TOS PROTOCOL AND PROCEDURE: LITTERFALL AND FINE WOODY DEBRIS

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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

TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

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 will provide essential data for understanding vegetative C fluxes over time. Litterfall and fine woody debris production will be estimated within Tower plots on an annual basis. Sampling point selection within a plot or subplot will be random; sampling points will be 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 will be targeted rather than random across the plot. This protocol will not be implemented at sites with overstory vegetation < 2 meters tall.

This design 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. However, sorting freshly collected litter is preferable because dry litter is easily fragmented and identifying small litter fragments to functional group will introduce uncertainty in sorting accuracy.

Elevated litter trap size has been selected to be consistent with existing standards and are the same dimensions (70 cm x 70 cm x 80 cm) as traps used by Smithsonian Tropical Research Institute Center for Tropical Forest Studies(CTFS. To minimize the number of clip strips dedicated to fine woody debris sampling, which are therefore unavailable for herbaceous biomass sampling, ground traps will have the same dimensions as a single clip strip cell, 3 m x 0.5 m. If it is apparent that the volume of litterfall biomass collected from elevated and ground litter traps is too great to efficiently dry and process given limitations on drying oven space in the NEON laboratory, trap size or number may be reduced by Science Operations based on sample optimization analysis.

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: Laboratory Processing for Dry Mass Measurement:** Covers laboratory processing including drying and weighing of samples.
- **SOP E: Data Entry and Verification:** Provides guidance for manual data transcription from paper data sheets to the Access database.
- **SOP F: Processing Litter Samples for Biogeochemistry:** Describes the steps for sub-sampling and grinding dried leaf and needle material.

- **SOP G: Sample Shipment for Biogeochemistry:** Provides science rationale for timelines and restrictions on sample handling and shipping to external facilities.

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.050005	Field Operations Job Instruction Training Plan
AD[05]	NEON.DOC.000914	TOS Science Design for Plant Biomass, Productivity, and Leaf Area Index
AD[06]	NEON.DOC.014051	Field Audit Plan
AD[07]	NEON.DOC.000824	Data and Data Product Quality Assurance and Control Plan

2.2 Reference Documents

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

RD[01]	NEON.DOC.000008	NEON Acronym List
RD[02]	NEON.DOC.000243	NEON Glossary of Terms
RD[03]	NEON.DOC.005003	NEON Scientific 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.014037	TOS Protocol and Procedure: Measurement of Herbaceous Biomass
RD[07]	NEON.DOC.001025	TOS Protocol and Procedure: Plot Establishment
RD[08]	NEON.DOC.001711	TOS Protocol and Procedure: Coarse Woody Debris
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

2.3 Acronyms

Acronym	Definition
ANPP	Aboveground Net Primary Productivity
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). Woody pieces with diameter ≥ 2 cm are considered coarse woody debris, and will be sampled according to the NEON Field and Lab Protocol for Coarse Woody Debris (RD[08]).

3 METHOD

To measure litterfall and fine woody debris, NEON will employ two types of sampling units: 1) square, elevated, mesh litter traps; and 2) rectangular, ground "traps" (Figure 3, SOP B). Elevated litter traps are designed to be large enough that the average size of abundant foliage and fine woody debris elements are easily intercepted by the trap. Ground traps are intended to intercept particularly large foliage elements that will not fit in elevated traps (e.g. palm fronds), and fine woody debris pieces that are too long to be sampled in elevated traps including small diameter branches.

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 problem reporting system to resolve any field issues associated with implementing this protocol.

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

The procedures described in this protocol will be audited according to the Field Audit Plan (AD[06]). Additional quality assurance will be performed on data collected via these procedures according to the NEON Data and Data Product Quality Assurance and Control Plan (AD[07]).

3.1 Sampling Methods

For both elevated and ground traps, only the portion of material that meets both the length and diameter criteria will be sampled (Muller-Landau and Wright 2010). Litter sampled from elevated traps will be sorted into functional groups following collection, using the groupings outlined in Table 1. Note that these functional groups differ from those used in NEON's herbaceous clip harvest protocol (RD[06]); litter material larger than described in Table 1 will be collected according to that protocol.

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. cones, bark, etc)	< 50 cm length	> 50 cm length
Seeds	All	N/A
Flowers	All	N/A
Other (lichen, mosses, unidentifiable material, etc.)	All	N/A

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

Leaf and needle litter from elevated traps from a single sampling bout will be shipped to external laboratories to be analyzed for C, N, δ13C and, δ15N once every five years.

3.2 Laboratory processing

Following collection and sorting in the field, litter will be transported back to the laboratory and dried at 65˚C until water weight has been removed, to within the allowed variance indicated in SOP D (minimum 48 hrs). The woody portion of litter will be cut to fit in the drying oven then dried at a higher temperature than litterfall, 105 °C to release bound water (Williamson and Wiemann 2010).

3.3 Equipment

Design of PVC elevated litter traps is adopted from the CTFS design. Non-oxidizable metal rods (e.g. aluminum, galvanized steel, or equivalent) will be used to hold elevated litter traps in place. The corners of ground traps will be marked with non-oxidizable metal or plastic stakes to facilitate precise re-measurement of the selected sampling area.

3.4 Spatial Distribution of Sampling

Consistent with existing protocols, NEON will establish 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. The selected subplots will be same ones used for all other plant productivity sampling in Tower plots.

Only plots with woody vegetation > 2 m tall, will be selected for litter sampling using this protocol. Vegetation surveys conducted during site characterization will inform plot selection. Initially, all tower plots will automatically be considered for litter sampling and then accepted according to the following criteria:

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

Plots that do not meet these criteria are not utilized for litter sampling.

3.5 Elevated traps

An elevated mesh litterfall trap (70.7 cm x 70.7 cm x 80 cm; 0.5 m^2, 0.8 m tall) will be placed at a random location within each accepted plot/subplot, with trap locations selected from the herbaceous clip harvest list. Once set, traps will remain in the same location within the plot for sampling in subsequent years unless traps are removed for optimization. These traps will reliably sample shed leaves, needles, reproductive parts, and fine woody debris with butt-end diameter < 2 cm *and* length < 50 cm. Traps will be sampled according to the guidelines outlined in section 9 of this document. Deciduous forests will be sampled once in the spring then once every two weeks during leaf senescence. Evergreen systems including coniferous, xeric and tropical forests will be sampled year-round; the ideal sampling interval is every 4 weeks but may be extended to 8 weeks if dictated by logistical constraints. Sites with both deciduous and evergreen vegetation will be 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 will target litter trap placement to randomly selected areas of the plot with woody cover, and then use remote sensing imagery from NEON's Airborne Observation Platform (AOP) to estimate woody vegetation percent cover of the plot to scale litter production from the trap to the plot level. Scaling of this data will occur as part of the preparation of data products and is not expressly part of this protocol.

3.6 Ground traps

Ground traps for collecting large leaves, fronds, and fine woody debris with butt-end diameter < 2 cm and length > 50 cm, will be 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 will be one randomly selected 0.5 m x 3 m herbaceous clip harvest grid cell within the same plot or subplot as the elevated trap (Figure 2, SOP B). Ground traps are cleared of all relevant litter 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 will be sampled; these sample locations will not move from year to year and will be excluded from consideration as locations for herbaceous clip harvest.

4 SAMPLING SCHEDULE

4.1 Sampling Frequency and Timing

The primary objective is to generate annual or per growing season estimates of litterfall and fine woody debris production within the dominant vegetation type (i.e. within Tower plots). Estimates of deciduous litterfall will be calculated on a per annum basis, with all of the litter produced in a given year contributing toward the yearly estimate. Evergreen litterfall estimates within a given calendar year do not necessarily reflect annual production due to the multi-year and somewhat variable lifespan of needles; however, the long-term average (n = at least 3 years) will be used to estimate per annum needle production.

Material left uncollected in the field for longer than the specified sampling interval may be subject to granivory by small mammals, herbivory by insects, or increased decomposition and resulting loss of mass. In deciduous forests, elevated traps must be checked at least 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 sufficient resources exist to support additional sampling; this is left to the discretion of the Domain manager and will not be dictated by Science Operations.

4.1.1 Elevated traps

In Tower airsheds dominated by deciduous vegetation with pronounced annual senescence, elevated litter traps will be sampled in the spring to account for winter production of fine woody debris, followed by biweekly sampling during the period of autumn senescence (Bernier et al. 2008). In systems dominated by plants that bear multi-year leaves or needles (e.g. D17 San Joaquin and D04 Guanica), elevated traps will be sampled throughout the year. Mixed forests, forests with both evergreen and deciduous species present, will be sampled according to a hybrid approach; sampling should occur once a month with increased, bi-weekly sampling during senescence. If the hybrid approach is selected,

monthly sampling may be extended to the maximum 8-week sampling interval as needed to account for additional sampling bouts during senescence.

Litterfall in coniferous forests (e.g. D10 Rocky Mountain Park and D16 Wind River) or in xeric shrub systems (e.g. D14 Santa Rita and Jornada LTER) may be sampled with less frequency than deciduous broadleaf forests, but since there is no clear 'litterfall season' sampling will occur year round. NEON will sample litterfall in arid desert systems on a monthly basis (Table 2).

Once a month sampling is considered ideal to ensure data quality; however, sampling frequency at coniferous, xeric, tropical or mixed forest sites may be reduced to once every 8-weeks if dictated by logistical constraints.

Table 2. Sample timing and frequency by vegetation type

Climate / Ecosystem	When to sample elevated traps
Temperate Deciduous	• Once in the spring, approximately same time each 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	• Once a month* • Every two weeks during leaf senescence period

* A 4 week sampling interval is ideal for purposes of data quality but may be decreased to once every 8 weeks if dictated by logistical constraints.

4.1.2 Ground traps

Ground traps will be sampled once annually in Tower plots.

4.2 Criteria for Determining Onset and Cessation of Sampling

Elevated trap sampling will vary depending on the vegetation present at a site (Table 2). Ground litter trap sampling will occur once a year and should occur within ± 2 weeks of the date on which sampling occurred the previous calendar year. Initiation of 2 week sampling intervals during leaf senescence may be determined by checking an elevated trap from a plot near the Tower; once litter material from senesced falling leaves begins to accumulate in the trap, begin fall sampling. Data do not need to be collected on the 'indicator trap' and the one selected for monitoring does not need to be recorded. The only stipulation is that the selected trap is surrounded by deciduous trees.

4.3 Timing for Laboratory Processing and Analysis

Samples should be sorted and placed in the drying oven as soon as logistically feasible upon return to the domain lab to minimize loss of mass.

In dry environments, since samples are oven dry, they may be placed in temporary storage prior to weighing; in humid environments however, there is a tendency for dried samples to reabsorb water so samples should be weighed soon after removal from the drying oven.For samples from collection events selected for chemical analysis and bioarchive there are no scientific limits on the time oven-dried samples may be placed in temporary storage prior to grinding, and subsampling for chemical analysis and bioarchive, however, samples stored prior to grinding must be dried an additional 24 hrs in the drying oven before grinding. .

4.3.1 Processing Samples for Biogeochemistry

Dried samples of leaf and needle material from elevated traps collected during a single collection bout will be processed and sent to an external lab for biogeochemistry isotope analysis and bioarchive once every five years. These samples will be shipped according to the process outlined in SOP G.

4.4 Sampling Timing Contingencies

When unexpected field conditions require deviations from this protocol, the following field implementation guidance must be followed to ensure quality standards are met:

Table 3. Contingent decisions

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 (okay), or sort and oven-dry as per protocol (best), 2. Resume collection of litter trap ASAP with new labeled bags 3. Combine dried biomass per functional group for weighing when all biomass is dry.	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 delay occurs between litter collection, resume harvest of next trap ASAP	Some litter mass may be lost from traps, increasing uncertainty in biomass and ANPP estimates.
	If all traps are not collected in a single bout, prioritize collection of litter from missed traps at the subsequent bout	

Within a given year or growing season, Metcalfe et al. (2008) point out that litterfall collection efforts often have high levels of uncertainty and require greater sample size to accurately estimate annual production than other biomass pools. Additional traps may be installed at additional random (clip strip) locations per plot should variance of the litterfall estimate be greater than ± 10% of the estimated mean based on analysis, conducted by Science Operations, of data from initial collection events (see AD[05] for details), and if technician labor is available.

If it is apparent that the volume of biomass collected from elevated and ground litter traps is too great to efficiently dry and process given limited drying oven space in the NEON domain laboratories, trap size or number may be reduced if justified based on sample optimization analysis conducted by Science Operations.

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. Technicians 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 attend Hand and Power Tool Safety Training and Machine Shop Safety. Personnel shall be trained in the safe use, maintenance and cleaning of the Wiley® Mill or equivalent.

6 PERSONNEL AND EQUIPMENT

6.1 Equipment

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

Table 4. Equipment list – Initial trap deployment

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
Durable Items					
0343220000	R	Aluminum stake	Secure trap	4 per trap	N
MX104361	R	Chaining pins or other suitable anchor	Anchor measuring tapes	2	N
	S	Coin	Randomize selection of patches at sites with targeted selection	1	N
MX100320	S	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	N
EG07670000	R	Elevated litter trap assembly	Collect litter sample	40-50	N
MX103218	R	Foliage filter	Allow laser rangefinder use in dense vegetation	2	N

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
MX100322	R	Laser Rangefinder, ½ foot accuracy	Locate X, Y coordinates of within-plot trap location	1	N
MX104742	S	Laser Rangefinder, 1 yard accuracy	Measure distances. May be used, in conjunction with handheld compass, as an alternative to TruPulse	1	N
MX100722	R	Measuring tape, minimum 30 m	Locate clip-harvest strips within plots/subplots.Plot slope < 10 deg; grassland, savannah	1	N
MX103491	R	PVC pipe cutter	Cut PVC to length	1	N
	R	Torpedo bubble level	Check the angle of the elevated trap	1	N
MX103238	S	White reflector or reflective tape	Reflective target for laser rangefinder; aids in measuring distance to target accurately	1	N
Consumable items					
MX104908	R	CR123A battery	Spare battery for laser rangefinder	2	N
	R	PVC pipe glue	Permanently attach PVC from the elevated trap kits	1 jar	N
MX105416 (Blue) MX105397 (orange)	R	Survey marking flag, PVC or fiberglass stake	Delineate sampling area	4 per trap	N

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
Resources					
RD[05]	R	Datasheets for Litterfall and Fine Woody Debris	Record required data and metadata	Variable	N
	R	Per plot or subplot Clip Lists	Identify random clip-strip locations		N
	S	Random number list	Randomize selection of patches at sites with targeted selection	1	N

[1] All permanent marker material and color selection is contingent on approval by the NEON site host or local land manager
[2] 1 meter is ideal but may be adjusted as needed to suit site conditions
R/S=Required/Suggested

Table 5. Equipment list – Field sampling elevated and ground litter traps

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
Durable Items					
MX103524	R	Nylon rope	Delineate ground trap	1, 8 m	N
0343220000	S	Aluminum stake	Replace stakes on damaged ground traps	4	N

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
	R	Cloth bags, two colors, numbered and marked	Carry fresh, potentially wet, litter samples	2 per trap pair	N
EG07670000	R	Elevated litter trap assembly	Replace damaged traps	2	N
MX103218	R	Foliage filter	Allow laser rangefinder use in dense vegetation	2	N
MX109491	R	Handheld caliper, 0.1 cm precision	Measure branch diameters	1	N
MX100322	R	Laser Rangefinder, ½ foot accuracy	Locate X, Y coordinates of trap if thick brush prevents visual trap location in Thick brush	1	N
MX100497	S	Measuring stick, 1 m	Measure and identify/discard litter > 50 cm	1	N
	S	Pruning lopper, heavy duty	Cut branches up to 2 cm diameter	1	N
	R	Torpedo bubble level	Check the angle of the elevated trap	1	N
MX109425 (wirecutter)	S	Screen patch kit (pieces of screen, wire, wirecutters)	Repair minor holes in screen material	1	N
Consumable items					
MX104908	S	CR123A battery	Spare battery for laser rangefinder	2	N

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
MX104502	R	General Purpose Tags	Label collection bags	2 per trap pair	N
Resources					
RD[05]	R	Datasheets for Litterfall and Fine Woody Debris	Record required data and metadata	Variable	N

† May also mark 50cm on plot frame with permanent marker.
R/S=Required/Suggested

Table 6. Equipment list – Laboratory processing and analysis

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
Durable Items					
MX103237	R	Hy back pan	Receive sub-samples generated by splitter	2 per splitter	N
MX103235	R	Sample microsplitter, small capacity	Subsample from small volumes of ground sample. Relatively little litter mass per litterCode per trap	1	N
MX107196	R	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	N
Consumable items					
MX105089	R	Paper bag, #8	Contain litter, sorted to functional group	50	N
MX101278	R	Scintillation vials with caps, 20 mL	Contain ground split samples for shipment to archive or chemical analysis	As needed	N
MX105583 (15 gal)	S	Trash bag	Contain oven-dried samples before they are weighed	Box of 100	N
Resources					
RD[05]	R	Datasheet Lab Drying QC	Record data	As needed	N

Item No.	R/S	Description	Purpose	Quantity*	Special Handling
RD[05]	R	Datasheet Lab Weighing	Record data	As needed	N

R/S=Required/Suggested

6.2 Training Requirements

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

Technicians must be proficient in the use of handheld GPS units in order to successfully navigate to plots for sampling.

6.3 Specialized Skills

The lead plant technician must possess the demonstrated ability to identify collected plant structures to functional group via visual inspection. Preferably, the technicians sorting litter are the same technicians who harvested the litter in the field.

6.4 Estimated Time

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

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

Lab processing time will depend heavily on the volume of material collected and number of functional groups present in a given collection. Sorting material prior to drying will likely take 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 function group per trap.

7 STANDARD OPERATING PROCEDURES

SOP A Preparing for Sampling

1. Print clip strip lists for the plots that will be visited
   - Litterfall sampling locations will be selected from the plot-specific randomized lists created for herbaceous clip harvest locations (RD[06]). These lists are therefore essential for the completion of the trap deployment procedure (SOP B), 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 will be utilized in the field regardless of selected trap placement strategy.
2. Gather all field equipment
3. Number cloth collection bags and mark with either an ‘E’ for elevated trap or a ‘G’ for ground trap.
4. Prepare GPS:
   a. Charge batteries
   b. Load plot locations
5. Prepare laser rangefinder (if using)
   a. Check battery and charge
   b. Clean lenses with lens cloth or lens tissue (if necessary)
   c. Check/set correct declination. See RD[11] for details.
   d. Calibrate tilt sensor.; see RD[11] for details.
   e. Calibrate internal compass.
6. Prepare compass (if using)
   a. 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/
7. Generate randomized number lists for sites with targeted selection
8. Print datasheets (RD[05]) on all-weather paper

SOP B Initial Deployment of Traps

B.1 Selecting litter trap location strategy

Litter traps will be deployed in pairs, one elevated and one ground trap per pair. There will be one set of paired traps 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.. Because litter sampling will primarily occur in forested sites where plot size is typically 1600 m^2 or more, most plots will have at least two pairs of traps. Trap placement will utilize the clip cell grid developed for the herbaceous clip harvest protocol(RD[06]). Refer to the TOS Protocol and Procedure: Plot Establishment (RD[07]) for details on handling measuring tapes and plot delineation tips.

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 is present throughout < 50% of the plot area.
- **Random selection** is employed in forested sites with relatively continuous canopy

Refer to Appendix D for recommended strategy; these recommendations are based on a combination of NLCD vegetation classification and satellite imagery. If the selected strategy seems inappropriate (based on the criteria listed above) for a particular site given the conditions on the ground, use NEON's problem reporting system to iterate with Science Operations about the trap placement strategy.

B.2 Locating targeted elevated trap location

1. Navigate to the desired plot and, if sampling in an 40 m x 40 m plot, the randomly selected subplot.
2. Assess location of patches of qualifying vegetation within the plot or subplot (depending on plot size)
3. Give each patch a numeric value. Assign values sequentially, left to right, bottom to top, beginning in the SW corner (Figure 1)
4. Use either a random number list or a series of coin flips to randomly select a patch to target for litterfall and fine woody debris sampling.
5. Once a patch is selected, select a location under the canopy, central to the patch to place an elevated litter trap.
6. Use the range finder to measure the distance to plot/subplot edges.
7. Determine where the nearest clip strip centroid is located.
   a. From the selected location, measure distance to the nearest N-S plot boundary to determine the x-coordinate of this point
   b. Measure the distance to the nearest E-W plot boundary to determine the y-coordinate

c. Use clip cell map to identify the clip cell located closest to the selected point
d. Navigate to the centroid of that cell.
    1) clipStrip x-coordinate + 5 cm, clipStrip y-coordinate + 1 m

8. If practical, center trap over that point, this will minimize the number of clips that will be removed from consideration for herbaceous clip harvest.

- In the example provided in Figure 1, the coordinates associated with nearest clip strip centroid from the center of patch 4 are: x = 3.7, y = 11.5.
- Not centering the trap over a centroid is acceptable but not ideal as there will be more cells excluded from consideration for herbaceous clip harvest.

9. Place a pin flag at the selected trap location.

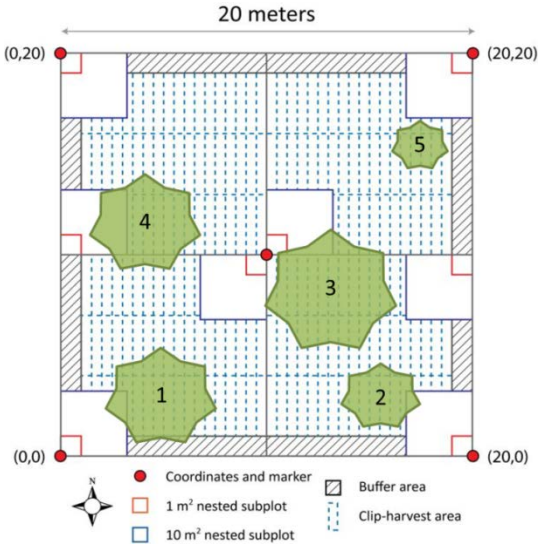

Figure 1. Example of numbering system for qualifying patches of vegetation within a plot

B.3 Locating random elevated trap location

Use the plot- or subplot-specific Clip List ([plotID]_[subplotID]_clipList.csv) 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 clipCell from the randomized list:

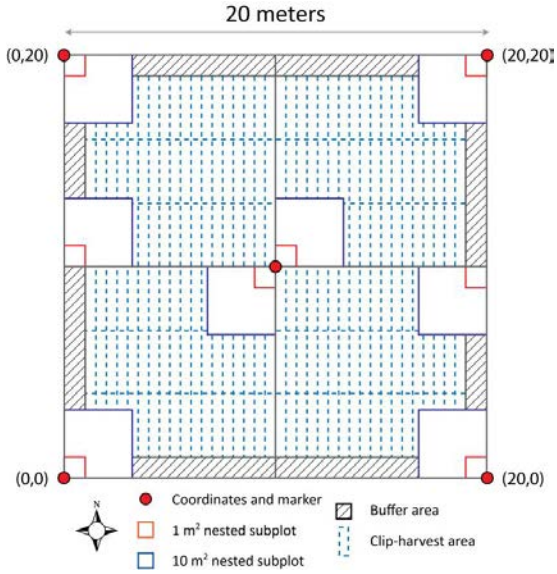

Figure 2. A 20 m x 20 m NEON plot showing the locations of 0.5m x 3m clip-harvest "cells" (dashed blue lines). Larger plots will have different nested subplots, but the coordinate numbering system for the 20 m subplot within these plots will follow the same conventions as shown above. 40m x 40m plot schematic available in Appendix F

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 2), and stretch the tape taut.
b. Place a pin flag at the desired relative X-coordinate.
c. Standing directly over the pin flag that was just placed 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]).
   - Note: if laser rangefinder is not available, the same routine described here may be completed using a handheld compass to verify azimuth and a laser rangefinder or additional tape measure for distance.
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 2) *Note: in 40 m x 40 m plots, subplot centroids are 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.
   - Make sure the azimuth is 0˚ (True North) when shooting the laser rangefinder to find the Y-coordinate (see RD[11]).
   - Note: if laser rangefinder is not available, the same routine described here may be completed using a handheld compass to verify azimuth and a laser rangefinder or additional tape measure for distance.
d. Place a pin flag at the clip-strip (X,Y) location.

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 strip for an elevated litter trap:
   - Accept the strip if no obstacles are present that prevent trap placement and anchoring (e.g. large shallow rock covering a majority of the clip cell, large boulders or impermeable vegetation)
   - If the strip is not acceptable for placement of an elevated litter trap, move to the next strip on the list but do NOT record the strip status as rejected for herbaceous biomass sampling.
3. Navigate to center of the cell (1 m north, 0.05 m east), place a pin flag. Elevated traps will be centered over this point.
   - If the trap cannot be anchored over the selected point, the trap may be shifted up to 1 meter North or South.

- Record a '1' in the **Status** column of Clip List sheet for clip-strip selected, Record the litter trap deployment date in the **Date** field, add a note that cell was used for litter collection.

B.4 Locating ground trap clip strip

Ground traps will be placed across one entire clipCell and may not be placed such more than one cell is occupied by a ground trap.

- **Targeted selection** – repeat the process described in B.2 for randomly selecting a patch in which to locate the ground trap. Do not exclude the patch selected for the elevated trap from consideration.
- **Random selection** – continue using the randomized clip strip locations in sequential order as described in B.3; assess the suitability of the next potential clip-strip location that has not previously been sampled or rejected.
- 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 clip cell

1. Navigate to the SW corner of the selected cell (0.5 m south, 0.2 m west from clipList x,y coordinates), place a pin flag.
2. Delineate the 3 m x 0.5 m clip strip 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 stakes in each of the four corners leaving ~20cm visible above ground.
4. Remove all large leaves, large fronds, and ALL fine woody debris from within the ground trap area.
    - It is not necessary to remove small leaves, fronds, etc. that are normally sampled with the elevated litter traps.

B.5 Elevated trap construction and installation

1. Center square trap frame over pin flag placed in the center of the selected clip strip cell
2. Mark plot corners with pin flags
   - 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, glue 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
   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. Attach screen to square frame with the provided zip ties (Figure 3).
   - The pre-cut screen is larger than the trap area and should not be taut across the trap, some sag is necessary to prevent litterfall from blowing away.
8. Record tagIDs of individuals overhanging the trap on the “Trap Deployment Datasheet” (RD[05]) or the multiple tree tag table if using a digital data collection tablet.

Figure 3. Fully constructed elevated litter trap. .

SOP C Field Sampling

C.1 Fine litter collection – Elevated traps

1. Navigate to plot
2. Assess and record the **litterTrapStatus** (Table 7)

Table 7. Prescribed litterTrapStatus 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 worse 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, requires replacement

3. If the trap is not in good condition (3-6), discard the litter and make necessary repairs. Broken traps should be replaced immediately if possible.
4. If the trap is in good condition (1) continue with collection procedure.
5. Discard litter > 50 cm in length, this material is not reliably collected in the elevated traps and will be sampled in ground traps
6. All woody material > 2 cm diameter will be measured according to the Coarse Wood Debris (CWD) protocol. Use calipers to measure diameter of woody branches
   a. Discard branches > 2 cm at narrowest point
   b. 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
7. Transfer all other material, including parts hanging out of the trap, into the cloth bag designated for elevated trap litter
8. Create label with clipID, date, trap type, and technician name (Figure 4), and attach to bag

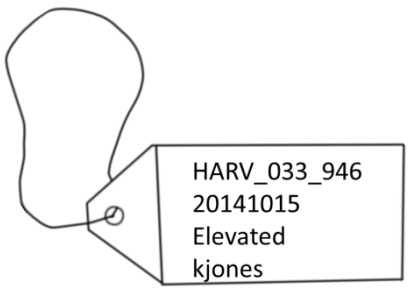

Figure 4. Example field collection label

9. Knot cloth bag to prevent material from falling out while in transport, do not use draw strings if present on bags
10. Using the 'SOP C: Field Sampling' datasheet or table (PDA) record:
    - **measuredBy/recordedBy**
    - **collectDate**: use YYYYMMDD format
    - **boutNumber**: bout number assigned by Field Operations, 01, 02…
    - **plotID:** xxxx_## - assigned by Science Operations
    - **subplotID:** see Appendix F for a plot map
    - **clipID:** unique identifier for trap location within the plot
    - **trapType:** Elevated
    - **trapCondition:** Table 7
    - **bagNumber:** transcribe from cloth bag
    - **trapMoved:** Yes, No
11. Record remarks if necessary

C.2 Woody litter collection – Ground

1. Locate stakes marking ground trap location
2. Assess and record **litterTrapStatus** (Table 8)

Table 8. Modified litterTrapStatus codes for ground traps

Code	Description
OK	Litter collected –Trap in good shape, no issues
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

- If trap condition is blocked (code=2), do not collect. If obstruction cannot be cleared, move ground trap to a new location from the clip strip list.
    - Record the new trap location/clip strip ID in the **newClipID** field
    - Clear all litter from the new strip
    - Do not collect

3. Wrap nylon cord around the four staked corners of the ground trap, delineating the trap edges.
4. Identify qualifying litter which is:
    - > 50 cm length and
    - < 2 cm diameter
5. 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.
6. Cut off and discard portions of woody branches > 2 cm diameter

7. Collect all remaining qualifying litter from within the ground trap, transfer material to the cloth bag designated for ground trap litter
    - Pieces may be cut to smaller lengths if they are too long to fit in the cloth collection bags.
8. Create a label with clipID, date, trap type, technician name (Figure 4), and attach to bag.
9. Knot cloth bag to prevent material from falling out while in transport, do not use draw strings if present on bags
10. Using the 'SOP C: Field Sampling' datasheet or table (PDA) record:
    - **measuredBy/recordedBy**
    - **collectDate**: use YYYYMMDD format
    - **boutNumber**: bout number assigned by Field Operations, 01, 02…
    - **plotID:** xxxx_## - assigned by Science Operations
    - **subplotID:** see Appendix F for a plot map
    - **clipID:** unique identifier for trap location within the plot
    - **trapType:** Ground
    - **trapCondition:** Table 7
    - **bagNumber:** transcribe from cloth bag
    - **trapMoved:** Yes, No
11. Record remarks if necessary

SOP D Laboratory Processing for Dry Mass Measurement

D.1 Sorting, drying and weighing litter samples

1. If litter and bags are very wet (i.e. dripping), hang bags to air dry before further processing.
2. Sort litter from each Elevated and Ground bag per trap pair to litter functional group.
   a. Clear adequate bench space in the laboratory.
   b. Empty the a cloth bag filled with litter onto the bench, and sort litter pieces to the functional groups in Table 9 (Elevated trap collection bags) or Table 10 (Ground trap collection bags).
   c. Clean off any dirt attached to litter from ground traps.

Table 9. Elevated trap litter functional group codes

litterCode	Description
ELVS	Leaves (including petioles, rachis and non-woody tendrils)
ENDL	Needles from coniferous species
ETWI	Twigs/branches < 2 cm diameter *and* < 50 cm length
EWDY	Woody material (e.g. bark, cones, etc.)
ESDS	Seeds
EFLR	Flowers (including pedicels)
EOTR	Other (lichen, mosses, unidentifiable material, etc.)

Table 10. Ground trap litter functional group codes

litterCode	Description
GLVS	Leaves and needles > 50 cm length (including petioles, rachis and non-woody tendrils)
GTWI	Twigs/branches < 2 cm diameter *and* > 50 cm length

   d. Label paper bags to hold sorted litter functional groups from each trap. Include sampling information from tag on cloth bag, as well as the appropriate litter **litterCode.** Choose either 8# or 25# kraft bags, depending on the quantity of litter.
3. Label the **ovenInTime** (24 hr time, e.g. 1645 for 4:45 pm) and **ovenInDate** (YYYYMMDD) that bags are placed in the drying oven on the back side of the tag.
   a. ***Critical step***: Labeling bags allows assessment of how long different batches of bags have been in the oven, especially when harvests 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; 48-hour samples may be located and removed for weighing without requiring a complete unloading of the contents of the oven.
4. Place bags of litter (excluding ETWI, EWDY and GTWI) in a drying oven set to 65° C for 48h – 120h (2d – 5d), until constant mass is attained.
5. Check the drying progress of litter bags using the "Lab Drying QC" datasheet.

a. Check the weight of the same subset of n=10 bags per date 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 (averaged across all n=10 bags, ± 0.05 g or 1%, whichever is greater)

6. Place bags of ETWI, EWDY and GTWI litter in a drying oven set to 101-105° C for 24-72 hours, until constant mass is attained. If multiple drying ovens are available, steps 5-6 and 8-9 may be occur simultaneously, otherwise, complete drying of litter material at 65° C before increasing the temperature to dry FWD. Woody material requires higher drying temperatures to release bound water.
7. Check the drying progress of litter bags using the "Lab Drying QC" datasheet.
   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 (averaged across all n=10 bags, ± 0.05g or 1%, whichever is greater)
   d. Upon removal, label bags with the **ovenOutDate** and **ovenOutTime**.
8. Weigh material from each functional group (i.e**. litterCode**) on mass balance (0.01g accuracy).
   - Weigh dried plant material immediately after removing from the drying oven, as it will absorb moisture from the air if left in ambient room conditions (particularly in humid environments). If practical to do so, remove bags from the oven and weigh one at a time.
   - 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)..
   - 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.
9. Record the **litterMass** to the nearest 0.01g on the "Litter Weight" datasheet. For large volumes of biomass that do not readily fit into a large weigh boat, use any of the following strategies:

   - Use an HDPE tray, 'larval tray' plastic box lid (or equivalent) instead of a weigh boat. *Note: paper bags or a large pieces of cardboard may absorb moisture and skew mass measurements and therefore should be avoided in humid environments.*
   - Crush or chop the biomass to reduce volume so it will fit into a weigh boat.
   - *Avoid splitting the biomass into subgroups for weighing, as uncertainty values must be added each time a subgroup is created.*

10. Once all weights have been recorded, if the collection event has been selected for bioarchive and analysis, return biomass from the leaves and needle functional groups to paper bags and store together in the large plastic bag, seal, and place in temporary storage. Samples in temporary storage can then be prepared as time permits for bioarchive and chemical analysis (SOP F).
    - All other material may be discarded
    - If the collection event has not been selected for bioarchive and analysis, all litter material may be discarded after weighing.

SOP E Data Entry and Verification

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.

The data ingest document contains at least the following spreadsheets:

- **ltrFieldSummary_in:** Spreadsheet summarizing each data ingest table, and defining table field names and ingest rules.
- **ltr_pertrap_in**: Metadata describing trap placement
- **ltr_fielddata_in**: Metadata describing individual sampling events on a per trap per plotID per sampling date basis.
- **ltr_massdata_in**: Oven-dried biomass data for each functional group per clipID per eventDate, as well as weighing QA data.

E.1 Entering and uploading field data

1. For data collected on paper datasheets: Transcribe data into appropriate MS Access database template or the protocol specific NEON data Web UI in accordance with data entry and data QA/QC protocols (AD[07]).
2. MS Access data entry fields mirror the datasheet, do not change formatting on the provided spreadsheet.
3. Example entries of each data field are provided in the 'litterfall and fine woody debris' ingest workbook (RD[09]) or the 'ltrFieldSummary' schema of the 'ltr_dataIngest_2014' MS Access database. Consult this table for appropriate values and formats for each field in the subsequent worksheets.
4. If this is the first bout at a site or a trap had to be moved to a new location (i.e. the **newClipID** field on the field collection datasheet is non-null)**,** transcribe data from the 'SOP B: Initial Deployment of Traps' Datasheets to the "ltr_pertrap_in" ingest table.
5. For collection events, record metadata for date, litterTrapStatus and bags used in the 'ltr_fielddata_in'. If the **newClipID** field is non-null, record 'yes' in the **trapMoved** field of the 'ltr_pertrap_in' table.
6. Following completion of lab processing, record the weights of each functional group in the 'ltr_labdata_in' ingest table.
7. Update permanent digital versions of the "clip-strip coordinate" lists with **status** and **date** grid cells were used.
8. Lab Drying QC data are not transcribed for ingest into the NEON CYI.
9. Once all data from the most recent sampling bout have been collected and transcribed, submit data for ingest by CYI according to the guidelines provided in RD[04].

10. For data collected on the NEON digital data collection device: Download all data according to the protocols for data handling. Address any QA/QC concerns.

E.2 Equipment maintenance, cleaning, and storage

1. Charge/replace laser rangefinder batteries, if necessary.
2. Charge GPS unit.
3. Clean grinding mill and splitters.

SOP F Processing Litter Samples for Biogeochemistry

Select samples from one elevated trap per plot from one collection event every five years to will be ground and submitted for bioarchive and chemical analysis. Two functional groups, leaves and needles, will be targeted, mass from all other functional groups and from ground traps, will be discarded.

Timing: In coniferous evergreen or broadleaf evergreen dominated systems, collect a sample for archive from the October collection event. In deciduous and mixed forest systems, select a sample from the period of peak senescence, this date may vary from site to site and from year to year. Refer to the site specific Appendix D for suggested sampling windows, use assessment of local conditions to ultimately drive this decision.

Following analysis, the remainder of samples will be sent to archive facility by the external lab. Domains will only generate one sample per trap.

1. Select dried leaves and needles from a single elevated trap per plot. In a mixed forest system, this will generate two samples for each selected clipID, one from the 'leaves' functional group, and one from the 'needles' functional group.
   a. Do not save and process for chemical analysis if dry mass is < 5mg
   b. If dry mass is > 20 g, subsample material by hand:
      1) coarsely crush material by hand into a clean container (e.g. bucket for large amounts of material, bowl for less)
      2) mix crushed material by hand to create an even blend
      3) haphazardly select one handful of crushed leaves, >5g to grind and process for chemical analysis
2. Coarsely grind material from each functional group per clipID (trap) with a Wiley Mill (0.85mm, 20 mesh size).
3. Use an appropriately sized splitter/microsplitter to generate one representative sub-sample of approximately 20 mL volume.

BEST PRACTICES

- If the split sub-sample is too large to fit into the vial in its entirety, continue splitting until a sub-sample of the desired size is generated.
- DO NOT create sub-samples with a scoopula or spatula. These tools should only be used to transfer an ENTIRE sub-sample into a vial.

4. Place the split sub-samples into 20 mL polypropylene scint vials, and label the vials with **sampleID**:
   clipID[no underscores].date.functional group code (ex. OSBS0250175.20151115.ELVS)

SOP G Sample Shipment for Biogeochemistry

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

G.1 Handling Hazardous Material

N/A

G.2 Biogeochemistry: Supplies/Containers

20 mL Scintillation vials with dried ground material in them do not require additional preservation. Vials will be shipped from the Domain lab to external labs for analysis:

1. place vial in a plastic bag
2. label bag with **collectDate, clipID** and analysis needed using a Sharpie.
3. Fill in 7x9x8 box with bubble wrap.
4. Send Ground if alone – can affix 'Up' stickers
5. place shipping inventory on top of bubble wrap

G.3 Timelines

There are no scientific limits on the time oven-dried, ground samples may be placed in temporary storage prior to shipping.

G.4 Conditions

Samples must be dry, ground, securely contained and clearly labeled.

G.5 Grouping/Splitting Samples

N/A

G.6 Return of Materials or Containers

N/A

G.7 Shipping Inventory

Shipments are to have a shipping inventory, cover letter and sample log (electronic and hardcopy) to be prepared for the contracted lab to which samples are sent. The sample log includes information such as sampleID, field metadata, and sample preparation information. Duplicate (electronic) copies of the

shipping inventory and sample log are to be sent to the contact in NEON Collections and Laboratory Analysis at the time of shipment. Also include the shipment tracking # in the email.

Shipping information for the external facility that will receive samples can be found on the CLA SharePoint page. The file is named Shipping Information for External Facilities. The NEON Collections and Laboratory Analysis contact is also listed in this file.

G.8 Laboratory Contact Information and Shipping/Receipt Days

See the CLA shipping document on CLA's NEON intranet site.

8 REFERENCES

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APPENDIX A DATASHEETS

The following datasheets are associated with this protocol:

Table 11. Datasheets associated with this protocol

NEON Doc. #	Title
NEON.DOC.002132	Datasheets for TOS Protocol and Procedure: Litterfall and Fine Woody Debris

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

APPENDIX B QUICK REFERENCES

B.1 Delineating the clip harvest strip used for litter trap placement

LOCATE AND ASSESS POTENTIAL CLIP AREA

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

STEP 2 – Select first available clip-strip location from Work Order list.

STEP 3 – Locate X-coordinate, anchor and stretch east-west tape, place pin flag.

Y-Coordinate	East-West Tape Location
1, 4, or 7	(0,0) →(20,0)
10, 13, or 16	(0,10) →(20,10)

STEP 4 – Locate Y-coordinate with laser rangefinder in HD mode (azimuth 0°), place pin flag.

STEP 5 – Locate clip cell centroid (elevated trap) 1 m North, 5 cm East

STEP 5b – Locate clip cell SW corner (ground trap) 0.5 m South, 20 cm West

STEP 5 – Assess suitability of clip-strip. Reject if not suitable.

DELINEATE 0.5 M X 2 M CLIP-STRIP

STEP 1 – Place north-south oriented string-and-stake set on west side of clip-strip. Use laser rangefinder to orient string.

STEP 2 – Place second string-and-stake set EXACTLY 50 cm to the east of first set.

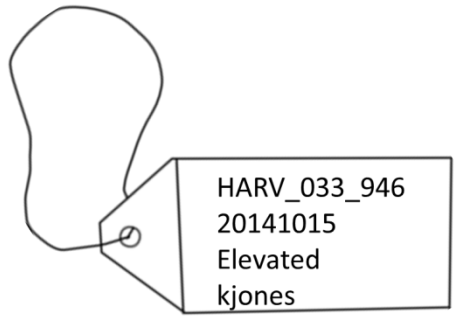
STEP 3 – Check distance between strings at both ends with ruler or tape measure.

STEP 4 – Monument clip strip corners with pvc or wood stakes

B.2 Liter 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 worse 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, requires replacement

B.3 Example field collection label



APPENDIX C REMINDERS

N/A

APPENDIX D ESTIMATED DATES FOR ONSET AND CESSATION OF SAMPLING

The dates listed here are estimated by MODIS averaged EVI values from 2001-2009 and are the 'average Greenness Increase' date a proxy for the beginning of spring, the time period when sampling for winter litterfall, and the beginning and average end of senescence. The sampling dates in this table are based on MODIS data for an area centered on the NEON flux tower; 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 will be sampled once a month all year or not at all.

Table 12. 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
01	BART	Mixed Forest	Random	Hybrid	120	220	300
01	SAWB	Deciduous Forest	Random	Spring + Senescence	110	220	315
01	HARV	Mixed Forest	Random	Hybrid	110	220	300
02	BLAN	Deciduous Forest/ Pasture Hay	Targeted	Spring + Senescence	75	210	310
02	SCBI	Deciduous Forest	Random	Spring + Senescence	85	150	320
02	SERC	Deciduous Forest	Random	Spring + Senescence	80	220	325
03	DSNY	Grassland Herbaceous		None			
03	JERC	Mixed Forest	Random	Hybrid			
03	OSBS	Evergreen Forest	Random	Monthly, Year Round			
04	GUAN	Evergreen Forest	Random	Monthly, Year Round			
04	LAJA	Cultivated Crops		None			

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence
04	MAME	Evergreen Forest	Random	Monthly, Year Round			
05	STEI	Deciduous Forest*	Random	Spring + Senescence	120	215	250
05	TREE	Deciduous Forest	Random	Spring + Senescence	120	215	250
05	UNDE	Woody Wetlands	Targeted	Spring + Senescence	125	215	285
06	KONA	Cultivated Crops		None			
06	KONZ	Grassland Herbaceous		None			
06	KUFS	Deciduous Forest	Random	Spring + Senescence	75	210	330
07	GRSM	Deciduous Forest	Random	Spring + Senescence	90	215	310
07	MLBS	Deciduous Forest	Random	Spring + Senescence	110	220	310
07	ORNL	Deciduous Forest	Random	Spring + Senescence	90	210	315
08	CHOC	Woody Wetlands	Targeted	Spring + Senescence	70	200	335
08	DELA	Woody Wetlands	Targeted	Spring + Senescence	60	205	330
08	TALL	Evergreen Forest	Random	Monthly, Year Round	75	195	330
09	DCFS	Grassland Herbaceous		None			
09	NOGP	Grassland Herbaceous		None			
09	WOOD	Grassland Herbaceous		None			
10	CPER	Grassland Herbaceous		None			
10	RMNP	Evergreen Forest	Random	Monthly, Year Round	120	210	315
10	STER	Cultivated Crops		None			
11	CLBJ	Grassland Herbaceous		None			

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence
11	KLEM	#N/A					
11	TBD	#N/A					
12	BOZE	Grassland Herbaceous		None			
12	PARA	Grassland Herbaceous		None			
12	YELL	Shrub Scrub	TBD	TBD	120	190	280
13	MOAB	Shrub Scrub	TBD	TBD	85	225	300
13	NIWO	Evergreen Forest	Random	Monthly, Year Round			
13	TBD	#N/A					
14	JORN	Shrub Scrub	TBD	TBD	80	245	320
14	SRER	Shrub Scrub	TBD	None	150	240	330
14	TBD	#N/A					
15	ONAQ	Shrub Scrub	TBD	TBD	75	170	280
15	TBD	#N/A					
15	RBUT	Deciduous Forest	Random	Spring + Senescence	105	190	310
16	ABBY	Grassland Herbaceous		None			
16	THAY	Evergreen Forest	Random	Monthly, Year Round			
16	WREF	Evergreen Forest	Random	Monthly, Year Round			
17	SJER	#N/A	Targeted	Monthly, Year Round	270	95	155
17	SOAP	Evergreen Forest	Random	Monthly, Year Round	90	185	290
17	TEAK	Evergreen Forest	Random	Monthly, Year Round	120	205	300

Domain	Site code	Primary Airshed NLCD	Trap Location Selection	Suggested Sampling Schedule	Average Greenness Increase	Beginning of Senescence	Average End of Senescence
18	BARO	Sedge Herbaceous		None			
18	TOOL	Dwarf Scrub		TBD	160	205	240
19	BONA	Deciduous Forest	Random	Spring + Senescence	135	*TBD*	250
19	DEJU	Evergreen Forest	Random	Monthly, Year Round			
19	HEAL	Shrub Scrub	TBD	TBD	135	210	245
19	POKE	Deciduous Forest	Random	Spring + Senescence	135	205	250
20	OLAA	Evergreen Forest	Random	Monthly, Year Round			
20	PUFO	Shrub Scrub	Random	TBD	0	NA	365
20	PUGR	Grassland Herbaceous		None			

* Site information has been updated from NLCD or MODIS data based on local observations

APPENDIX E SITE-SPECIFIC INFORMATION

At burn sites, litter will need to be collected and traps removed before planned management activities, then replaced soon after. Burn sites include, but may not be limited to, the following:

Table 13. Burn sites

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

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 technicians (red 'X') are supplied on a per subplot basis. 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.