, 3-way elbow, 0.5 in	Replace damaged elevated trap corner pieces	As needed
	N	122 cm x 122 cm, 1mm polyester window screen	Replace or repair damaged elevated trap screen	As needed
	N	6 in long, UV resistant zip-ties	Replace damaged elevated trap zip ties	As needed
	N	PVC slip coupling	Replace damaged elevated trap leg spacers	As needed
Compass Tools #703512	Y	Foliage filter	Allow laser rangefinder use in dense vegetation	2

Item No.	Exact Brand Y/N	Description	Purpose	Quantity
Forestry Suppliers #90998				
	N	Handheld caliper, 0.1 cm precision	Measure branch diameters	1
Forestry Suppliers #91567	Y	Laser Rangefinder, ½ foot accuracy	Locate X, Y coordinates of trap if thick brush prevents visual trap location in Thick brush	1
Grainger #3KMZ6 Forestry Suppliers #71166	N	Measuring stick, 1 m^2	Measure and identify/discard litter > 50 cm	1
Home Depot #300450094	N	Pruning lopper, heavy duty	Cut branches up to 2 cm diameter	1
Home Depot #EM81.9	N	Torpedo bubble level	Check the angle of the elevated trap	1
Fastenal #294561	N	Flush cut clippers	Cutting screen material or zip ties	1
	N	Screen patch kit (pieces of 1 mm screen, wire, window screen repair tape, wirecutters)	Repair minor holes in screen material	1
Grainger #2RVU2	N	CR123A battery	Spare battery for laser rangefinder	2
Herbarium Supply #361	N	General Purpose Tags, may use rite in-the-rain	Label collection bags	2 per trap pair
ULINE #S-21339	N	Sample warning pictogram label	Identify acute toxins that may cause serious eye or skin irritation. Samples contain *Toxicodendron* spp.	1 per container

[1] recommended size ~ pillowcase dimensions

[2] May also mark 50cm on plot frame with permanent marker.

Table 20. Equipment list – Laboratory processing and analysis SOPs D & F

Item No.	Exact brand	Description	Purpose	Quantity*
Grainger #1TTX2 #2AJP4	N	Paintbrushes, various sizes	For use in sorting litter	4
Fisher #S90203 #02-401-7	N	Timer	Track sorting time and limit to one hour per field sample	1
	N	Domain specific litter sorting guide	Assist with identification of litter functional groups	1
Fisher #8732115 #8732112	N	Weigh boats, various sizes	For weighing sorted material	4
Fisher #NC0516918	N	Hy back pan	Receive sub-samples generated by splitter	2 per splitter
Fisher #NC9052925	Y	Sample microsplitter, small capacity	Subsample from small volumes of ground sample. Relatively little litter mass per litterCode per trap	1
Fisher #040G-010	Y	Sample splitter, large capacity	Subsample from relatively large volumes of ground sample. Useful with fibrous leaves. Relatively large litter mass per litterCode per trap	1
ULINE #S-7798 #S-6285 #S-14719 #S-6286 #S-14720 #S-5623 #S-11485 #S-12775 #S-17208 #S-11486 #S-17209 #S-11487	N	Paper coin envelopes, assorted sizes	Contain very small masses of sorted litter for drying	20

Item No.	Exact brand	Description	Purpose	Quantity*
Grainger #12R027 #12R024 ULINE #S-7630 #S-13236 #S-11538 #S-13241	N	Paper bags, assorted sizes	Contain litter, sorted to functional group	50
ULINE #S-15706	N	12 x 18 blank newsprint paper	Clean, high contrast surface for sorting	As needed
Fisher #03-337-23C	Y	Scintillation vials with caps, 20 mL	Contain ground split samples for shipment to archive or chemical analysis	As needed
Fisher #19-176-550	N	Ethanol wipes	Quickly clean gloves, buckets, sample splitter, etc. between samples	
Fisher #15-930-C	Y	Adhesive cryo-labels (0.5 x 1.25 in)	Labeling sample containers with bgc sampleID labels	1 sheet
	Y	Type I adhesive barcode labels	Labeling sample containers with barcode-readable labels	1 sheet

R/S=Required/Suggested

APPENDIX F   PLOT MAPS

40m x 40m (top image) and 20m x 20m (lower image) Tower Plots showing the location of 0.5m x 3m clip-harvest cells (dashed blue lines). Subplot IDs are listed in gray for the 40m x 40m plot. The clip-strip coordinates provided to domain staff are supplied on a per subplot basis (red 'X' in the figures). For plot centroids, navigate 1 m North and 5 cm East from this point. To locate the clip cell / ground trap SW corner, navigate 0.5 m South and 20 cm West from the provided coordinates. Exclusion areas in 40m x 40m Tower Plots selected for Plant Diversity sampling are consistent with a 20m x 20m plot centered on the plot centroid. Clip cells in exclusion areas are not included in the randomized clipLists provided by NEON Science.
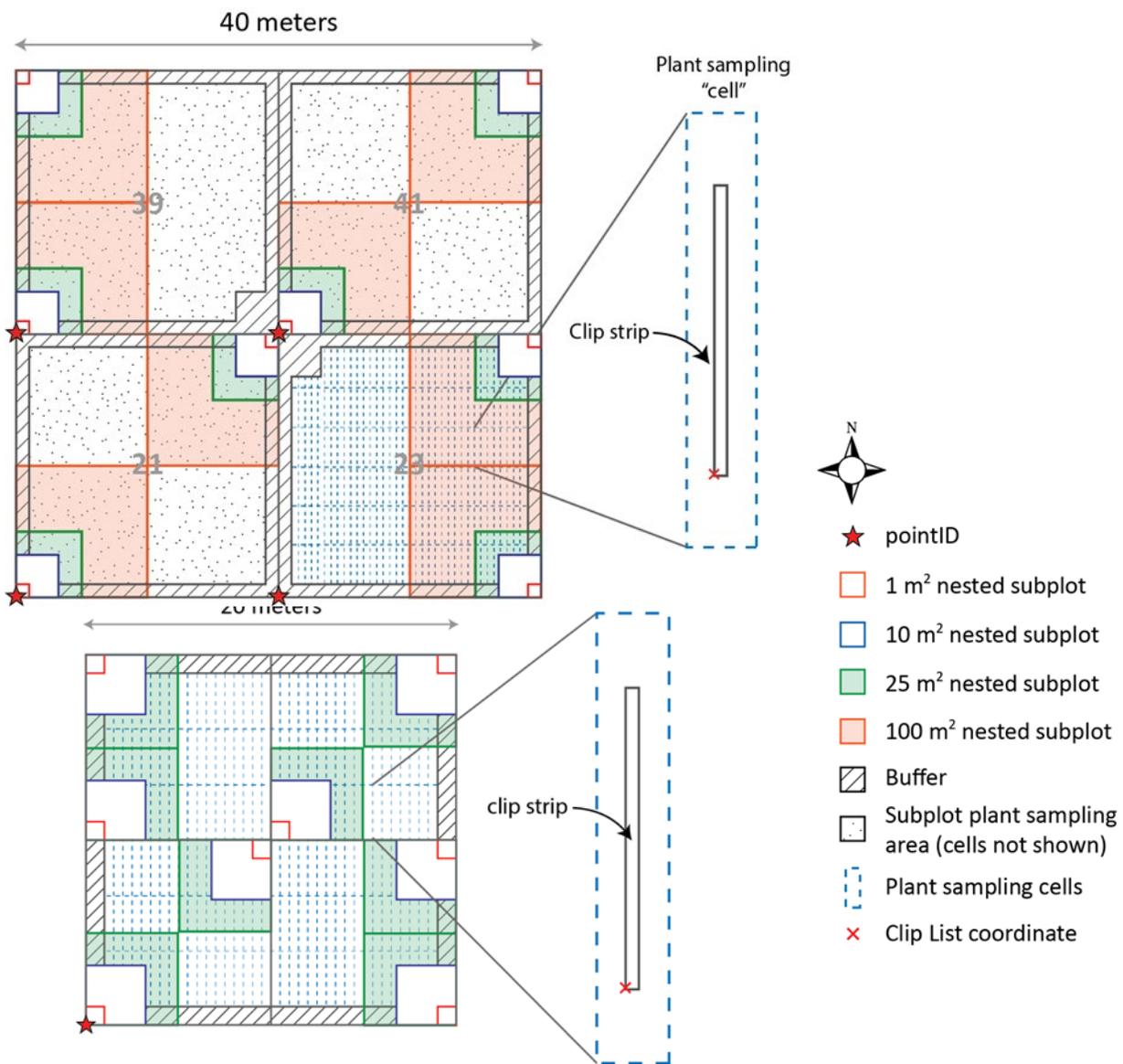
APPENDIX G   CLIP CELL NUMBER COORDINATES AND MAPS

Targeted deployment of ground and elevated litter traps (SOP B) in habitats with non-continuous cover (< 50% of the plot area) of woody vegetation requires locating Clip Strips within "patches" of vegetation with overstory species ≥ 2 m. To identify trap location within woody "patches," first map out the location of patches within a selected subplot, use a random selection procedure to pick an individual patch then use the appropriate map in this Appendix to determine which clipCellNumber should be sampled. Use **Table 21** in to find the easting and northing values associated with that Clip Strip so that it can be delineated at a known location relative to the SW corner of the 20m x 20m plot / subplot.
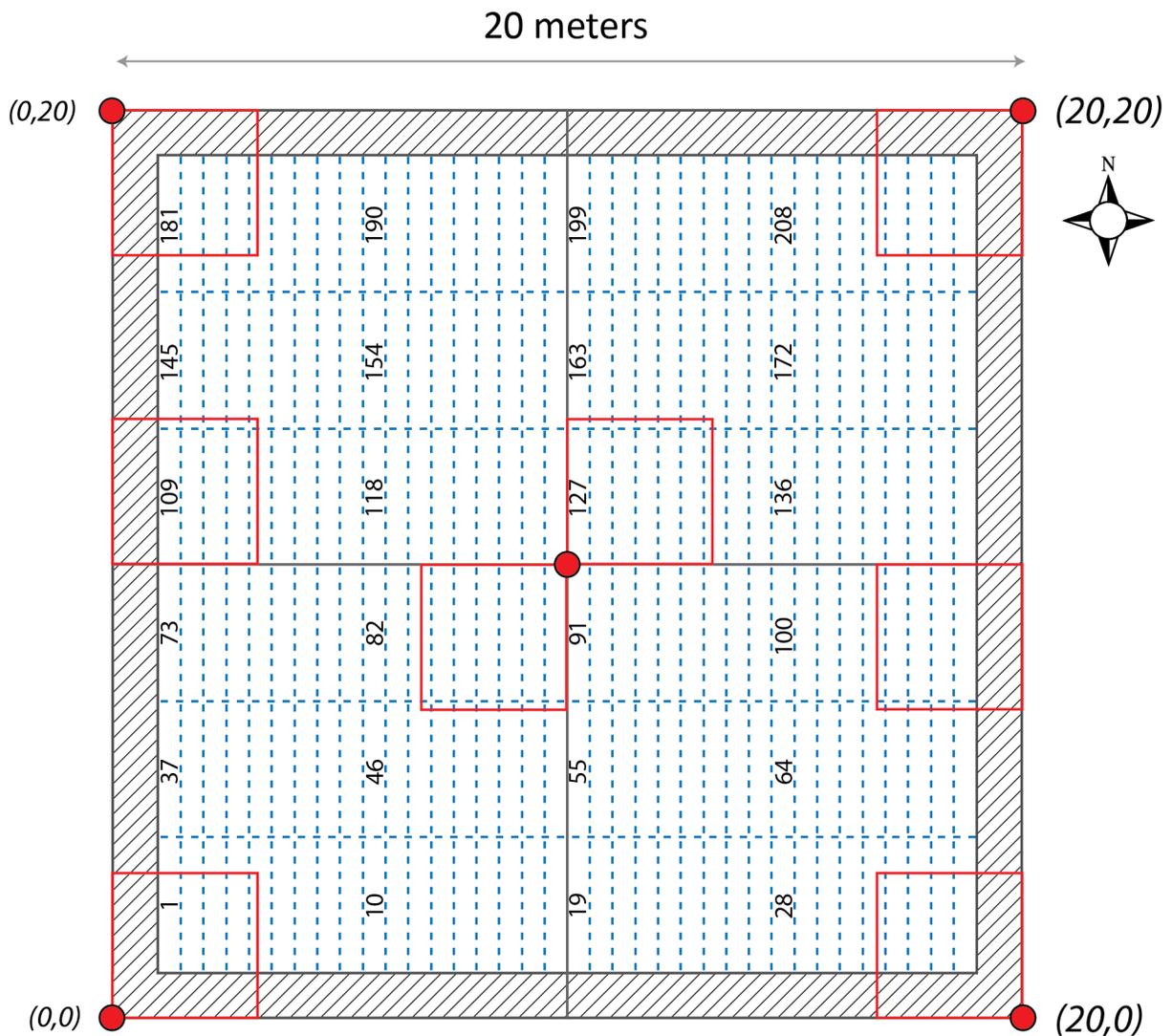
Figure 20. Map of clipCellNumbers in a 20m x 20m base plot (subplotID = 31 in provided Clip Lists). Red squares indicate nested subplots used for diversity sampling; clip cells that significantly overlap red squares are not used for litter sampling. However, clip cells with minimal overlap (e.g., 48-54, 68-72, 145-149) are considered for litter sampling.
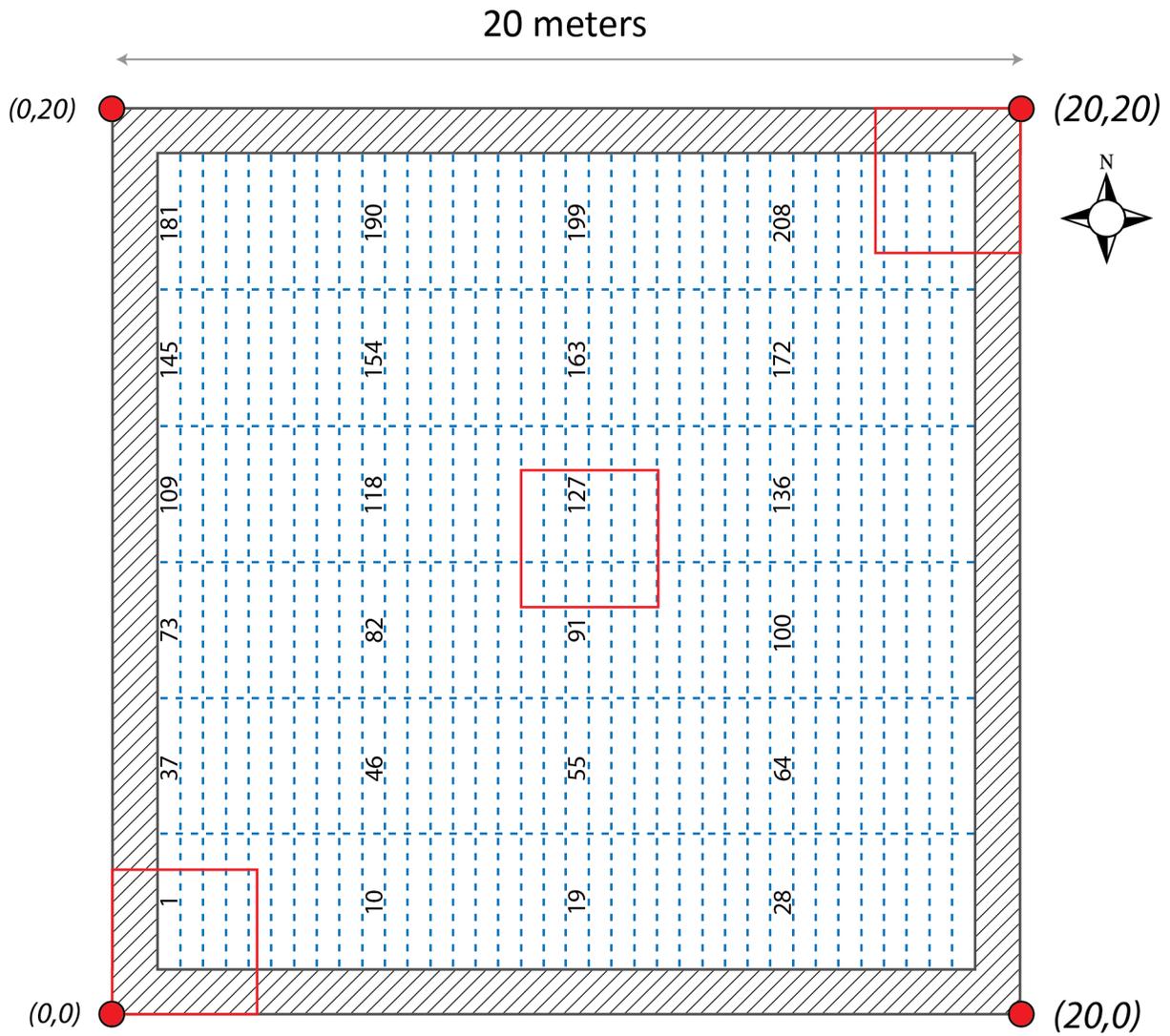
Figure 21. Map of clipCellNumbers for **subplotID = 21** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for litter sampling.
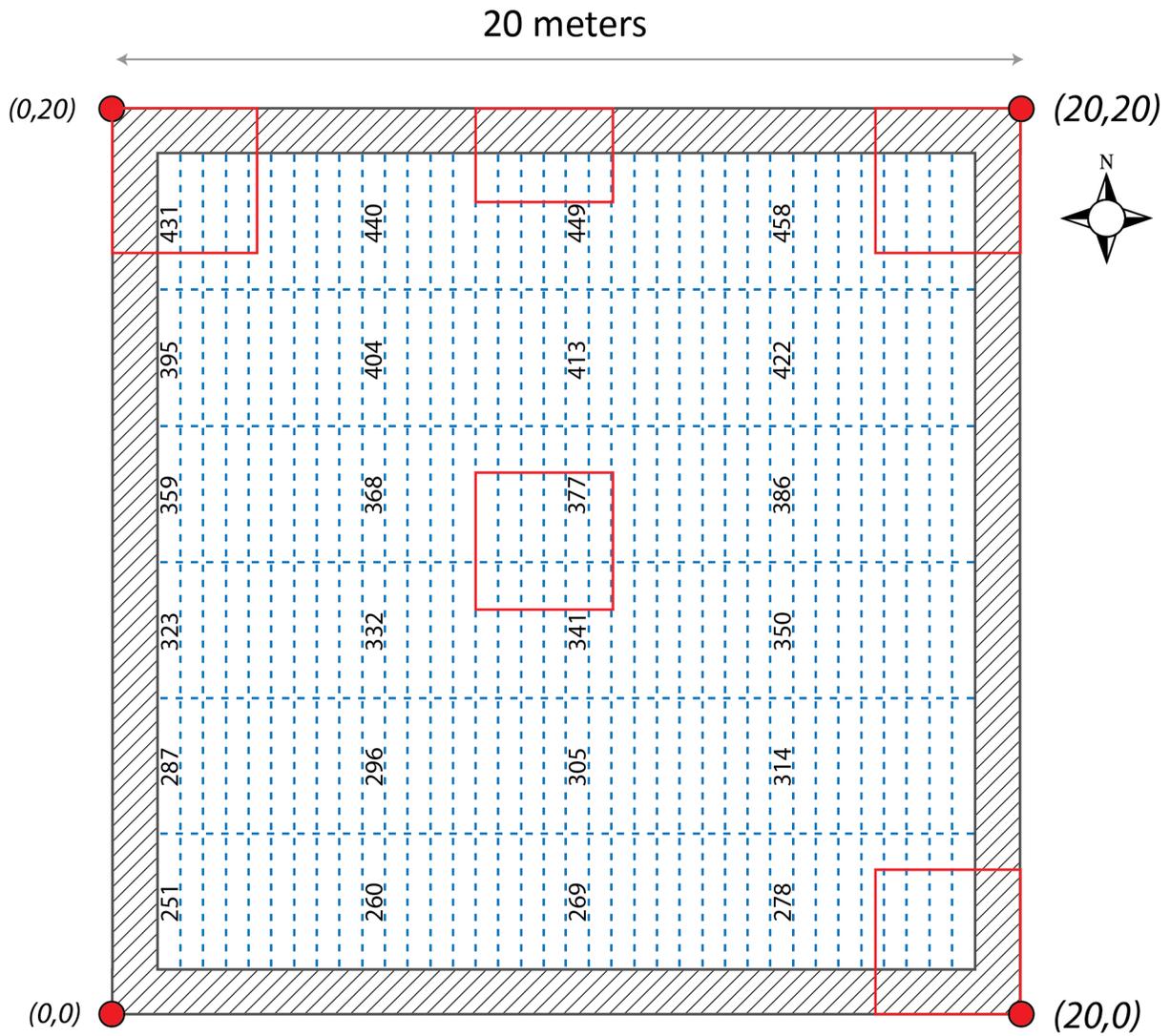
Figure 22. Map of clipCellNumbers for **subplotID = 23** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for litter sampling.

20 meters
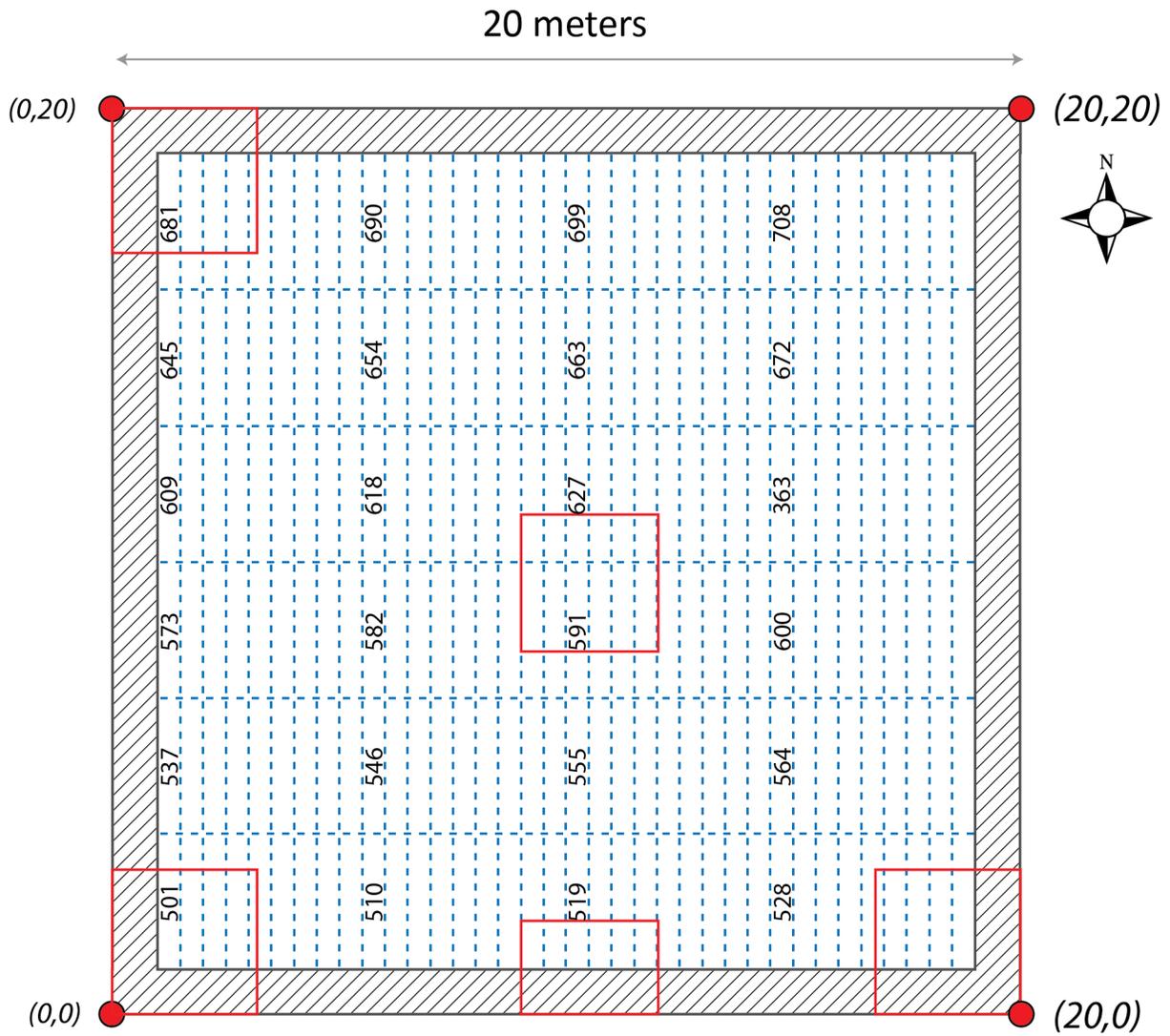
Figure 23. Map of clipCellNumbers for **subplotID = 39** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for litter sampling.

20 meters
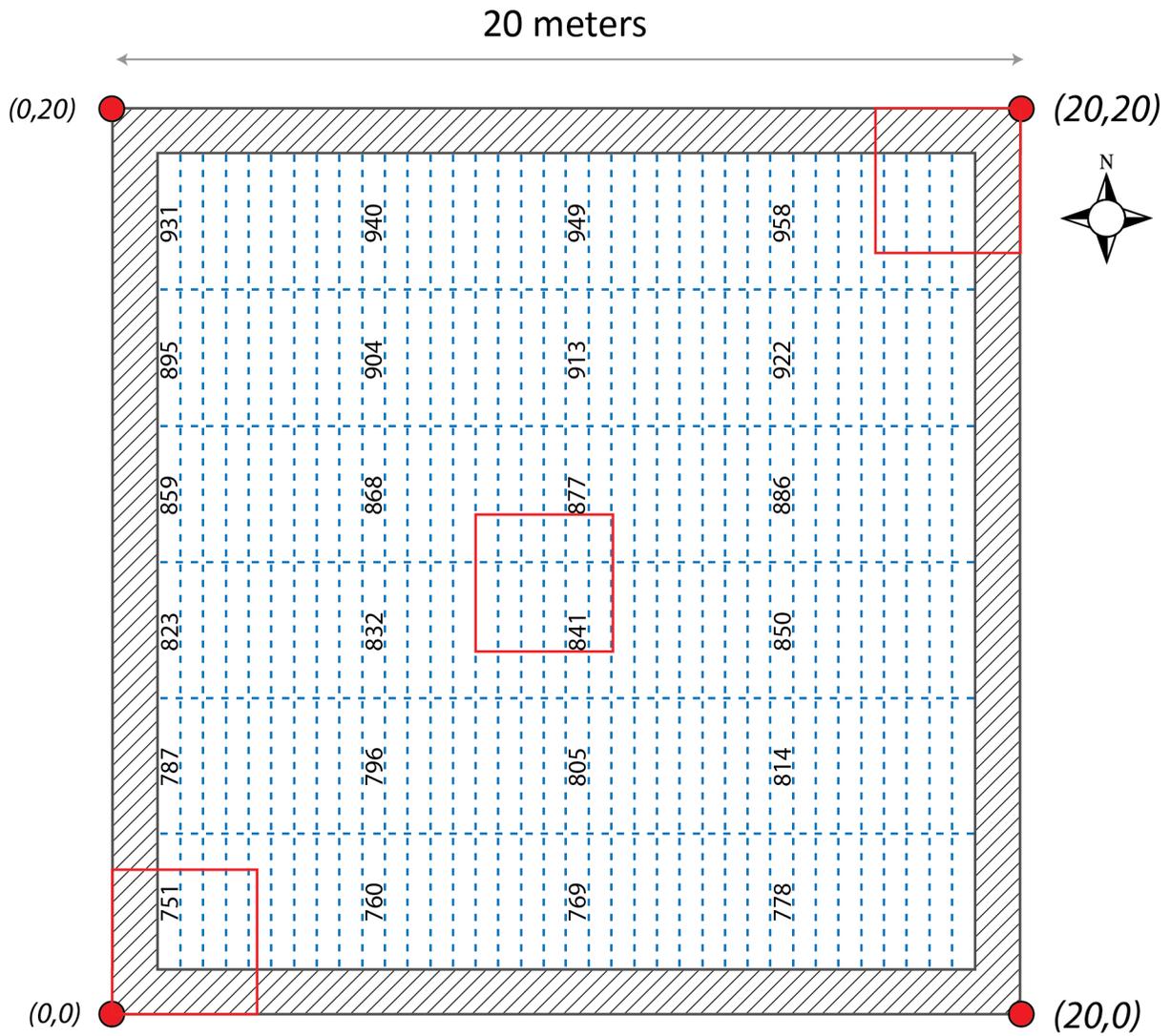
Figure 24. Map of clipCellNumbers for **subplotID = 41** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for litter sampling.

G.1 Coordinates for litter trap placement by clipCellNumber and subplotID

Table 21. List of clipCell coordinates by subplotID.

Coordinates correspond to the either 1) SW corner of the 0.5m x 3m ClipCell for ground trap placement, or 2) the centroid of the clip cell over which an elevated trap would be placed. Offsets indicate the distance in meters relative to the SW corner of the plot (subplotID = 31) or subplot. These are **not** the same coordinates used in the herbaceous clip harvest protocol. Print this Appendix separately for use with this protocol

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
1	1	251	501	751	1	1	1.25	2.5
2	2	252	502	752	1.5	1	1.75	2.5
3	3	253	503	753	2	1	2.25	2.5
4	4	254	504	754	2.5	1	2.75	2.5
5	5	255	505	755	3	1	3.25	2.5
6	6	256	506	756	3.5	1	3.75	2.5
7	7	257	507	757	4	1	4.25	2.5
8	8	258	508	758	4.5	1	4.75	2.5
9	9	259	509	759	5	1	5.25	2.5
10	10	260	510	760	5.5	1	5.75	2.5
11	11	261	511	761	6	1	6.25	2.5
12	12	262	512	762	6.5	1	6.75	2.5
13	13	263	513	763	7	1	7.25	2.5
14	14	264	514	764	7.5	1	7.75	2.5
15	15	265	515	765	8	1	8.25	2.5
16	16	266	516	766	8.5	1	8.75	2.5
17	17	267	517	767	9	1	9.25	2.5
18	18	268	518	768	9.5	1	9.75	2.5
19	19	269	519	769	10	1	10.25	2.5
20	20	270	520	770	10.5	1	10.75	2.5
21	21	271	521	771	11	1	11.25	2.5
22	22	272	522	772	11.5	1	11.75	2.5
23	23	273	523	773	12	1	12.25	2.5
24	24	274	524	774	12.5	1	12.75	2.5
25	25	275	525	775	13	1	13.25	2.5
26	26	276	526	776	13.5	1	13.75	2.5
27	27	277	527	777	14	1	14.25	2.5
28	28	278	528	778	14.5	1	14.75	2.5
29	29	279	529	779	15	1	15.25	2.5
30	30	280	530	780	15.5	1	15.75	2.5
31	31	281	531	781	16	1	16.25	2.5
32	32	282	532	782	16.5	1	16.75	2.5

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
33	33	283	533	783	17	1	17.25	2.5
34	34	284	534	784	17.5	1	17.75	2.5
35	35	285	535	785	18	1	18.25	2.5
36	36	286	536	786	18.5	1	18.75	2.5
37	37	287	537	787	1	4	1.25	5.5
38	38	288	538	788	1.5	4	1.75	5.5
39	39	289	539	789	2	4	2.25	5.5
40	40	290	540	790	2.5	4	2.75	5.5
41	41	291	541	791	3	4	3.25	5.5
42	42	292	542	792	3.5	4	3.75	5.5
43	43	293	543	793	4	4	4.25	5.5
44	44	294	544	794	4.5	4	4.75	5.5
45	45	295	545	795	5	4	5.25	5.5
46	46	296	546	796	5.5	4	5.75	5.5
47	47	297	547	797	6	4	6.25	5.5
48	48	298	548	798	6.5	4	6.75	5.5
49	49	299	549	799	7	4	7.25	5.5
50	50	300	550	800	7.5	4	7.75	5.5
51	51	301	551	801	8	4	8.25	5.5
52	52	302	552	802	8.5	4	8.75	5.5
53	53	303	553	803	9	4	9.25	5.5
54	54	304	554	804	9.5	4	9.75	5.5
55	55	305	555	805	10	4	10.25	5.5
56	56	306	556	806	10.5	4	10.75	5.5
57	57	307	557	807	11	4	11.25	5.5
58	58	308	558	808	11.5	4	11.75	5.5
59	59	309	559	809	12	4	12.25	5.5
60	60	310	560	810	12.5	4	12.75	5.5
61	61	311	561	811	13	4	13.25	5.5
62	62	312	562	812	13.5	4	13.75	5.5
63	63	313	563	813	14	4	14.25	5.5
64	64	314	564	814	14.5	4	14.75	5.5
65	65	315	565	815	15	4	15.25	5.5
66	66	316	566	816	15.5	4	15.75	5.5
67	67	317	567	817	16	4	16.25	5.5
68	68	318	568	818	16.5	4	16.75	5.5
69	69	319	569	819	17	4	17.25	5.5
70	70	320	570	820	17.5	4	17.75	5.5
71	71	321	571	821	18	4	18.25	5.5
72	72	322	572	822	18.5	4	18.75	5.5
73	73	323	573	823	1	7	1.25	8.5

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
74	74	324	574	824	1.5	7	1.75	8.5
75	75	325	575	825	2	7	2.25	8.5
76	76	326	576	826	2.5	7	2.75	8.5
77	77	327	577	827	3	7	3.25	8.5
78	78	328	578	828	3.5	7	3.75	8.5
79	79	329	579	829	4	7	4.25	8.5
80	80	330	580	830	4.5	7	4.75	8.5
81	81	331	581	831	5	7	5.25	8.5
82	82	332	582	832	5.5	7	5.75	8.5
83	83	333	583	833	6	7	6.25	8.5
84	84	334	584	834	6.5	7	6.75	8.5
85	85	335	585	835	7	7	7.25	8.5
86	86	336	586	836	7.5	7	7.75	8.5
87	87	337	587	837	8	7	8.25	8.5
88	88	338	588	838	8.5	7	8.75	8.5
89	89	339	589	839	9	7	9.25	8.5
90	90	340	590	840	9.5	7	9.75	8.5
91	91	341	591	841	10	7	10.25	8.5
92	92	342	592	842	10.5	7	10.75	8.5
93	93	343	593	843	11	7	11.25	8.5
94	94	344	594	844	11.5	7	11.75	8.5
95	95	345	595	845	12	7	12.25	8.5
96	96	346	596	846	12.5	7	12.75	8.5
97	97	347	597	847	13	7	13.25	8.5
98	98	348	598	848	13.5	7	13.75	8.5
99	99	349	599	849	14	7	14.25	8.5
100	100	350	600	850	14.5	7	14.75	8.5
101	101	351	601	851	15	7	15.25	8.5
102	102	352	602	852	15.5	7	15.75	8.5
103	103	353	603	853	16	7	16.25	8.5
104	104	354	604	854	16.5	7	16.75	8.5
105	105	355	605	855	17	7	17.25	8.5
106	106	356	606	856	17.5	7	17.75	8.5
107	107	357	607	857	18	7	18.25	8.5
108	108	358	608	858	18.5	7	18.75	8.5
109	109	359	609	859	1	10	1.25	11.5
110	110	360	610	860	1.5	10	1.75	11.5
111	111	361	611	861	2	10	2.25	11.5
112	112	362	612	862	2.5	10	2.75	11.5
113	113	363	613	863	3	10	3.25	11.5
114	114	364	614	864	3.5	10	3.75	11.5

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
115	115	365	615	865	4	10	4.25	11.5
116	116	366	616	866	4.5	10	4.75	11.5
117	117	367	617	867	5	10	5.25	11.5
118	118	368	618	868	5.5	10	5.75	11.5
119	119	369	619	869	6	10	6.25	11.5
120	120	370	620	870	6.5	10	6.75	11.5
121	121	371	621	871	7	10	7.25	11.5
122	122	372	622	872	7.5	10	7.75	11.5
123	123	373	623	873	8	10	8.25	11.5
124	124	374	624	874	8.5	10	8.75	11.5
125	125	375	625	875	9	10	9.25	11.5
126	126	376	626	876	9.5	10	9.75	11.5
127	127	377	627	877	10	10	10.25	11.5
128	128	378	628	878	10.5	10	10.75	11.5
129	129	379	629	879	11	10	11.25	11.5
130	130	380	630	880	11.5	10	11.75	11.5
131	131	381	631	881	12	10	12.25	11.5
132	132	382	632	882	12.5	10	12.75	11.5
133	133	383	633	883	13	10	13.25	11.5
134	134	384	634	884	13.5	10	13.75	11.5
135	135	385	635	885	14	10	14.25	11.5
136	136	386	636	886	14.5	10	14.75	11.5
137	137	387	637	887	15	10	15.25	11.5
138	138	388	638	888	15.5	10	15.75	11.5
139	139	389	639	889	16	10	16.25	11.5
140	140	390	640	890	16.5	10	16.75	11.5
141	141	391	641	891	17	10	17.25	11.5
142	142	392	642	892	17.5	10	17.75	11.5
143	143	393	643	893	18	10	18.25	11.5
144	144	394	644	894	18.5	10	18.75	11.5
145	145	395	645	895	1	13	1.25	14.5
146	146	396	646	896	1.5	13	1.75	14.5
147	147	397	647	897	2	13	2.25	14.5
148	148	398	648	898	2.5	13	2.75	14.5
149	149	399	649	899	3	13	3.25	14.5
150	150	400	650	900	3.5	13	3.75	14.5
151	151	401	651	901	4	13	4.25	14.5
152	152	402	652	902	4.5	13	4.75	14.5
153	153	403	653	903	5	13	5.25	14.5
154	154	404	654	904	5.5	13	5.75	14.5
155	155	405	655	905	6	13	6.25	14.5

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
156	156	406	656	906	6.5	13	6.75	14.5
157	157	407	657	907	7	13	7.25	14.5
158	158	408	658	908	7.5	13	7.75	14.5
159	159	409	659	909	8	13	8.25	14.5
160	160	410	660	910	8.5	13	8.75	14.5
161	161	411	661	911	9	13	9.25	14.5
162	162	412	662	912	9.5	13	9.75	14.5
163	163	413	663	913	10	13	10.25	14.5
164	164	414	664	914	10.5	13	10.75	14.5
165	165	415	665	915	11	13	11.25	14.5
166	166	416	666	916	11.5	13	11.75	14.5
167	167	417	667	917	12	13	12.25	14.5
168	168	418	668	918	12.5	13	12.75	14.5
169	169	419	669	919	13	13	13.25	14.5
170	170	420	670	920	13.5	13	13.75	14.5
171	171	421	671	921	14	13	14.25	14.5
172	172	422	672	922	14.5	13	14.75	14.5
173	173	423	673	923	15	13	15.25	14.5
174	174	424	674	924	15.5	13	15.75	14.5
175	175	425	675	925	16	13	16.25	14.5
176	176	426	676	926	16.5	13	16.75	14.5
177	177	427	677	927	17	13	17.25	14.5
178	178	428	678	928	17.5	13	17.75	14.5
179	179	429	679	929	18	13	18.25	14.5
180	180	430	680	930	18.5	13	18.75	14.5
181	181	431	681	931	1	16	1.25	17.5
182	182	432	682	932	1.5	16	1.75	17.5
183	183	433	683	933	2	16	2.25	17.5
184	184	434	684	934	2.5	16	2.75	17.5
185	185	435	685	935	3	16	3.25	17.5
186	186	436	686	936	3.5	16	3.75	17.5
187	187	437	687	937	4	16	4.25	17.5
188	188	438	688	938	4.5	16	4.75	17.5
189	189	439	689	939	5	16	5.25	17.5
190	190	440	690	940	5.5	16	5.75	17.5
191	191	441	691	941	6	16	6.25	17.5
192	192	442	692	942	6.5	16	6.75	17.5
193	193	443	693	943	7	16	7.25	17.5
194	194	444	694	944	7.5	16	7.75	17.5
195	195	445	695	945	8	16	8.25	17.5
196	196	446	696	946	8.5	16	8.75	17.5

clipCell Number subplotID = 31	clipCell Number subplotID = 21	clipCell Number subplotID = 23	clipCell Number subplotID = 39	clipCell Number subplotID = 41	Ground Trap easting offset	Ground Trap northing offset	Elevated Trap easting offset	Elevated Trap northing Offset
197	197	447	697	947	9	16	9.25	17.5
198	198	448	698	948	9.5	16	9.75	17.5
199	199	449	699	949	10	16	10.25	17.5
200	200	450	700	950	10.5	16	10.75	17.5
201	201	451	701	951	11	16	11.25	17.5
202	202	452	702	952	11.5	16	11.75	17.5
203	203	453	703	953	12	16	12.25	17.5
204	204	454	704	954	12.5	16	12.75	17.5
205	205	455	705	955	13	16	13.25	17.5
206	206	456	706	956	13.5	16	13.75	17.5
207	207	457	707	957	14	16	14.25	17.5
208	208	458	708	958	14.5	16	14.75	17.5
209	209	459	709	959	15	16	15.25	17.5
210	210	460	710	960	15.5	16	15.75	17.5
211	211	461	711	961	16	16	16.25	17.5
212	212	462	712	962	16.5	16	16.75	17.5
213	213	463	713	963	17	16	17.25	17.5
214	214	464	714	964	17.5	16	17.75	17.5
215	215	465	715	965	18	16	18.25	17.5
216	216	466	716	966	18.5	16	18.75	17.5

APPENDIX H   COLLECTING LITTERFALL FROM *TOXICODENDRON* SPECIES

This appendix deals with protocol-specific mitigation strategies for dealing with *Toxicodendron* in the course of litterfall and fine woody debris sampling. General strategies for preventing *Toxicodendron* exposure are described in detail in the *Toxicodendron* SOP (RD[12]).

H.1   Equipment and Materials

Table 22. Equipment and materials required for a team of two to minimize exposure to toxic oils from *Toxicodendron* spp. during litter collection

Item Description	Qty	Example Item	Purpose
Small paper bags, pre-weighed, labeled with bag weight	Variable	8# or lunch sack type	*Toxicodendron* biomass never handled directly again after it is placed in pre-weighed bag if not being processed for archive and chemistry analysis
Cotton gloves, single use	Box of 12	http://www.globalindustrial.com/p/safety/hands/cotton-canvas-gloves/anchor-4501v-8-oz-cotton-canvas-knit-wrist-1110	Prevent oil contact with skin.
Disposable PPE outer-wear	Case of 24	Coveralls; http://disposable-garments.com/shop/koolguard/koolguard-coveralls/	Prevent oil contact with skin, normal clothing.
Large, single-use plastic bags	Box	Trash bag or large Ziploc type bag	Transport used gloves and PPE and minimize toxic oil transfer.
Cleanser, urushiol-specific	1	Tecnu or equivalent; http://www.teclabsinc.com/products/poison-oak-ivy/tecnu	Clean equipment and surfaces after use.

H.2   Minimizing Exposure to Toxic Oil in the Field and Lab

Plot locations with *Toxicodendron spp.* present require a modified sampling strategy to collect and weigh litter dry mass. There are two possible approaches to collection, either of which is acceptable from a science perspective.

Option A: sort all litter material in the field.

Field processing litter requires extra time in the field but all functional groups from the trap can then be treated in a similar manner to *Toxicodendron*. That is, weighed and discarded without removing material from bags.

Option B: sort non-*Toxicodendron* material in the lab

Sort *Toxicodendon* from the trap, bulk the remainder in a cloth collection bag, sort in the lab with disposable cotton gloves (optionally on butcher paper) then decontaminate all surfaces with Urushiol-specific cleanser.

Label all sample bags with warning sticker.

The following are best-practice techniques for minimizing exposure to *urushiol* oils during litterfall collection of *Toxicodendron* species.

1. Prior to field work:
    - Count out bags for storing and drying *Toxicodendron* biomass **and** other functional groups (include enough for collection of leaves, fruit and stems in separate bags). Don't mix *Toxicodendron* biomass with any other biomass.
    - Pre-weigh (to nearest 0.01 g) and label each paper bag that will be used for storing and drying litter material from traps that include *Toxicodendron* biomass. Once the weight of each empty bag is included on the bag label, the biomass inside the bag will never have to be touched after it is initially placed in the bag unless collected during a chemistry bout.
2. To handle *Toxicodendron* biomass in the field:

    - Wear cotton gloves and dispose after single use. Toxic oils can pass through nitrile or latex gloves.
    - Bring a clean, new plastic bag to the field for storing and transporting contaminated gloves after use.
    - Wear a thin outer layer of disposable PPE over clothes and shoes.

3. After field work is complete, wash clothing and collection bags according to these guidelines or similar:
    - While handling and loading unwashed clothing exposed to toxic oils, wear gloves or use a clean cloth to prevent direct contact between your skin and the clothing.
    - Wash with ordinary laundry detergent at the highest recommended water temperature.
    - Do not overload the machine; the clothes must be allowed to agitate freely.

4. To process *Toxicodendron* biomass for dryMass measurement in the laboratory:

    - Wear cotton gloves while handling *Toxicodendron* or any litter material that may have come in contact with *Toxicodendron* litter in traps, including sorting of non *Toxicodendron* material.
    - Disinfect all tools and lab surfaces used in the sorting process with Tecnu. Discard gloves.
    - Minimize potential spread of toxic oil by putting *Toxicodendron* biomass bags into the same drying oven every time.
    - When drying is complete, clean drying oven shelves used for drying *Toxicodendron* biomass bags with hot water and Tecnu. Wear appropriate PPE when cleaning.

- Record weight dried biomass, minus weight of the bag, to nearest 0.01 g. Dried *Toxicodendron* biomass should never leave the bag.
- After weighing, if the bout is not being processed for archive and chemistry analyses, dispose of all biomass bags from traps that contained *Toxicodendron*.

5. To process *Toxicodendron* biomass or leaf/needle for Bioarchive and Leaf Chemistry
   - If sample contains *Toxicodendron spp*, or was collected from a trap that also contained *Toxicodendron*, no grinding s take place. However, subsampling for chemical analyses and archive will still occur.
   - Conduct all subsampling activities in a clean fume hood. Use caution when handling the sample so as not to expose yourself or others to leaves containing toxic oils.
   - Wear single-use cotton gloves as described in RD[12] and follow the guidelines in RD[12] to clean any equipment, clothing, or skin that comes in contact with foliage.
   - For the leaves sample, combine *Toxicodendron* and non-*Toxicodendron* material
   - Homogenize the sample prior to manual subsampling by crushing/shaking the contents of the brown paper sample bag(s). It may be helpful to transfer sample to a larger-size paper bag first if it is held in a small paper bag.
     - If the sample is very large (> 20 g), haphazardly subsample ~ 20 g first, then use this for further subsampling. The rest may be discarded.
   - Split the homogenized foliar material into three subsamples. Try to ensure that the splits are fairly representative but with minimal handling of the foliage.
   - Sample mass < 10 g: follow guidelines in section F.2, to apportion material for the different subsample types. Use forceps to avoid having to touch the material where possible.
   - Do not grind the archive subsample.
   - Label all samples with the warning sticker

APPENDIX I   TROUBLESHOOTING

Sampling Challenge	Proposed solution
Plot is seasonally inundated.	*Deployment* – Initial deployment of traps cannot occur while plots are inundated. If a site experiences areas of seasonal flooding, deployment may occur in all dry plots prior to deployment in inundated plots. Trap deployment is not all or nothing though it is preferable, once all traps are deployed, to sample all plots at the same time. Production is reported as per/year mass/area so there is wiggle room on collection dates. Tracking is easier if everything is on the same schedule. Though PVC is not buoyant, a sealed frame of an elevated trap could be lifted and moved by water in the plot. If it appears likely that a plot may occasionally experience periods of inundation act preemptively by weighting the elevated traps (consider bricks or large rocks) or drilling a couple small holes in the top of the frame to allow air to escape, minimizing float potential. *Collection* - Though at the time of deployment of ground traps the clip strip is cleared of all woody material, inundation will move litter laterally across the landscape, it is likely in these plots; total annual production of fine woody debris will be overestimated since material > 1yr will float into the trap area. It is not practical to attempt to distinguish new litter from old, so all qualifying litter present in the trap area should be collected; record trapCondition = PF to indicate that the trap location was previously flooded. This way a user can search records and identify those at which estimates of annual production are affected by flooding.
Atypical structures in litter samples slow down sorting time	At sites with high diversity of species, it may be difficult to identify structures that are only occasionally encountered in litter samples. One solution may be to create a reference collection to make sorting more efficient. Collections may include: pollen cones, seed cones, seeds, or flower parts. Creation of a litter reference collection is at the discretion of domain staff and is not a requirement imposed by Science. For distinguishing structures from flowers vs. fruits, one approach may be to use phenological cues to sort unattached flower/seed structures into the appropriate functional group.
Quarantine in effect at site	Discontinue sampling, document quarantine issues via a problem ticket. Coordinate with Domain Manager, HQ Permitting, and regulatory agency to determine how sampling should proceed.
Elevated traps overtopped by plant growth	Manually remove plants growing up and on elevated litter traps, as well as plants growing beneath and immediately around the trap that are likely to grow up trap legs. Ground tarps may be used to minimize growth of particularly aggressive plants beneath elevated traps. Additionally, adding weights (or a rock) to the screen may help traps remain upright if vegetation does threaten to disturb the trap between maintenance bouts.

Sampling Challenge	Proposed solution
Unexpected material collected in litter trap	All qualifying plant material present in the elevated traps should be collected. Galls, for example, shouldn't be removed from litter but should be sorted with the functional group from which the source tissue originates; a gall on a twig should be sorted with twigs and branches, a gall on a leaf should be sorted with leaves.

Plant material that may not originate from overhanging vegetation but does qualify according to the guidelines provided in this protocol should be collected. For example nest material including grass, twigs, herbaceous plants, and moss, collected and transported by birds or small mammals still represents material produced within a given year, presumably from the plot or nearby areas. Nest material likely contains many different tissue types some of which may not be identifiable, it is therefore acceptable to sort all nest material in the 'other' category.

However, material growing up through the mesh from below an elevated trap should be excluded and trimmed back as part of regular trap maintenance before reaching the height of the trap.

Seeds from fruits consumed elsewhere then deposited by birds represent plant material produced in the current year that would otherwise have landed in the 0.5m2 patch of ground, these seeds should be collected and sorted in the 'seeds' category.

 An exception is made for sap. Do not place pieces of sap or any other plant exudate, in the drying ovens, under heat, these materials will be lost to melting or pose safety concerns due to natural flammability. Exudates are not explicitly accounted for in net primary productivity calculations. Small amounts of sap bound in woody seed cones does not generally pose a fire hazard. However, if the volume of exudate is great enough to saturate a paper bag or there is any risk of sap dripping and collecting on heating elements in the oven, exclude the material from the sample and record # of female cones were discarded.

Non-plant material, including invertebrates and animal by-products, found in a field trap should be removed when collected and discarded according. Dead vertebrates found in the trap should be collected and processed according to the guidelines in the State Collection Permit. |
| Snow in elevated trap | If snow is present in an elevated trap at collection, collect all snow and litter; do not attempt to separate snow from litter while in the field. If snow is mounded such that disturbing the pile would result in snow falling outside of trap, or the volume of snow is > the volume of your collection bag, do not collect.

In the case where there is too much snow to collect, skip the trap, do not record any data. Use field notes or draft records to manage internal accounting. |

	Melt snow at DSF and air dry before sorting.
Trap incorrectly constructed or incorrectly marked	If the trap dimensions are not correct, if the area is < 15% different from intended trapSize, correct the trap and continue sampling the selected location. If the trap dimensions and the area is >=15% different from the intended trapSize, drop the sampling location, move to the next random clipcell and construct a trap with the correct dimensions and submit a data update request to enter a non-standard trapSize for old data.

APPENDIX J   ALTERNATIVE TRAP MATERIALS

Based on site conditions, it may be necessary to modify materials used in construction of the elevated litter trap.

Here are suggestions employed by some NEON domains to address specific issues:

- **Destruction by bears or cattle.** Trap frame material. The design specifies PVC but at some sites, this material may be attractive to bears resulting in widespread damage to traps. Wood traps and galvanized conduit traps constructed at the domain office are an approved alternative (**Figure 25** and **Figure 26**, and **Figure 27**). Additionally, larger anchors such as t-posts may be employed to discourage animals from attempting to move and destroy elevated litter traps
  - Conduit traps are deployed at YELL tower plots and are constructed using the following materials:
    - 1-3/4" x 3-1/2" x 5' Green Steel Fence T-Posts
    - 3/4" Electric Metallic Tube (EMT) Conduit
    - 3/4" 3-way thru canopy roof fittings
- Metals with potential to oxidize and leach into the soil may only be used in plots not scheduled for soil biogeochemistry sampling.

Figure 25. PVC elevated trap destroyed by bears at SCBI.

Figure 26. Wood elevated trap frame at Konza.

Figure 27. Conduit trap from D17- San Joaquin

- **Destruction by rodents.** Application of a non-toxic capsaicin rodent deterrent spray on trap surfaces may render the trap material un-palatable without causing undue harm to surrounding vegetation or wildlife. Spray must be re-applied to maintain efficiency. If zip ties are targeted, mesh may be secured to trap with the aluminum wire used to attach numbered tags to shrubs and saplings.
- **Removal of material by wind.** Traps may be weighted by placing baseball-size rocks in the elevated trap to prevent wind from disturbing the mesh and forcing collected material out of the trap. Additionally, using a larger piece of mesh than the 4ft x 4ft piece provided in the kits to create more sag, a deeper bowl (i.e., >20 cm specified in SOP A) that may trap material more effectively in windy conditions.

Elements that may **not** be modified:

- Trap shape, elevated traps must be square

- Trap size, elevated traps must be 0.5m^2 (70 cm x 70 cm)

Use of non-oxidizing materials in plots scheduled for soil biogeochemistry sampling, if metal is used for any portion of the trap, it must resistant to rust (aluminum, stainless steel)