TOS PROTOCOL AND PROCEDURE: MEASUREMENT OF HERBACEOUS BIOMASS

PREPARED BY	ORGANIZATION	DATE
Courtney Meier	FSU	12/05/2014

APPROVALS	ORGANIZATION	APPROVAL DATE
Mike Stewart	PSE	8/18/2015
Dave Tazik	SCI	6/04/2015

RELEASED BY	ORGANIZATION	RELEASE DATE
Anne Balsley	CM	8/24/2015

See configuration management system for approval history.

Change Record

REVISION	DATE	ECO #	DESCRIPTION OF CHANGE
A_DRAFT	03/25/2011	ECO-00280	Initial Draft Release
B_DRAFT	01/13/2014	ECO-01140	Updates from 2013. Will be finalized in next rev.
C	03/24/2014	ECO-01664	Production release, template change, and other changes as detailed in Appendix C
D	04/10/2014	ECO-01792	Updated Appendix D with site-specific information
E	10/01/2014	ECO-02309	Migration to new protocol template
F	8/24/2015	ECO-02532	• New guidance for determining WST biomass to clip • Streamlined SOP C by removing information already provided in SOP B and improving reference to SOP B. • Created new SOP F 'Herbaceous Clip for Biogeochemistry' that includes grinding and subsampling for chemical analysis • Added timing information for SOP F to Section 4 • Added SOP H with shipping information for biogeochemistry samples; added Appendix F with supporting information for SOP F. • Updates from FOPS feedback: sorting WST, accounting for Toxicodendron mass, explanation of MODIS data in site-specific Appendix. • Section 4: Clarified intended temporal sampling strategy at agricultural sites with multiple crop rotations.

TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

1 OVERVIEW

1.1 Background

Herbaceous vegetation is operationally defined in this protocol as non-woody plants (i.e. grasses, sedges, forbs, some bryophytes, and non-woody vines such as *Convolvulus spp.* and certain *Rubus spp*.), as well as woody-stemmed plants with diameter at decimeter height (ddh) < 1 cm at the time of sampling. The net primary productivity (NPP) of this plant group dominates the total NPP of grassland sites, and can contribute significantly to NPP in savannahs and some forests, even though total herbaceous biomass is low relative to that of large woody stems.

Understanding long-term trends in herbaceous community structure and biomass is very important in grazed ecosystems where these plants constitute a critical food source for wildlife and livestock. In addition, members of the herbaceous plant community can respond relatively rapidly to various global change drivers. For example, it is predicted that cool-season C3 graminoids may decrease in abundance relative to warm-season C4 graminoids in more northern latitudes as global temperatures and CO_2 concentrations continue to rise, and water availability becomes more variable.

It is standard practice for herbaceous biomass and productivity to be assessed via clip harvests, followed by sorting clipped material into current-year and previous years' growth in order to estimate annual NPP for this plant growth form. Current-year growth is often sorted by species into additional categories based on plant functional traits – e.g. cool-season vs. warm-season graminoids, or leguminous vs. non-leguminous forbs. In order to engender cross-compatibility with existing research, NEON will sort clipped biomass into functional categories that are broadly similar to those employed by the global Nutrient Network research group (http://www.nutnet.umn.edu/exp_protocol).

In sites where grazing is an important part of the management practice, it is standard practice to use grazing exclosures to estimate the productivity that is consumed by grazing herbivores. NEON will employ a standard approach where clip-harvests are performed with paired grazed/exclosed areas per plot.

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

Thanks to Daniel Milchunas of Colorado State University and Mary Ashby of the Central Plains Experimental Range USDA-ARS for valuable advice and insight.

2 RELATED DOCUMENTS AND ACRONYMS

2.1 Applicable Documents

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

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

2.2 Reference Documents

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

RD[01]	NEON.DOC.000008	NEON Acronym List
RD[02]	NEON.DOC.000243	NEON Glossary of Terms
RD[03]	NEON.DOC.005003	NEON Scientific Data Products Catalog
RD[04]	NEON.DOC.001271	NEON Protocol and Procedure: Manual Data Transcription
RD[05]	NEON.DOC.001574	Datasheets for TOS Protocol and Procedure: Measurement of Herbaceous Biomass
RD[06]	NEON.DOC.005005	Level 0 Data Products Catalog

RD[07]	NEON.DOC.000987	Measurement of Vegetation Structure
RD[08]	NEON.DOC.001788	Grazing Exclosure Assembly Instruction
RD[09]	NEON.DOC.001920	NEON Raw Data Ingest Workbook for TOS Herbaceous Plant Biomass
RD[10]	NEON.DOC.014038	TOS Protocol and Procedure: Core Sampling for Plant Belowground Biomass
RD[11]	NEON.DOC.001024	TOS Protocol and Procecure: Canopy Foliage Chemistry and Leaf Mass per Area Sampling
RD[12]	NEON.DOC.001717	TOS Standard Operating Procedure: TruPulse Rangefinder Use and Calibration
RD[13]	NEON.DOC.001271	NEON Protocol and Procedure: Manual Data Transcription

2.3 Acronyms

Acronym	Definition
ddh	Diameter at decimeter height
NPP	Net Primary Productivity

2.4 Definitions

None given.

3 METHOD

Herbaceous biomass clip-harvests occur within randomly located clip-harvest strips located in 20m x 20m plots or subplots. The goal of the clip-harvesting procedure is to estimate the amount of herbaceous biomass produced within the delineated Clip Strip area. This means that only those herbaceous plants whose stems enter the ground within the Clip Strip are clipped (exceptions to this are woody-stemmed plants with diameter at decimeter height [ddh] < 1 cm; the SOPs describe in more detail how to deal with these plants). There will typically be one clip-harvest per plot or subplot per sampling event, although sites managed for grazing receive two clips per plot or subplot per bout; see SOP C.

There are two types of plots where clip-harvests will occur: Distributed and Tower plots. Clip-harvests in Distributed and Tower plots are organized into 0.5 m x 3m gridded, numbered “cells” that cover the available sampling area within the plot. Within a cell, technicians perform clip-harvests in north/south-facing strips with dimensions of 0.1 m x 2 m (**Figure 1**). Those cells that overlap 1 m^2 and 10 m^2 nested subplots are omitted from clip-harvest sampling. Relative coordinates are assigned to the Southwest corner of each Clip Strip, which enable technicians to find the desired Clip Strip location for a given sampling bout. For reference, the Southwest corner of each 20m x 20m plot or subplot is defined as (0,0), and the Northeast corner of the plot or subplot is (20,20) (**Figure 1**). Within Distributed plots, the herbaceous biomass and productivity clip-harvest protocol is carried out only in non-forested plots with

> 50% herbaceous cover as seen from the air. Forested plots are considered to be those with NLCD Class = deciduous forest, mixed forest, or evergreen forest.

Clip harvests in 20m x 20m Tower plots will be carried out identically to those performed in Distributed plots, with the exception that all plots are harvested regardless of NLCD vegetation type. For Tower plots 40m x 40m and larger, the herbaceous clip-harvest protocol is implemented in two randomly selected 20m x 20m subpots per plot (**Figure 2**). Again, similar to clip-harvesting in Distributed plots, 1 m^2 and 10 m^2 nested subplots are not clip-harvested in Tower plots, but larger sized nested subplots within 40m x 40m and larger Tower plots may support clip-harvesting.

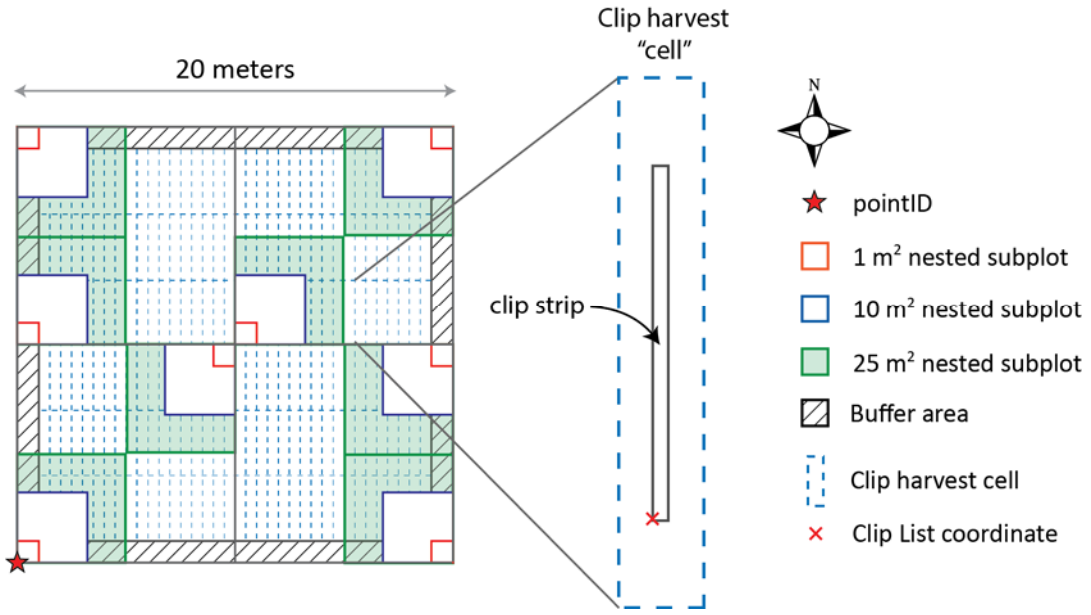

Figure 1. A Distributed plot showing the locations of 0.5m x 3m clip-harvest "cells" (dashed blue lines) that contain potential 0.1m x 2m clip-harvest strips. Coordinates corresponding to the SW corner of the Clip Strip (red 'X' in blowup) are provided to technicians in plot-specific Clip Lists. Clip List coordinates are always relative to the SW corner of the plot (red star).

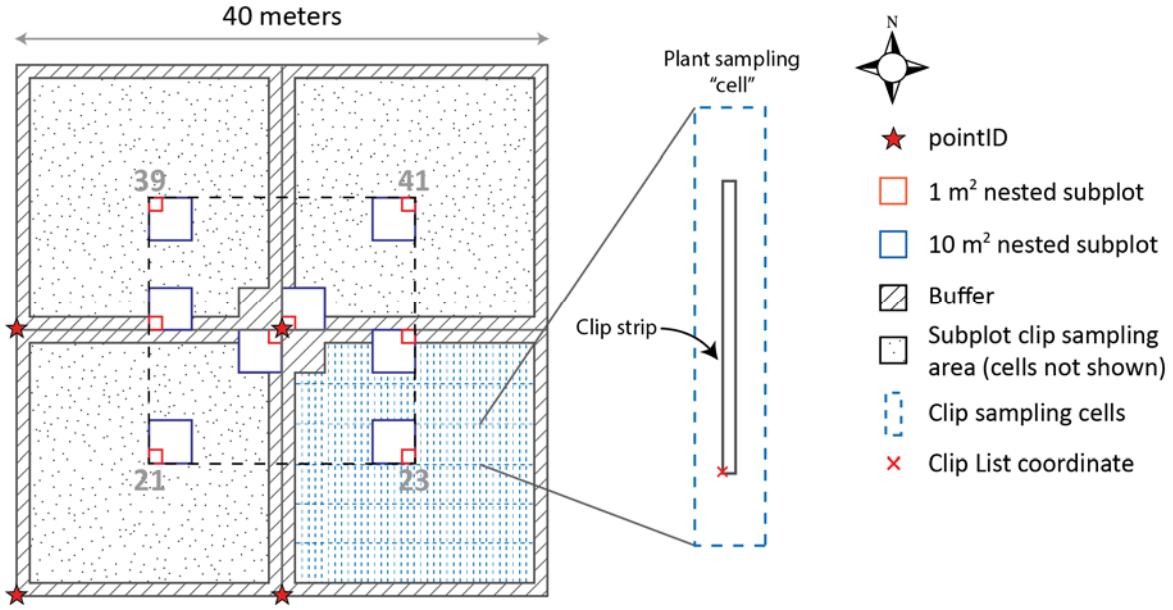

Figure 2. A 40m x 40m Tower plot showing the location of 0.5m x 3m clip-harvest cells (dashed blue lines) within a 20m x 20m subplot. Cells from the other subplots have been omitted for clarity. The Clip Strip coordinates provided to technicians (red 'X') are supplied on a per subplot basis. Clip List coordinates are always relative to the SW corner of subplots (red stars). Note that a standard 20m x 20m plot is superimposed over the centroid; this configuration allows for standardized plant diversity sampling to occur in a randomly selected subset of 40m x 40m Tower plots.

To determine a clip-harvest location within a given sampling bout, technicians are provided with a randomized list of potential Clip Strip coordinates for each 20 m x 20 m plot or subplot (referred to as "Clip Lists" hereafter). An excess number of potential clip-harvest locations within a particular plot or subplot are randomly determined a priori by NEON Science Operations, with the knowledge that not all potential locations will be suitable for clip-harvesting. That is, there may be obstacles such as rocks, trees, ant nests, etc. at any given location that will prevent carrying out a clip-harvest. Technicians should work down this list through time on a per plot or subplot basis, crossing off harvested and rejected strips on the list as work progresses from bout to bout, so that re-sampling of a given Clip Strip over the lifetime of the Observatory is minimized or eliminated.

LINKED PROTOCOLS: HERBACEOUS BIOMASS AND BELOWGROUND BIOMASS

In Tower plots, the Herbaceous Biomass and Belowground Biomass Core (RD[10]) protocols are spatially linked, and should co-occur in the same cell used for the peak aboveground biomass harvest. The Clip List should indicate whether the Belowground Biomass Core protocol has been implemented before the peak biomass clip; if the the Belowground Core sampling precedes the peak biomass clip, always clip peak biomass from the cell used for Core sampling.

Upon arriving at a plot, it is the field technician's responsibility to first locate the proposed random clip-harvest location, assess its suitability (rejecting and moving on to the next location if necessary), delineate the area for harvesting, and then perform the clip-harvest and biomass sorting. Within each plot or subplot, harvest strips are moved each year to minimize effects of harvest on subsequent biomass data. Additional clip-harvest bouts are required if grazing exclosures are employed at the site. Instructions for utilizing exclosures are provided in SOP C.

Once field work is complete at the plot, technicians must keep harvested biomass cold until sort checking is performed. Best practice is to place clipped biomass into a cooler containing -20 ˚C cold packs immediately after clipping: keeping clipped biomass cold is critical to prevent wilting, so that species' diagnostic features are preserved. Within 24-h of harvest, the same technicians who harvested and sorted the biomass in the field must then check each bag of clipped material to make sure that sorting was done properly, and in particular, that no previous years' biomass is mixed with current-year biomass. Sorted biomass is then either returned to the cooler with fresh -20 ˚C cold packs, or oven-dried as soon as possible in the laboratory and weighed.

Properly accounting for grazing, the contribution of different plant growth forms to overall aboveground biomass (sorting biomass to sub-shrubs, graminoid functional type, etc.), and determining whether biomass was produced in the current year or a previous year are the most important requirements for generating quality data from this field work.

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.

It is 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 (currently JIRA). Be sure to keep apprised of any changes made to the protocol that are documented in JIRA, but that have not yet been incorporated into the protocol.

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

4 SAMPLING SCHEDULE

4.1 Sampling Frequency and Timing

A given sampling bout should ideally be concluded within 10-14 days of initiation (Table 1), so that the plant community does not change appreciably during the time that all target plots are sampled. This guideline ensures that data collected across all plots within a given bout are comparable. The number of field technicians assigned to the clip-harvesting task should be optimized so that this goal is feasible. Herbaceous clip-harvests must be performed within Tower Plots on an annual basis, and sampling these plots is a priority.

After herbaceous plants are clipped from a given Clip Strip, the following points are critical with respect to timing:

- Place clipped biomass immediately into a cooler, and keep stored cool until it can be placed in a drying oven.
- Check field-sorted biomass for sorting accuracy within 24 h of harvest.
- On the "Field" datasheet, record the date and time that the samples were placed into cold storage after being clipped in the field, and the date and time that the samples were placed in the drying oven. These data will enable automatic calculation of the number of hours that samples were kept in cold storage.

The frequency and timing of herbaceous biomass clip-harvests depends on the type of site being sampled, as well as the type of plots sampled within the site (Table 1). Of special note are agricultural sites with target plots in different parcles that support multiple crops in different phases of rotation. Each crop that matures within the given sampling year will need to be sampled for biomass and productivity, and multiple sampling bouts will be required to capture the biomass and productivity of crops planted and harvested in different seasons. Communicating and collaborating with the site host to determine and coordinate harvest dates and sampling activities is highly advantageous in agricultural systems.

> ***Example:*** A north temperate site has 10 plots fallow, 10 plots planted with spring wheat, and 10 plots planted with corn. Plots planted with spring wheat would be harvested for bout1 (likely late May or early June); fallow plots and corn plots would be harvested for bout2 (likely mid to late August). Fallow plots may be harvested on a different schedule than corn if a cover crop was planted that reaches peak biomass later than the corn harvest.

The Herbaceous Biomass Sampling for Biogeochemistry procedure (SOP F) is a separate clip harvest effort performed every 5 years in conjunction with and at the same time as the Canopy Foliage Chemistry protocol (RD[11]) (**Table 1**). If there is no woody vegetation at a site, and RD[11] is therefore

not implemented, the Herbaceous Biomass Sampling for Biogeochemistry SOP should still be implemented every 5 years, and be timed to coincide with the AOP flight as indicated in RD[11].

Table 1. Sampling frequency and timing guidelines for herbaceous biomass clip-harvesting based on sampling type and plot type.

Sampling Type	Plot Type	Plot Number	# Events per Sampling Year	Yearly Interval[1]	Sampling Start	Sampling Stop
Agricultural	Distributed, Gradient	n=10 (min)	Once per crop cycle	Annual	2-4 weeks before crop harvest	Within 14d of sampling start
	Tower	n=10		Annual		
Ungrazed (SOP B)	Distributed, Gradient (non-forest NLCD types only)	n=20 (max)	1X per sampling year	Every 3y (one site per domain per year)	Peak biomass (APPENDIX D provides site-specific dates)	Within 14d of sampling start
	Tower	n=20 or n=30[2]	1X or 2X per sampling year	Annual	Peak biomass[3] (APPENDIX D provides site-specific dates)	Within 14d of sampling start[4]
Grazed[5] (SOP C)	Distributed, Gradient (non-forest NLCD types only)	Approach utilized is identical to that used at ungrazed sites.				
	Tower	n=20 or n=30[3]	Every 4 weeks	Annual	10-14d before animal stocking	Senescence
Biogeo-chemistry (SOP F)	Distributed, Gradient	n=20 (max)	1X per sampling year	Every 5y (coincident with Canopy Chemistry protocol)	Coincident with Canopy Chemistry protocol	Within 14d of sampling start

[1] The schedule determining which years a protocol is implemented; all sites in a domain are sampled at the given interval, unless otherwise indicated; 'annual' means a protocol is implemented every year, 'every 3 y' means there are two 'off' years following every 'on' year. This field DOES NOT indicate the number of times within an 'on' year the protocol should be implemented; intra-year frequency is provided in the '# Events per Sampling Year' field.

[2] Plot number may be reduced following initial data collection at a given site.

[3] When two clip-harvests are performed per year, Sampling Start is per bout. For example, there would be one Sampling Start date for an early season peak in cool-season graminoid biomass (May), followed by another Sampling Start date for a late-season peak in warm-season graminoid biomass (August).

[4] When two clip-harvests are performed per year, "Sampling Stop" is per bout.

[5] Grazed ecosystems are defined as those actively managed for livestock grazing. The 4 wk sampling interval should only be applied when exclosures are present and livestock are present.

4.2 Criteria for Determining Onset and Cessation of Sampling

Site-specific sampling start windows are provided in Appendix D. It is incumbent upon Field Operations to select sampling onset dates within these windows. Sampling the herbaceous biomass at agricultural sites poses an additional issue because sampling must be timed such that it occurs before crop harvest takes place. Field Operations Managers responsible for agricultural sites must determine anticipated harvest dates that will influence when sampling should take place.

4.3 Timing for Laboratory Processing and Analysis

Because herbaceous biomass continues to be biologically active after clipping and before drying (i.e. plant cells continue to respire and therefore lose mass), it is important to place clipped samples into the drying oven as soon as possible after clipping occurs. Ideally, samples will be placed in the drying oven within 24 h of clipping in the field, and must be kept in cold storage the entire time between clipping in the field and drying in the laboratory. Keeping samples in cold storage mitigates mass loss by slowing cellular activity. However, when it is not possible to dry samples in the laboratory within 24 h of clipping, it is acceptable to keep samples in cold storage for up to a maximum of 5 days following clipping.

Once samples are dry, they may be weighed immediately (SOP E.1), or placed in temporary storage prior to weighing. There are no scientific limits on the time oven-dried samples may be placed in temporary storage prior to weighing and processing. However, samples should be stored temporarily for no more than 30 days to prevent backlogs from forming (SOP E.1).

4.4 Sampling Timing Contingencies

Table 2. Contingency decisions

Delay/Situation	Action	Outcome for Data Products
Hours	If delay prevents completion of clip-harvest strip: 1. Ensure all small bags of sorted biomass are labeled, 2. Place small bags into a 25# bag and label, Resume harvest of same clip-harvest strip ASAP	No adverse outcome.
	If delay occurs between plots, resume harvest of next clip-harvest strip ASAP.	
1-7 days	If delay prevents completion of clip-harvest strip: 1. Ensure all small bags of sorted biomass are labeled, 2. Place small bags into a 25# bag and label,	May create potential change in observed NPP, and may increase uncertainty in consumption estimates at grazed sites.

Delay/Situation	Action	Outcome for Data Products
	3. Store already clipped biomass in a cooler/refrigerator (okay), or oven-dry as per protocol (best), 4. Resume harvest of same clip-harvest strip ASAP with new labeled bags, and 5. Combine dried biomass per functional group for weighing when all biomass is dry.	May also be difficult to complete clip harvest of all plots in 10-14 day window if delay approaches 7 days.
	If delay occurs between clip-harvest strips, resume harvest of next strip ASAP.	
8-13 days or longer	If delay prevents completion of clip-harvest strip: 1. Ensure all small bags of sorted biomass are labeled, 2. Place small bags into a 25# bag and label, 3. Store already clipped biomass in a cooler/refrigerator (okay), or oven-dry as per protocol (best), 4. Resume harvest of same clip-harvest strip ASAP with new labeled bags, and 5. Combine dried biomass per functional group for weighing when all biomass is dry.	More uncertainty in biomass and NPP estimates, especially in grazed systems. Aboveground biomass per unit area may change in the field over this length of time.
	If delay occurs between clip-harvest strips, resume harvest of next strip ASAP	

For QA/QC of the weighing and data entry portion of the laboratory work, 10% of the previously dried, weighed samples are selected for QA/QC per sampling bout. For these samples, technicians re-weigh and record the QA mass in the **qaDryMass** field on the "Lab Weighing" datasheet.

5 SAFETY

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

Personnel working at a NEON site must be compliant with safe field work practices as outlined in the Operations Field Safety and Security Plan (AD[02]) and EHS Safety Policy and Program Manual (AD[01]). Additional safety issues associated with this field procedure are outlined below. The Field Operations Manager and the Lead Field Technician have primary authority to stop work activities based on unsafe

field conditions; however, all employees have the responsibility and right to stop their work in unsafe conditions.

Additional safety issues associated with this field procedure include potential exposure to *Toxicodendron* oils (discussed in APPENDIX E).

6 PERSONNEL RESOURCES

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.

See also APPENDIX E, section E.1 for equipment related to minimizing exposure to toxic oils from *Toxicodendron spp.*

Table 3. Equipment list – durable items required for per-plot biomass clip harvesting and sorting (quantities are for two technicians)

Item No.	R/S	Description	Purpose	Quantity	Special Handling
Durable Items					
MX100703	S	GPS receiver, recreational accuracy	Navigate to sampling location	1	N
MX100320	S	Compass with mirror and declination adjustment	Locate clip-harvest strips (with measuring tape)	1	N
MX100322	R	Laser Rangefinder, 0.3 m accuracy	Locate clip-harvest strips within plots/subplots; use when plot slope > 20% or brushy	1	N
MX103218	R	Foliage filter	Allow laser rangefinder use in dense vegetation	2	N
MX104359	R	White reflector or reflective tape	Reflective target for laser rangefinder; aids in measuring distance to target accurately	1	N

Item No.	R/S	Description	Purpose	Quantity	Special Handling
EG07570000 EG07570001	R	Grazing exclosure, tall or short grass system	Prevent herbivory at clip location in actively grazed sites		N
	S	Hammer	Install and remove grazing exclosure stakes	1	N
	R	Pruning shear	Clip plants		N
MX106656	R	Magnifier hand-lens, 10X	Aid in species identification	1	N
MX103211	R	Magnifier hand-lens, 20X	Aid in species identification	1	N
MX100358	R	Cold packs	Chill perishable samples in field		N
	R	Pre-marked string and stake sets	Delineate clip harvest strip		N
MX104361	R	Chaining pins or other suitable anchor	Anchor measuring tapes	2	N
MX100722	R	Measuring tape, minimum 30 m	Locate clip-harvest strips within plots/subplots .Plot slope < 20%; grassland, savannah	1	N
MX100543	R	Ruler, 30 cm	Delineate clip-harvest strip	1	N
	R	Forceps	Identify and sort plants	2	N
	S	Work gloves	Protect hands	2	N
	R	Cooler	Chill perishable samples in field	1	N

Item No.	R/S	Description	Purpose	Quantity	Special Handling
Consumable Items					
	R	Survey marking flag, PVC or fiberglass stake	Delineate sampling area	4	N
MX105089	R	Paper bags, #8[1]	Contain clipped herbaceous biomass, sorted to functional group	50[2]	N
MX103232	R	Paper bags, #25[1]	Organize smaller bags from a given clip strip	10[2]	N
	R	Permanent marker	Label paper bags	2	N
	S	CR123A battery	Spare battery for laser rangefinder	2	N
	S	AA battery	Spare battery for GPS receiver	2	N
Resources					
RD[05]	R	Herbaceous Biomass Field Datasheets	Record sampling metadata		N
	R	Per plot or subplot Clip Lists	Identify random Clip Strip locations	As needed	N
	R	Field guide, regional flora reference guide and/or key	Identify leguminous forbs and graminoids to species	1	N

R/S=Required/Suggested

[1] Bag size may be adjusted as necessary based on size/bulk of plants being clipped. For example, sites with tall grasses will require larger bags.
[2] Quantity may be adjusted as necessary based on field experience at a given site.

Table 4. Equipment list – Post-field sampling tasks

Item No.	R/S	Description	Purpose	Quantity	Special Handling
Durable Items					
MX103211	R	Magnifier hand-lens, 20X	Aid in species identification	1	N
MX106656	R	Magnifier hand-lens, 10X	Aid in species identification	1	N
	R	Cooler	Chill perishable samples	1	N
	R	Cold packs	Chill perishable samples	Vari-able	
Resources					
RD[05]	R	Completed Herbaceous Biomass Field Datasheets	Contains field-collected sampling metadata	Vari-able	N
	R	Field guide, regional flora reference guide and/or key	Aid in distinguishing morphologically similar species to functional groups	1	N

R/S=Required/Suggested

Table 5. Equipment list – Processing herbaceous biomass clip-harvest samples in the laboratory

Item No.	R/S	Description	Purpose	Quantity	Special Handling
Durable Items					
MX100230	R	Drying oven	Dry samples	2 (typically)	N
MX100265	R	Balance, 0.01 g accuracy	Weigh samples	1	N
MX100689	R	Weigh boats, large	Contain dried sample while weighing	Variable	N
MX103931	S	Plastic tray	Contain oversized samples while weighing	1	N
Consumable Items					
	R	Pre-printable labels	Label scintillation vials	As needed	N
RD[05]	R	Datasheets: • Lab Drying QC Datasheet • Lab Weighing Datasheet	Recording dry weight of herbaceous biomass	As needed	N

6.2 Training Requirements

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

For the field component of this protocol, technicians must be trained in navigating to points in the field with a GPS and manual methods. Most critically, technicians must be trained to quickly identify common herbaceous species at the sites within the region of employment. Because different herbaceous functional groups can be sensitive indicators of ecosystem responses to global change (e.g. N deposition, warming, rising CO_2), it is very important that field technicians within a domain can accurately and quickly identify C3 and C4 graminoids as well as identify leguminous and non-leguminous forbs within that domain.

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

6.3 Specialized Skills

The lead plant technician must possess the demonstrated ability to identify required plants to functional group – either via visual inspection, or via visual inspection in combination with a dichotomous or polyclave key.

- Identification of all leguminous forbs to functional group, in the absence of flowers, is required.
- Identification to species is not required for non-leguminous forbs and woody stemmed plants.
- Identification to species is required for cool-season (C3) and warm-season (C4) graminoid functional groups. Technicians should be able to identify graminoids vegetatively.

Ideally, each team member should know how to use diagnostic traits and a dichotomous or polyclave key to identify unknown species.

6.4 Estimated Time

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

An experienced two-person team will require between 1-3 hours to perform clip-harvest sampling within a given Clip Strip when biomass is sorted to functional group. This time range includes identifying

an acceptable clip-harvest location, delineating the clip-harvest strip, and clipping qualifying herbaceous biomass. The clip-harvest may take closer to 1 h per plot if herbaceous vegetation is sparse.

For those harvests at sites managed for grazing that are not sorted to functional group, the time required to clip-harvest herbaceous biomass from a Clip Strip should be < 1 hour (including all the tasks described above).

7 STANDARD OPERATING PROCEDURES

SOP A Preparing for Sampling

A.1 Sampling Equipment Preparation and Checklist

Table 6. Sampling equipment preparation checklist

✓	Item	Action
	GPS unit	Charge Load target locations
	Compass, mirror-sight	Check/set correct declination[1]
	TruPulse 360R	• Check battery, charge (if possible) • Clean lenses with lens cloth or lens tissue (if necessary) • Check/set correct declination[1]. See RD[12] for details. • Calibrate TruPulse tilt-sensor (only necessary after severe drop-shock; see RD[12] for details).
	Hand clippers	Clean and sharpen blades (if necessary)
	Re-usable cold packs	Place in –20 ˚C freezer
	Pre-marked string and stake sets[2]	Fabricate if necessary: 1. Cut 2.5m of ⅛" diameter nylon cord 2. Mark cord at each end with a sharpie so that the center section of the cord measures exactly 2m between the markings. 3. Tie each end to an 8" or longer tent stake.
	Herbaceous Biomass Field Datasheets (RD[05])	Print on all-weather paper
	Per plot or subplot Clip Lists[3]	Print on all-weather paper

[1] Declination changes with time and should be looked up annually per site: http://www.ngdc.noaa.gov/geomag-web/
[2] Pre-marked string and stake sets are used to temporarily delineate Clip Strip boundaries, and require fabrication prior to field work. Each set consists of two tent stakes connected by nylon cord.
[3] Provided separately by Science Operations once plot establishment has been completed.

A.2 Early-season preparation at a grazed site

Grazing exclosures are deployed only within Tower plots, and one exclosure should be constructed for each 20m x 20m plot or subplot.

- Inspect existing grazing exclosures for wear and damage.
- Construct additional grazing exclosures as needed according to plans outlined in RD[08].
  - The drawings in RD[08] depict two different styles of grazing exclosure, with heights optimized for low-stature grassland vegetation (plants ~ 30cm height), and mid-stature grassland vegetation (plants ~ 1 m height).
  - Choose the exclosure height so that the exclosure height is approximately equal to or just greater than the height of the vegetation.
  - Provide feedback to Science Operations if the assembly document requires updating to include an expanded range of exclosure heights.
- Deploy exclosures within Tower plots or subplots prior to the onset of grazing.
  - The Field Operations Manager must communicate with the site host to ascertain when grazing begins in a given growing season.
  - For each Tower plot or subplot, place an exclosure over the first suitable Clip Strip, and stake the exclosure to the ground.

Follow steps in SOP C to locate clip-harvest strips and assess suitability.

SOP B Field Sampling Sites Not Managed for Grazing

At non-grazed sites, which are defined here to mean those that are not actively managed for grazing, one Clip Strip per plot or subplot per bout is harvested. Distributed plots are harvested once per year at peak biomass (on a 3 y interval), and Tower plots are harvested annually either once or twice per growing season, depending on whether a particular plant community shows seasonally distinct biomass peaks (e.g. C3 peak in spring, C4 peak in summer).

At non-grazed sites, sorting clipped biomass to functional group is required. If two harvests are performed in Tower plots, both harvests must be sorted to functional group.

If plant diversity sampling occurs prior to herbaceous clip-harvest sampling in a given sampling year, identify and demarcate a suitable Clip Strip prior to performing diversity sampling. This will ensure that the Clip Strip is not trampled during diversity sampling.

B.1 Sample Collection in the Field

1. Navigate to the plotID to be sampled (using the GPS if necessary).
2. Use the plot- or subplot-specific Clip List ([plotID]_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.
   - The Clip List provides the randomized list of potential Clip Strips per plot or subplot.
   - The Clip List is also updated by NEON technicians to provide a record of which Clip Strips have already been harvested or rejected.
   - For Tower plots 40m x 40m and larger, herbaceous biomass sampling is performed in a randomly selected subset of available subplots.
     - Clip Lists are only provided for these randomly selected subplots.
3. Locate the relative X,Y-coordinates of the Clip Strip SW corner within the plot or subplot. The procedure used to locate the X-coordinate depends on the value of the relative Y-coordinate and the different procedures are detailed below:
4. ***If the 'offsetNorthing' coordinate for the clipID 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 1), and stretch the tape taut.*
   b. Place a pin flag at the desired relative X-coordinate (offsetEasting).
   c. Standing directly over the pin flag that was just placed at the X-coordinate, use either a compass and tape, or the TruPulse in **HD** mode with a reflective surface, to locate the Y-coordinate (offsetNorthing).

- Make sure the azimuth is as close to 0˚ as possible (True North) when finding the Y-coordinate (see **Figure 1**).
- Avoid trampling the potential clip-harvest strip as much as possible.

d. Place a pin flag at the Clip Strip (X,Y) location; this point corresponds to the SW corner of the Clip Strip.

5. If the 'offsetNorthing' coordinate for the clipID is > 10:

a. Run a tape* East/West from the plot centroid (10,10) to either the (0,10) position or the (20,10) position (**Figure 1**):

offsetEasting	Tape Layout[1]
1 < offsetEasting < 10	From (10,10) to (0,10)[1]
10 < offsetEasting < 20	From (10,10) to (20,10)[1]

[1] Use a compass or the TruPulse in **AZ** mode to guide the tape along the correct azimuth. For plots < 20% slope and lacking brush, an additional tape can be run N/S connecting the SW/NW or SE/NE plot markers to help find the (0,10) and (20,10) points if desired.

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

c. Standing directly over the pin flag that was just placed at the X-coordinate, use either a compass and tape, or the TruPulse in **HD** mode with a reflective surface, to locate the Y-coordinate (offsetNorthing).

- Make sure the azimuth is 0˚ (True North) when shooting the TruPulse to find the Y-coordinate.
- Avoid trampling the potential Clip Strip as much as possible.

d. Place a pin flag at the Clip Strip (X,Y) location; this point corresponds to the SW corner of the Clip Strip.

BEST PRACTICE TIPS

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

6. Assess whether the Clip Strip location is suitable, and accept or reject the location (see **Figure 3** flow-chart). Obstacles, disturbances, and/or irregularities may include trees, large rocks, ant nests, etc. Strips should also be rejected if clipping a particular plant specimen in the strip would

influence plot-level diversity. That is, the plant in question exists nowhere else in the plot or subplot.

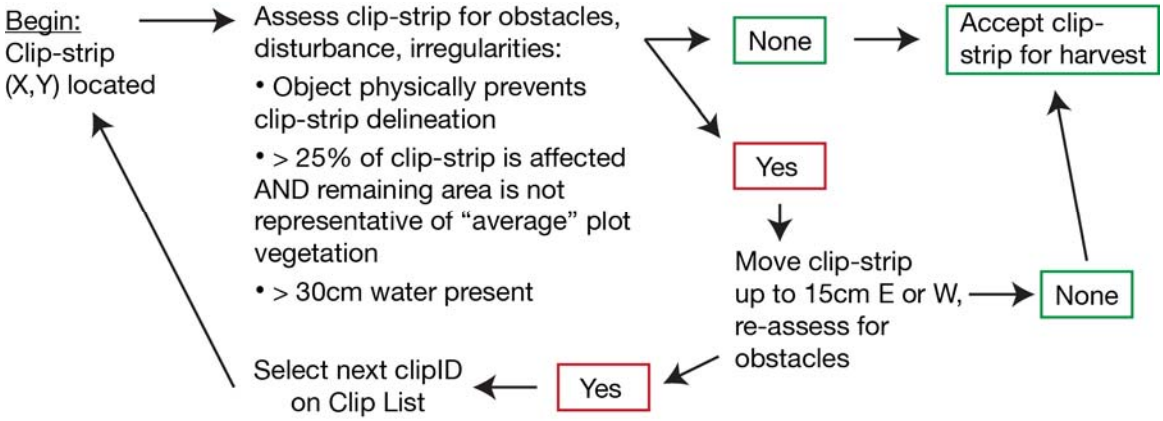

Figure 3. Flow chart to guide assessing potential clip cells for clip-harvest suitability.

7. **If the clipID is rejected, record why in the "status" column on the Clip List, select the next clipID on the list, and return to step (3) above. Otherwise, update the "status" column and proceed to step (6).** Update the "status" column in the Clip List using codes in Table 7.

Table 7. Codes to document acceptance/rejection of clip-harvest strips on the list of Clip Strip coordinates.

Code	Definition
0	Rejected; disturbance, obstacle, and/or irregularity encountered within the clipID cell
1	Accepted, no exclosure
2	Accepted, exclosure
3	Rejected temporarily, inundated
4	Rejected temporarily, uncommon plant
5	Co-located belowground biomass core sampling

8. Delineate the accepted Clip Strip for harvesting.
    a. Using one of the pre-marked string and stake sets, line up one of the marks with the pin flag, and push one stake into the ground.
    b. Stretch the string and second stake from the South to the North end of the Clip Strip, using the compass or the TruPulse to orient the string in a North/South direction.
    c. Keep the compass or TruPulse at least 50 cm from non-aluminum metal plot markers, eyeglasses, wristwatches, tent stakes, etc.
    d. Use a ruler to place the second string-and-stake set 10 cm to the right (east) of the first set. Check that the distance between the two strings is exactly 10 cm at both ends of the Clip Strip.

e. The two sets of marks on the two string-and-stake sets now clearly delineate a 0.1 m x 2 m area for clip-harvesting.

9. If there is no herbaceous biomass in a Clip Strip, AND the Clip Strip is deemed representative of the plot:
    - Record 'targetTaxaPresent = N' in the **remarks** field.
10. Using a sharpie, label 8# kraft paper bags with the following information (use larger bags if vegetation is large-stature; if vegetation is wet, cloth bags may be used):
    - **boutNumber**; use *XX* format, where XX is the bout number assigned by Field Operations, e.g. '01'. Values of **boutNumber** reset every year.
    - **date**; use *YYYYMMDD* format
    - **clipID**; as provided in Clip List; use "*plotID_clipCellNumber*" format, e.g. CPER_001_0126
    - **bagCount**; the total # of bags generated from a given clipID, does not include OSD since OSD bags are discarded after the sort-check.

Table 8. Herbaceous clip-harvest functional groups with corresponding herb codes, descriptions of functional groups, and clipping guidelines.

herb Code	Description	Clipping Guidelines
ALL	Clipped herbaceous biomass not sorted to any of the functional groups below; use for non-peak-biomass harvests at sites managed for grazing with more than one sampling bout per growing season	Clip 1-2 cm above the ground; plants < 1-2 cm tall are ignored.
BRY	Bryophytes; lichens are not part of this group, and are ignored	Clip only species that show distinct annual growth, and only clip current year growth
CSG	Cool-season graminoids; includes all grasses, sedges, rushes, etc. with the C3 photosynthetic pathway	Clip 1-2 cm above the ground; plants < 1-2 cm tall are ignored. DO NOT clip the crowns of perennial graminoids, as this will damage or kill the plant (**Figure 4**)
WSG	Warm-season graminoids; includes all grasses, sedges, rushes, etc. with the C4 photosynthetic pathway	Clip 1-2 cm above the ground; plants < 1-2 cm tall are ignored. DO NOT clip the crowns of perennial graminoids, as this will damage or kill the plant (**Figure 4**)
LFB	Leguminous forbs; includes all herbaceous annual and perennial members of the Fabaceae family	Clip tissue produced in the current year; DO NOT clip any aboveground perennial parts. Plants < 1-2 cm tall are ignored.
FRB	Non-leguminous herbaceous annual and perennial forbs	Clip tissue produced in the current year; DO NOT clip any aboveground perennial parts. Plants < 1-2 cm tall are ignored.
WST	Woody-stemmed plants with ddh < 1 cm	Treat nodes where current-year woody

herb Code	Description	Clipping Guidelines
		growth emerges from previous years' woody growth as a "rooting point". Clip only leaves and twigs produced in the current year attached to nodes that lie within the Clip Strip. Harvested current-year material may leave the Clip Strip so long as the node lies within it. It is not necessary for the actual rooting point to lie within the Clip Strip. See training materials for example diagram.
OSD	Old standing dead material produced in a previous growing season	Make sure standing dead material produced in the *current* growing season is sorted into the correct group above

11. If required to facilitate clipping, remove any cactus biomass lying within the Clip Strip. If the individual is rooted within the Clip Strip, clip off the cactus plant at the soil level. ***Only clip cactus plants that prevent access to herbaceous biomass that must be clipped.***

    - Dispose of the cactus biomass outside of the plot.

12. Clip and sort all herbaceous aboveground biomass rooted within the Clip Strip into the functional groups in Table 8. See Table 9 at end of this SOP for common points of confusion and guidelines for problem plants.

- Do **NOT** clip herbaceous vegetation that passes through/leans over the Clip Strip but is not rooted in the strip (this includes non-woody vines; WST group is an exception, see below).
- **DO** clip all herbaceous biomass of plants rooted within the strip > 1-2 cm in height. That is, include leaves in the harvest that exit the strip but originate from stems rooted within the strip.
- **DO** clip leaves and twigs of woody stemmed plants with ddh < 1cm that are produced in the current year AND originate from nodes that fall within the clip strip. It is not necessary that the individuals are rooted in the clip strip as long as the most recent node from which they originated falls within the strip.
- Working in pairs, technicians may split the clipping labor one of two ways:
    - Divide the 2 m Clip Strip into 1 m sections, label two bags for each herbCode so that each technician has a set of bags, and then combine the biomass for each herbCode when clipping is finished.
    - Divide the clipping labor among the herbCodes. For example, one technician clips cool- and warm-season graminoids while second technician clips leguminous and non-leguminous forbs.

BEST PRACTICE TIPS

- Target one herbCode at a time.
- Clip slowly, and immediately sort clipped vegetation into labeled bags.
- Place full bags immediately into a cooler with cold packs.

13. When clipping is finished, group all bags from the current clipID into a larger 25# bag and return to the cooler.
14. Record the total number of bags from the Clip Strip in the **bagCount** field and the time the bags were placed in the cooler in the **time** field on the "Field" datasheet.
15. If there is no biomass associated with a given herbGroup, note in the **remarks** field so it is clear the given herbGroup was not forgotten.
    - Record 'absent = XXX, YYY' etc., where XXX and YYY are the 3-letter codes for the absent herbGroups.

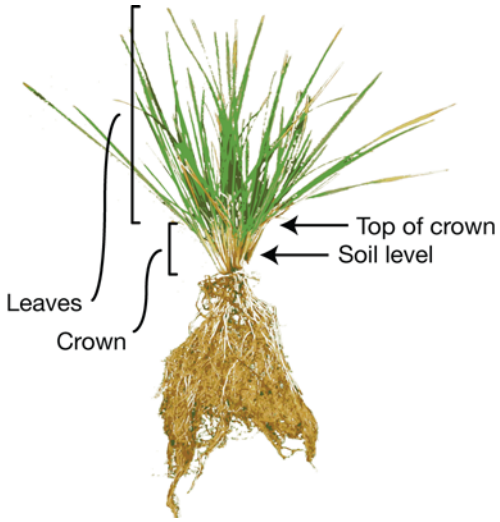

Figure 4. A perennial graminoid, showing the location of crown material relative to leaves and the soil surface

B.2 Points of Confusion

Table 9. Additional guidelines for plant growth forms that require special handling or consideration.

Growth Form	Guidelines
Barrel- and saguaro-type cactus species	DO NOT CLIP; clip around these plants.
Cholla- and pad-type cactus species	DO NOT CLIP for biomass; these plants are counted and measured with the Vegetation Structure protocol. Clip and remove pad cactus only if their presence hinders the removal of other herbaceous functional groups.
Toxicodendron spp.	Handle according to procedure in APPENDIX E
Agave, Yucca, and related species	DO NOT CLIP; these plants are counted and measured with the Vegetation Structure protocol
Ferns	DO NOT CLIP; these plants are counted and measured with the Vegetation Structure protocol
Clumped plants (caespitose graminoids, large rosette forbs, etc.)	Clip only the part of the clump that is rooted within the Clip Strip. See training materials for examples.
Litter, prostrate on the ground	Ignore; prostrate litter material produced in a previous year is not sampled as part of this protocol.
Evergreen herbaceous plants for which distinguishing current-year from past-year growth is difficult	Do your best to distinguish current-year from past-year growth, and be conservative. Make sure all technicians make consistently similar decisions when clipping.
Epiphytes	DO NOT CLIP
Multiple rooting points, at least one of which is in the Clip Strip	Only clip biomass associated with rooting points located within the strip..
WST that are leguminous (N fixers)	Leguminous WST should be grouped in with the leguminous forb functional group (LFB); the functional aspect of the grouping is more important than the morphological aspect.
Small sub-shrubs (e.g., Ericaceous plants) that are difficult to differentiate between WST or FRB	Use the USDA plants database as a consistent means of determining the growth form for a given species. If denoted a ‘shrub’ or ‘sub-shrub’ by USDA, classify as WST

B.3 Sample Preservation

- Keep paper bags with clipped vegetation in a cooler with cold packs to minimize wilting and to preserve diagnostic features for the post-harvest sort-check.
- Change cold packs for fresh ones every 12 h or transfer to a 4 ˚C refrigerator if a drying oven is not immediately available.

- Transfer bags to the drying oven as soon as possible after the post-harvest sort-check. Monitor drying progress with the "Lab Drying QC" datasheet.

IMPORTANT: Record the **clipDate** and **time** on the "Field" datasheet AND **ovenInDate** and time on both the sample bags and the "Lab Weighing" datasheet so that the number of hours the bags were stored cold can be automatically calculated.

SOP C Field Sampling with Grazing Management

At "grazed" sites, which are defined here to mean those that are actively managed for grazing, two Clip Strips per plot or subplot per bout are harvested. One Clip Strip per plot or subplot is protected from grazing with a grazing exclosure, and the second Clip Strip per plot or subplot is exposed to the managed level of grazing pressure. Exclosures should be placed at a suitable random grid-cell location prior to grazing onset in the current season. After each clip-harvest sampling event, the exclosure is moved to the next suitable random location, and clipping occurs on a 4 week interval. A 4 week clipping interval is ONLY employed when grazing exclosures are in use.

There are several common scenarios that affect the deployment of grazing exclosures at sites managed for grazing (Table 10):

Table 10. Appropriate exclosure strategies for atypical grazing management scenarios.

Grazing Management Scenario	Exclosure Strategy
Animals are stocked after senescence has occurred, and removed before plants begin to grow again the next season	Not necessary to deploy exclosures
A portion, but not all, of the Tower plots are managed for grazing	• Place exclosures only over those Tower plots that are actively managed for grazing. • When applicable, the clip that is sorted to functional group is chosen to be consistent with the other Tower plots.
Animals are stocked year-round	Deploy grazing exclosures year-round, even if sampling bouts cannot be performed in the absence of seasonal labor.

At grazed sites, sorting clipped biomass to functional group is only required for one harvest per year, and the harvest that is sorted to functional group should be selected to coincide with peak biomass. If there are multiple biomass peaks, choose the time point with the highest peak biomass for functional group sorting. All other harvests are not sorted to functional group, and are assigned **herbCode** = 'ALL'.

C.1 Sample Collection in the Field

1. Navigate to the plotID to be sampled (using the GPS if necessary).
2. ***You will sample two Clip Strips per plot.*** The first Clip Strip is the one underneath the grazing exclosure that was previously put in place. The second Clip Strip location will be selected from the Clip List provided by Science Operations.
    - The Clip List provides the randomized list of potential clip-harvest strips per plot or subplot, and also indicates which Clip Strips have already been harvested or rejected.

3. ***For the Clip Strip without an exclosure:*** Locate the SW corner of the Clip Strip using the method described in SOP B.
4. ***For both <u>Clip Strips:</u>*** Perform the clip-harvest as indicated in SOP B.
5. When you are finished harvesting the Clip Strip that was protected by the grazing exclosure, move the exclosure to the next suitable Clip Strip location on the Clip List.
6. Stake the exclosure to the ground, and proceed to the next plot.

SOP D Post-Field Sampling Tasks

D.1 Check Sorting of Clipped Biomass

The objectives for this task are:

- ***Within 24-h of harvest,*** check bags of field-sorted biomass for sorting accuracy.
- Remove any litter produced in a previous year from material produced in the current year. The most common error, particularly for clip-harvests performed late in the growing season, is to confuse material that was produced in the current year and has already died with material that was produced in a previous year.
- Remove any material produced in the current year that belongs in another biomass group. For example, leguminous forbs should not be mixed with non-leguminous forbs.
- It is not necessary to check sorting accuracy for bags with herbCode = OSD. The OSD material is used as a reference during the laboratory sort-check, and is then discarded.

To sort-check clipped biomass:

1. Select a 25# kraft bag (or bags) containing all of the biomass from a given clipID.
2. Make sure that all bags from a given clipID are collated in the correct 25# kraft bag(s).
3. Choose a smaller bag containing clipped biomass from one herbCode only, and carefully check the biomass that was sorted in the field.
4. Set aside biomass that does not belong in the bag into separate piles (i.e. one pile for each herbCode).
5. Place any previous years' litter into the "OSD" bag.
6. Place sorted, checked biomass back into the original bag.
7. Clean the work area of any debris, and proceed to the next herbCode from the same clipID, sorting again as in step (3).
8. Once all herbCodes have been checked for sorting accuracy, place piles of resorted biomass into the appropriately labeled herbCode bags, and place all smaller bags back into the 25# bag(s).
    - Exception: Discard the OSD bag.
9. Place the 25# bag(s) into the drying oven (SOP E), or return to the cooler for continued temporary storage.
10. Clean the workspace, and proceed to checking herbCode bags from the next clipID.

BEST PRACTICE TIPS

- The lead plant technician or botanist should spot-check 10% of the re-sorted biomass piles before they are re-bagged.
- Spot-checks from a person skilled in plant species identification is particularly important early in the field season when seasonal field technicians may be less familiar with local flora.

D.2 Refreshing the Sampling Kit

- Make sure the following consumables are available in sufficient quantity for the next round of clip-harvests:
  - Paper bags, 8# and 25# kraft (or the necessary size given site vegetation stature)
  - Rite-in-the-Rain paper for printing field datasheets
  - Permanent markers for labeling bags in the field
- Return cold packs to the -20 ˚C freezer to refresh.

D.3 Equipment Maintenance and Cleaning

- Clean blades of hand clippers with an appropriate solvent (oil, ethanol, water), and dry thoroughly.
- Recharge batteries for the GPS unit (if necessary).
- Recharge batteries for the TruPulse (if applicable).

SOP E Laboratory Processing of Herbaceous Biomass Samples

E.1 Drying and Weighing Clipped Herbaceous Biomass

Clipped herbaceous biomass should be placed in the drying oven as soon as possible following the post-field-sampling sort check.

1. Label each 25# bag (with smaller bags for each herbCode inside) with the date and time it is placed in the drying oven.
   - These data are the **ovenInDate** and time required during data entry.
   - ***Critical step***: Labeling bags allows assessment of how long different batches of bags have been in the oven, especially when harvests from multiple days occupy the same oven.
2. Place labeled 25# bags into a **60 ˚C drying oven for 48h – 120h (2d – 5d)**.
3. After placing all bags from one clipDate in the oven, check the drying progress of clipped biomass using a subset of 10 - herbCode samples, and the "Lab Drying QC" datasheet.
4. Check the weight of the same selected subset of n=10 herbCode sample bags per clipDate after day 1, 2, 3, etc. Record these weights each day on the "Lab Drying QC" datasheet.
5. Calculate the difference in weight between the latest two time points for each bag.
6. Samples are dry when the average weight difference between the latest two timepoints = 0 (averaged across all n=10 bags, ± 0.05 g)

DRYING TIPS

- A spreadsheet calculator is useful for calculating the average weight difference. Ask your Domain Manager for the calculator already created by Field Operations staff.
- To save time, select and weigh larger 25# bags from a given clipID that contain all of the smaller herbCode bags for that clipID. Plant material may be weighed WITH the bag, and it is not necessary to remove and weigh individual bags for a single herbCode (write herbCode = 'ALL' on the Lab Drying QC Datasheet).
- Focus on the heaviest bags, as these will likely take the longest to dry.

7. Remove bags of dried biomass from the drying oven, and label bags with **ovenOutDate**/Time.
   - Dried plant material should be weighed immediately after removing from the drying oven, as it will absorb moisture from the air if left in ambient room conditions (particularly in humid environments).
     - If using this method, it is helpful to remove bags from the oven and weigh one at a time.

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

8. Weigh biomass from each herbCode from each clipID using a mass balance (0.01g accuracy) and a large weigh boat.
    - Record **dryMass** on the "Lab Weighing" datasheet: nearest 0.01 g, plant material ONLY (without the bag).
    - For large volumes of biomass that do not readily fit into a large weighboat, use the following strategies:
        - Use a large plastic tray (or equivalent) instead of a weigh boat (see equipment list).
        - 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.
9. Record required metadata for each sample in the "Lab Weighing" datasheet:
    - **clipCellNumber** = last four digits of the clipID
    - **clipDate** = date biomass was clipped in the field
    - **ovenInDate / Time** = date and time (24-h format) the sample was placed in the drying oven
    - **ovenOutDate / Time** = date and time the sample was removed from the drying oven
    - **weighDate** = date sample was weighed in the laboratory
    - **herbGroup** = 3-letter code associated with sorted herbaceous functional groups (**Table 8**)
10. Once all weights have been recorded, QA may be performed on a subset of samples (SOP E.2), or return biomass to temporary storage at ambient conditions. Samples in temporary storage can then be weighed for QA as time permits.

E.2 Data Verification and QA

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

1. Per unique boutNumber for each site, select 10% of dried, previously weighed samples for re-weighing.
    - If QA weighing does not occur within several hours of the initial weighing, return the selected samples to the drying oven for 24 h prior to QA weighing. In humid environments, samples will pick up moisuremoisture from the atmosphere (especially bryophytes).

2. Record QA weight data to the nearest 0.01 g in the **qaDryMass** field of the "Lab Weighing" datasheet.
3. Return plant material to temporary storage until all data have been successfully entered to the NEON database. Once data have been successfully entered and QC checked at NEON headquarters, samples may be discarded.
    - ***Exception***: Do NOT discard Herbaceous Biomass for Biogeochemistry samples collected according to SOP F. These samples must be ground and shipped to archive and analytical facilities.

SOP F Herbaceous Biomass Sampling for Biogeochemistry

The herbaceous biomass sampling strategy for biogeochemistry is broadly similar to that employed for ungrazed sites described in SOP B. However, there are important high-level differences related to the spatial sampling requirements (below), and temporal sampling requirements (Section 4). In addition, unlike standard herbaceous biomass samples, samples generated via this procedure are ground and subsampled for chemical analysis once they are dry, and subsampling metadata must be tracked so that data returned from external analytical laboratories may be linked back to the samples.

Spatial sampling requirements

- 20 plots total will be sampled per bout. 4 of these plots are Tower plots, and the remaining 16 are either Distributed or Gradient plots. Randomly selected plots are provided by NEON Science Operations.
- The 4 Tower plots that support Herbaceous Biogeochemistry sampling are the same 4 plots used for biogeochemistry soil core sampling. ***If grazing exclosures are employed at a site, exclosures must NOT be deployed in these 4 Tower plots.*** Ferrous metals may interfere with soil biogeochemistry measurements.
- One Clip Strip is harvested per 20m x 20m plot or subplot. How the location of the Clip Strip is selected depends on the visually-estimated % aerial cover that is herbaceous vegetation in the plot:
    - *Herbaceous % aerial cover > 50% of the plot*:
        - Randomly select clip harvest locations using the plot-specific Clip List.
        - Reject potential clip harvest locations if they are under an overstory canopy.
        - Record which Clip Strip was chosen for biogeochemistry sampling on the Clip List.
    - *Herbaceous aerial % cover is < 50% AND > 25% of the plot*: Select clip harvest location using a targeted non-random procedure.
        1. Assign a number to each continuous “patch” of herbaceous vegetation.
        2. Randomly select a patch for sampling, using either a coin flip, or random number list.
        3. Find the center of the patch to the best of your ability, and then use a map of the clip cells (APPENDIX F) to select a Clip Strip that is as close as possible to the center of the patch.
        4. Record which Clip Strip was chosen for biogeochemistry sampling on the Clip List.
    - *Herbaceous aerial % cover is < 25% of the plot*: Do not perform herbaceous biomass sampling for biogeochemistry in the plot, and do NOT substitute another plot. Only canopy sampling for biogeochemistry will be performed in the plot (RD[11]).

F.1 Field Sampling for Herbaceous Biogeochemistry

1. Navigate to the plotID to be sampled (using the GPS if necessary).

2. Using the Spatial Sampling Requirements provided above, determine whether:
   - The Clip Strip location should be selected randomly (herbaceous % cover ≥ 50%)
   - Non-randomly (herbaceous % cover is < 50% but > 25%); or
   - Whether Herbaceous Biogeochemistry sampling should not be performed in the plot (herbaceous % cover < 25%)
3. Use the Clip List to locate the desired target coordinates for the selected Clip Strip, and record code = '1' in the 'status' column of the Clip List.
   - If selecting Clip Strips randomly, do NOT record code = '0' for any Clip Strips rejected because they lie underneath a canopy. These Clip Strips may still be used for regular herbaceous biomass sampling (SOP B and SOP C), and should therefore not be permanently rejected.
4. Locate the relative X,Y-coordinates of the Clip Strip SW corner within the plot or subplot using the method outlined in steps (3) – (5) in SOP B.1.

5. If the Clip Strip location was chosen randomly because the plot is > 50% cover herbaceous vegetation, assess according to the steps below, otherwise skip to step (6):

   - Assess whether the Clip Strip location is broadly representative of the herbaceous vegetation in the plot that is visible from the air, and accept or reject the location (see **Figure 3** flow-chart).
   - A Clip Strip may also be rejected if obstacles, disturbances, and/or irregularities are encountered, particularly those that prevent delineation of the Clip Strip. These may include trees, large rocks, ant nests, etc.
   - If the Clip Strip is rejected, record why in the 'status' column of the Clip List using the codes in **Table 7**, and proceed to the next Clip Strip on the Clip List.

UNCOMMON SPECIES

Random AND non-randomly located Clip Strips should also be rejected if clipping a particular plant specimen in the strip would influence plot-level diversity. That is, the plant in question exists nowhere else in the plot or subplot.

6. Delineate the accepted Clip Strip for harvesting:
   a. Using one of the pre-marked string and stake sets, line up one of the marks with the pin flag, and push one stake into the ground.

b. Stretch the string and second stake from the South to the North end of the Clip Strip, using the compass or the TruPulse to orient the string in a North/South direction.

c. Keep the compass or TruPulse at least 50 cm from non-aluminum metal plot markers, eyeglasses, wristwatches, tent stakes, etc.

d. Use a ruler to place the second string-and-stake set 10 cm to the right (east) of the first set. Check that the distance between the two strings is exactly 10 cm at both ends of the Clip Strip.

e. The two sets of marks on the two string-and-stake sets now clearly delineate a 0.1 m x 2 m area for clip-harvesting.

7. Using a Sharpie, label kraft paper bags (8# or 25#, depending on the amount of herbaceous vegetation) with the information below. If vegetation is wet, cloth bags may be used, and labels may be fashioned from pieces of waterproof paper placed inside the bags.
   - **boutNumber**; use *XX* format, where XX is the bout number assigned by Field Operations, e.g. '01'. Values of **boutNumber** reset every year.
   - **date**; use *YYYYMMDD* format
   - **clipID**; as provided in Clip List; use "*plotID_clipCellNumber"* format, e.g. CPER_001_0126
   - **bagCount**; the total # of bags within a given clipID.
8. Clip all herbaceous aboveground biomass rooted within the Clip Strip, and place into the labeled bags as you go. See Table 9 in SOP B.2 for common points of confusion and guidelines for problem plants.
   - ***DO NOT sort biomass into functional groups when clipping Herbaceous Biomass for Biogeochemistry.***
   - Do **NOT** clip herbaceous vegetation that passes through/leans over the Clip Strip but is not rooted in the strip (this includes non-woody vines; WST group is an exception, see bullet below).
   - **DO** clip all herbaceous biomass of plants rooted within the strip > 1-2 cm in height. That is, include leaves in the harvest that exit the strip but originate from stems rooted within the strip.
   - **DO** clip leaves and twigs of woody stemmed plants with ddh < 1cm that are produced in the current year AND originate from nodes that fall within the clip strip. It is not necessary that the individuals are rooted in the clip strip as long as the most recent node from which they originated falls within the strip.
9. When finished clipping, place all bags into a cooler with cold packs as soon as possible.
10. Record required data in the standard "Field Datasheet" (RD[05]):

- **plotID**
- **subplotID** – either 31, 21, 23, 39, or 41
- **clipCellNumber** – these are the right-most digits after the last '_' in the clipID
- **clipDate**
- **time** – the time clipped biomass was placed in the cooler
- **exclosure** – this should always be 'N' for this SOP
- **bagCount** – the total number of bags from the Clip Strip

11. Continue to the next required plot.

F.2 Laboratory Processing for Herbaceous Biogeochemistry

1. Samples processed for Herbaceous Biogeochemistry are dried and weighed according to the standard procedure described in SOP E, with two notable exceptions:
    - Use the "Biogeochemistry Lab" datasheet (RD[05]).
    - Herbaceous biomass from a given Clip Strip remains pooled, and is NOT sorted.
2. Based on the mass of each sample, decide whether enough sample is available for chemical analysis only, or whether sufficient mass is available for both chemical analysis and archive.
    - If total sample mass is < 250 mg:
        - No further processing is required. The sample will not be sent for chemical analysis or archive.
        - Record **subSampleType** = 'NA'
    - If total sample mass is ≥ 250 mg but < 750 mg:
        - The sample will be subsampled for chemical analysis only.
        - Record **subSampleType** = 'C' on the Biogeochemistry Lab datasheet
    - If total sample mass is ≥ 750 mg:
        - The sample will be subsampled for chemical analysis and archive.
        - Record **subSampleType** = 'C, A' on the Biogeochemistry Lab datasheet
3. Enter the completed Biogeochemistry Lab datasheet into the webUI.
    - The webUI will generate a .csv as part of the data entry process.
    - Review the .csv for data entry errors
    - Save a local working copy of the .csv file.
4. Copy the **subsampleID** information automatically generated in the .csv file into a label printing template. Label 20 mL plastic or glass scint vials with pre-printed labels.
    - Do not create pre-printed labels when **subSampleType** = 'NA'
5. Grind oven-dried biomass with a mill (0.85 mm, mesh size = 20).
    - Grind the entire sample if the total mass is ≤ 100 g
    - If the total mass is > 100 g, mix the sample well by hand, in a bag or tray depending on total sample volume, and select approximately 100 g for grinding.
    - The portion of the sample selected for grinding **must be representative**.
6. Mix the ground sample thoroughly with a spatula, and use an appropriately sized splitter or microsplitter to generate two representative subsamples to fill two pre-labeled 20 mL scint vials.
    - For samples with < 1 g total mass, split the sample with a spatula instead of a microsplitter, and use the information in step (2) to allocate material to the 'C' and (possibly) 'A' subsamples.
    - If there is >> 1 g of sample, but not enough to fill two 20 mL scint vials, split evenly between two vials.

BEST PRACTICE TIPS

- 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.

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

F.3 Equipment Maintenance

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

SOP G Data Entry and Verification

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

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

Before entering data, all personnel ***MUST*** read RD[13] for complete instructions regarding manual data transcription. Prior to entering data via a web user interface (webUI), each technician shall enter a plot (or subplot) of data from one bout into the protocol-specific webUI housed on the Training portal, as described in RD[13].

Protocol-specific instructions and the associated data ingest workbook for entering Herbaceous Biomass data can be found on the NEON intranet in the "FSU-FOPS" folder. Be sure to enter data for all plots within a bout. If an entire bout was missed, no data need be entered. There should be three data ingest tables in the CYI Herbaceous Biomass webUI associated with herbaceous biomass and productivity clip harvest data:

- **Perbout**: Metadata describing individual sampling events on a per clipID per plotID per boutNumber basis.
- **Massdata**: Oven-dried biomass data for each herbCode per clipID per eventID, as well as weighing QA data.
- **Biogeochemistry**: Oven-dried biomass data for pooled biomass (not sorted) per clipID per eventID, as well as a record of whether subsamples were produced for archive and chemical analysis.

G.1 Field Datasheets

1. Transcribe data from the Herbaceous Biomass Field Datasheets to the "perbout" ingest table.
   - Consult the herbaceous biomass and productivity data ingest document (RD[09]) to determine appropriate values and formats for each field in the ingest table.
2. If a representative clipID contained no herbaceous biomass (noted as 'targetTaxaPresent = N' in the **remarks** field of the Field Datasheet), enter the following values in the "perbout" ingest table:
   - **targetTaxaPresent** = 'N'
3. Update permanent digital versions of the "Clip Strip coordinate" lists with "date" and "status" data recorded in the field.

G.2 Lab Datasheets

Lab Drying QC data are not transcribed for ingest into the NEON database.

- Transcribe data from the “Lab Weighing” datasheet to the “massdata” ingest table.
    - Consult the herbaceous biomass and productivity data ingest document (RD[09]) for additional information about the appropriate values and formats for each field.
    - If a representative clipID contained no herbaceous biomass in one or more herbGroups (noted in the ‘remarks’ column of the Field Datasheet), enter the following in the “massdata” ingest table:
        - Enter the appropriate **herbGroup** code
        - Enter ‘0’ in the **dryMass** field
- Transcribe data from the “Biogeochemistry Lab” datasheet into the “biogeochemistry” ingest table.
    - The **herbGroup** code is always ‘ALL’ (pooled)

SOP H Sample Shipment

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

H.1 Timelines

Dried, ground samples may be stored indefinitely before shipping.

H.2 Storage / Shipping Conditions

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

H.3 Grouping/Splitting Samples

Samples originating from the same clipID should be grouped together. If there is more than one clipID per plotID, samples originating from the same plotID should be grouped together if possible.

H.4 Procedure

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

H.5 Laboratory Contact Information and Shipping/Receipt Days

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

8 REFERENCES

The Nutrient Network Experimental Protocol page (http://www.nutnet.umn.edu/exp_protocol). Accessed 2013-09-19.

APPENDIX A DATASHEETS

Table 11. Datasheets associated with this protocol

NEON Doc. #	Title
NEON.DOC.001574	Datasheets for TOS Protocol and Procedure: Measurement of Herbaceous Biomass

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

APPENDIX B QUICK REFERENCES

B.1 Delineating the Clip Harvest Strip

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 the Clip List. Be sure to check if Belowground Biomass Core sampling has already occurred in the current season, and choose Clip Strips accordingly.

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

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

STEP 4 – Locate the offsetNorthing coordinate with TruPulse in HD mode (azimuth 0°), place pin flag.

STEP 5 – Assess suitability of Clip Strip. Relocate 15 cm west or east OR reject if not suitable.

STEP 6 – Remove and relocate exclosure to next suitable random location, if applicable.

DELINEATE 0.1 M X 2 M CLIP STRIP

STEP 1 – Place north-south oriented string-and-stake set on west side of Clip Strip. Use TruPulse to orient string.

STEP 2 – Place second string-and-stake set EXACTLY 10 cm to the east of first set.

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

CHARACTERIZATION

STEP 1 – If cactus are present, remove and discard only those cactus pads that would physically prevent clipping herbaceous biomass.

B.2 Clipping and Sorting

STEP 1 – Label 8# kraft paper bags (lunchbag size), use multiple bags as needed.

STEP 2 – Clip biomass ROOTED in Clip Strip area, sorting vegetation into coded bags as you go.

STEP 3 – Record on Field Datasheet the total number of bags harvested per clip strip.

STEP 4 – Place all 8# bags from single Clip Strip into one 25# bag.

STEP 5 – Store bag in cooler with cold packs (or sealed ice) for transport back to lab.

STEP 6 – Transfer clip bags to 4° C refrigerator in domain lab (if possible).

STEP 7 – Check sorting at end of day or next morning in lab.

STEP 8 – Confer with lead plant technician to check that all biomass is correctly sorted.

STEP 9 – Place clipped biomass in a drying oven as soon as possible after clipping and sorting.

STEP 10 – On the appropriate datasheets, record the clipDate and time biomass was placed in the cooler in the field, as well as the ovenInDate and time biomass was placed in the drying oven.

QUALITY DEPENDS ON PROPER:

- Sorting into groups.
- Separation of previous and current years' growth.
- Labeling of all samples.

CLIPPING GUIDELINES

- Only clip biomass ROOTED in the Clip Strip area (with the exception of WST).
- Sort clipped vegetation into appropriate bags as you go.
- Clip as close to the ground as possible (i.e., 1-2 cm above ground).
- DO NOT CLIP crowns of perennial grasses, as this will kill or damage the plant.
- Do not clip ANY cactus unless required to access herbaceous vegetation
- Clip qualifying *Toxicodendron spp.* according to APPENDIX E.

APPENDIX C REMINDERS

Collecting Quality Biomass Data with the Clip Harvest Technique

At the plot: Be sure to…

- ☑ Avoid walking on targeted clip area, plant diversity subplots, and area surrounding plot centroid.
- ☑ Assess suitability of potential Clip Strip and accurately delineate.
- ☑ Relocate exclosure to next suitable location, if applicable.

Label Information

- boutNumber
- date
- clipID
- exclosure (Y or N)
- herbCode (3 letter code)
- bagNumber (e.g. 2 of 3)

Functional Group Name	Biomass Code
Bryophtyes (not lichens), with discernable annual growth	BRY
Cool-season graminoids (C3)	CSG
Warm-season graminoids (C4)	WSG
Leguminous forbs	LFB
Non-leguminous forbs	FRB
Woody-stemmed plants, ddh < 1.0 cm	WST
Previous years' old standing dead	OSD
Unsorted herbaceous biomass, potentially containing all functional groups	ALL

- ☑ Fill out 'status' for sampled or rejected clip-harvest locations on Clip List

Clip harvesting: Be sure to…

- ☑ Clip and discard pad-forming cactus only if necessary.
- ☑ Fill in Field Datasheet and check that all bags are accounted for.
- ☑ Store bags in cooler or refrigerator at all times prior to oven drying.
- ☑ Check sorting in lab at end of field day or next morning.
- ☑ Confer with lead plant technician to check biomass sorting.

Using the TruPulse: Pay close attention to…

- ☑ Declination – Is it set for your current location?
- ☑ Selection choices in drop-down menu.
- ☑ Battery charge (replace when low-charge indicated).
- ☑ Transcription of measurements onto data sheet.
- ☑ Metal objects – Keep them at least 2 feet away from instrument when using internal compass.

APPENDIX D SITE-SPECIFIC ESTIMATED DATES FOR ONSET AND CESSATION OF SAMPLING

The dates in the table below are estimated from satellite MODIS-EVI phenology data averaged from 2001-2009, and correspond to the average date after which greenness begins to decrease at each site. By using the average greenness decrease date, we ensure that there is a high probability that all herbaceous biomass has been produced for the current season prior to clipping. However, these dates are only a guide, and 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.

At sites managed for grazing, Distributed plots should be clip-harvested at approximately the same peak biomass date that functional groups are sorted in the Tower plots (see "Additional Sampling Information" field in the table below).

Table 12. Site-specific grazing status and per bout sampling start dates for herbaceous clip-harvest.

Domain Number	Site ID	Grazing Mgmt	Bouts per Growing Season	Start Date (dd/mm)	Additional Sampling Information
01	BART	No	1	08/08	
	BURL	No	1	08/08	
	HARV	No	1	08/08	
02	BLAN	No	1	07/29	
	SCBI	No	1	08/08	
	SERC	No	1	08/08	
03	DSNY	No	1	07/09	Tower plot parcel may be managed for grazing. Candidate site for continuous growth/decomp SOP.
	JERC	No	1	08/08	
	OSBS	No	1	07/09	
04	GUAN	No	2	Bout1: 5/30 Bout2: 9/12	
	LAJA				
05	STEI	No	1	08/03	
	TREE	No	1	08/03	
	UNDE	No	1	08/03	
06	KONA				
	KONZ	Yes	Every 4 wks	03/31	End sampling by 10/27, or when livestock removed. Sort clip to functional group on first bout occurring after 07/29.
	KUFS				
07	GRSM	No	1	08/03	
	MLBS	No	1	08/08	
	ORNL	No	1	07/29	
08	CHOC	No	1	07/19	

Domain Number	Site ID	Grazing Mgmt	Bouts per Growing Season	Start Date (dd/mm)	Additional Sampling Information
	DELA	No	1	07/24	
	TALL	No	1	07/14	
09	DCFS	Yes	Every 4 wks	04/30	End sampling by 10/17, or when livestock removed. Sort clip to functional group on first bout occurring after 07/24.
	NOGP	Yes	Every 4 wks	04/24	End sampling by 10/17, or when livestock removed. Sort clip to functional group on first bout occurring after 07/19.
	WOOD	No	1	07/29	Intermittent, low-level grazing. Domain Mgr must communicate with site host to determine whether Tower plots are grazed in a given year.
10	CPER	Yes	Every 4 wks	03/31	End sampling by 12/16, or when livestock removed. Sort clip to functional group on first bout occurring after 07/29.
	RMNP	No	1	06/29	
	STER	No	1 bout per crop, per growing season	2-4 wks before crop harvest	Domain Mgr communicates with site host to determine crop harvest dates.
11	CLBJ				
	OAES	Yes	2	Bout1: 6/9 Bout2: 10/17	Cows typically in one pasture early Sept through winter, light grazing pressure. For affected plots only: • Deploy exclosures prior to stocking (late August) • Sample every 4 wks until 11/11, or until livestock removed. • Only sort to functional group for 10/17 bout
	TBD				
12	BOZE				
	PARA				
	YELL	No	1	07/09	
13	MOAB	Yes	Every 4 wks	03/26	End sampling by 10/27, or when livestock removed. Sort clip to functional group on first bout occurring after 08/13.

Domain Number	Site ID	Grazing Mgmt	Bouts per Growing Season	Start Date (dd/mm)	Additional Sampling Information
	NIWO	No	1	08/08	
	TBD				
14	JORN	No	1	09/02	Grazing not anticipated in Tower plots, but may occur. Domain Mgr communicates with site host to determine if Tower plots grazed.
	SRER	No	1	08/28	Grazing not anticipated in Tower plots, but may occur. Domain Mgr communicates with site host to determine if Tower plots grazed.
	TBD				
15	ONAQ	Yes	Every 4 wks	03/16	End sampling by 10/07, or when livestock removed. Sort clip to functional group on first bout occurring after 06/19.
	RBUT				
	TBD				
16	ABBY				
	THAY				
	WREF	No	1	07/29	
17	SJER	Yes	Every 4 wks	09/27	End sampling by 06/04 of next calendar year (Mediterranean growing season). Sort clip to functional group on first bout occurring after 04/05.
	SOAP	No	1	07/04	
	TEAK	No	1	07/24	
18	BARO	No	1	07/29	
	TOOL	No	1	07/24	
	TBD				
19	DEJU	No	1	07/29	
	HEAL	No	1	07/29	
	POKE	No	1	07/24	
20	OLAA				Candidate site for continuous growth/decomp SOP.
	PUFO				Candidate site for continuous growth/decomp SOP.
	PUGR				Candidate site for continuous growth/decomp SOP.

APPENDIX E CLIP-HARVESTING *TOXICODENDRON* SPECIES

E.1 Equipment and Materials

Table 13. Equipment and materials required for a team of two to minimize exposure to toxic oils from *Toxicodendron spp.* that should be clip-harvested.

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.
Labeled hand clippers, dedicated to *Toxicodendron* clipping	1	Same item type as indicated in equipment lists	Minimize spread of oils to other equipment.
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 clippers after use.

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

The following are best-practice techniques for minimizing exposure to toxic oil during clip-harvest of Toxicodendron species.

1. Prior to field work:
   a. Count out bags for storing and drying ONLY Toxicodendron biomass. Don't mix Toxicodendron biomass with any other biomass.
   b. Pre-weigh (to nearest 0.01 g) and label each paper bag that will be used for storing and drying clip-harvested *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.
2. To handle and clip *Toxicodendron* biomass in the field:

- Wear cotton gloves and dispose after single use. Toxic oils can pass through nitrile or latex gloves.
- Use a pair of clippers dedicated solely to clipping *Toxicodendron spp*, and clean with Tecnu (or equivalent) after each use. Store separately from other clippers to prevent accidental contact.
- Bring a clean, new plastic bag to the field for storing and transporting contaminated gloves and clippers after use.
- Wear a thin outer layer of disposable PPE over clothes and shoes.

3. After field work is complete, wash clothing acccording 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 in the laboratory:
   - 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 of bag + dried biomass to nearest 0.01 g, and also record weight of individual empty bag (to 0.01 g) on data sheets. Dried *Toxicodendron* biomass should never leave the bag.
     - Use a spreadsheet to calculate the mass of *Toxicodendron* by difference. The Herbaceous Biomass webUI will only accept one 'dryMass' value, so you must subtract out the weight of the bag prior to data entry.
     - Use herbCode = 'WST' for *Toxicodendron* biomass.
   - After weighing, dispose of all *Toxicodendron* biomass bags.
     - *Toxicodendron* tissue will not be specimen mounted, or processed for Herbaceous Biogeochemistry (i.e., archived and sent for external chemical analysis).

APPENDIX F CLIPCELLNUMBER COORDINATES AND MAPS

The Herbaceous Biomass Sampling for Biogeochemistry procedure (SOP F) requires locating Clip Strips within "patches" of herbaceous vegetation when the % cover of herbaceous vegetation over the entire plot is ≥ 25% AND < 50%. To identify the location of clip harvests within herbaceous "patches," first use the appropriate map in Appendix F.1 to determine which clipCellNumber should be sampled, and then use **Table 14** in Appendix F.2 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.

F.1 Maps of clipCellNumber by subplotID

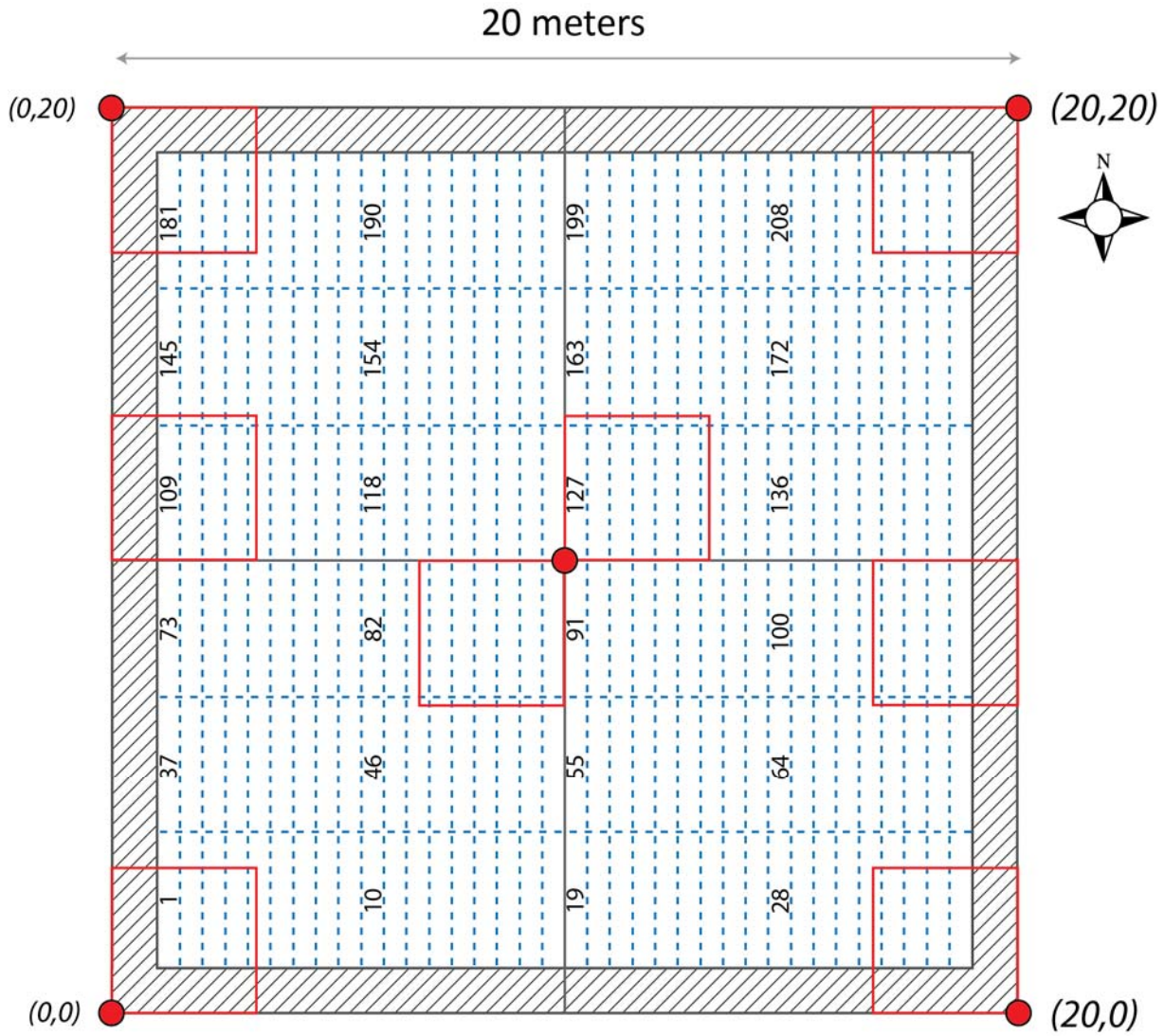

Figure 5. Map of clipCellNumbers in a 20m x 20m base plot (subplotID = 31 in provided Clip Lists). Red squares indicate nested subplots used for diversity sampling; clip cells that significantly overlap red squares are not used for clip sampling. However, clip cells with minimal overlap (e.g., 48-54, 68-72, 145-149) do support clip harvest sampling.

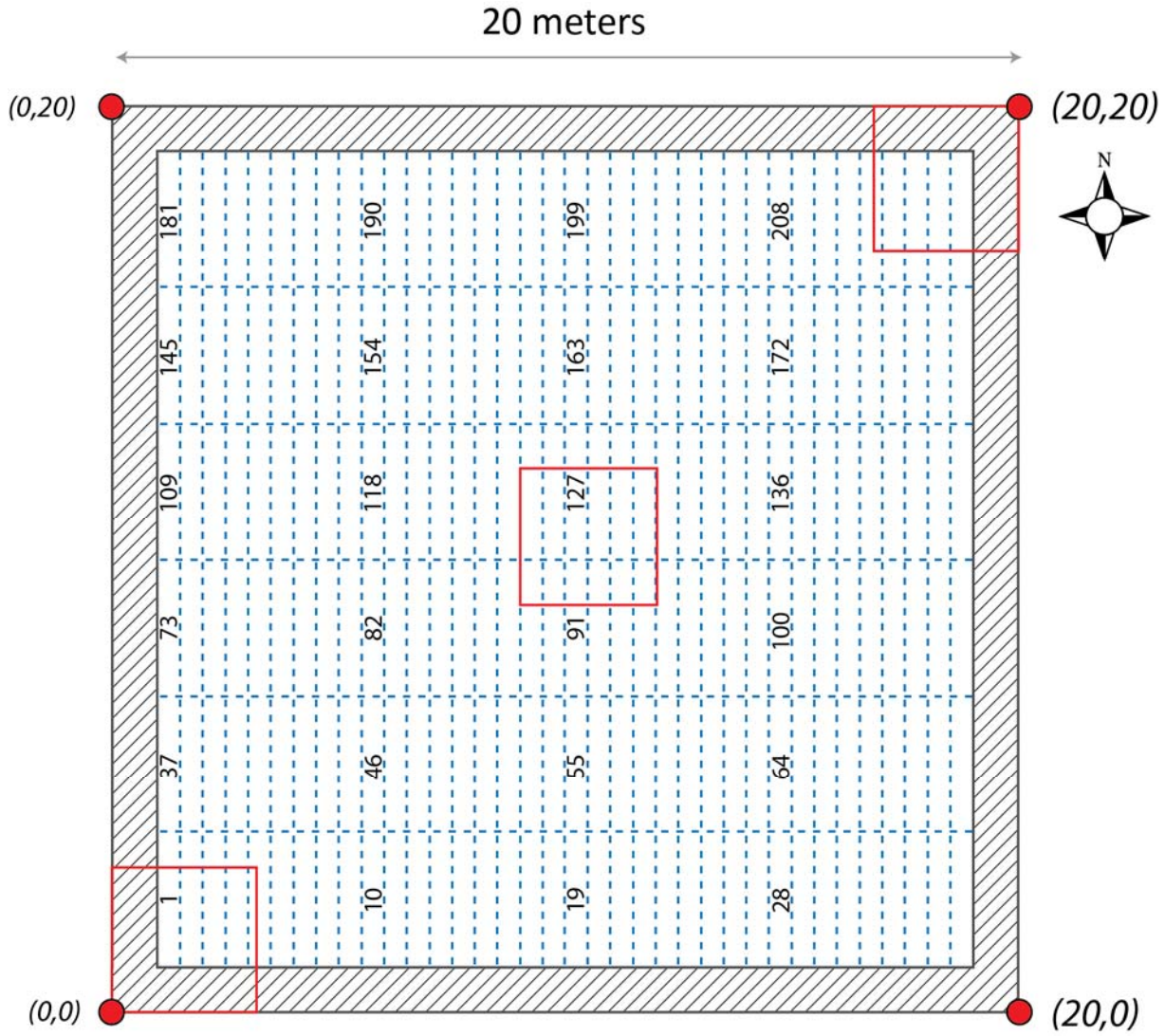

Figure 6. Map of clipCellNumbers for **subplotID = 21** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for clip sampling.

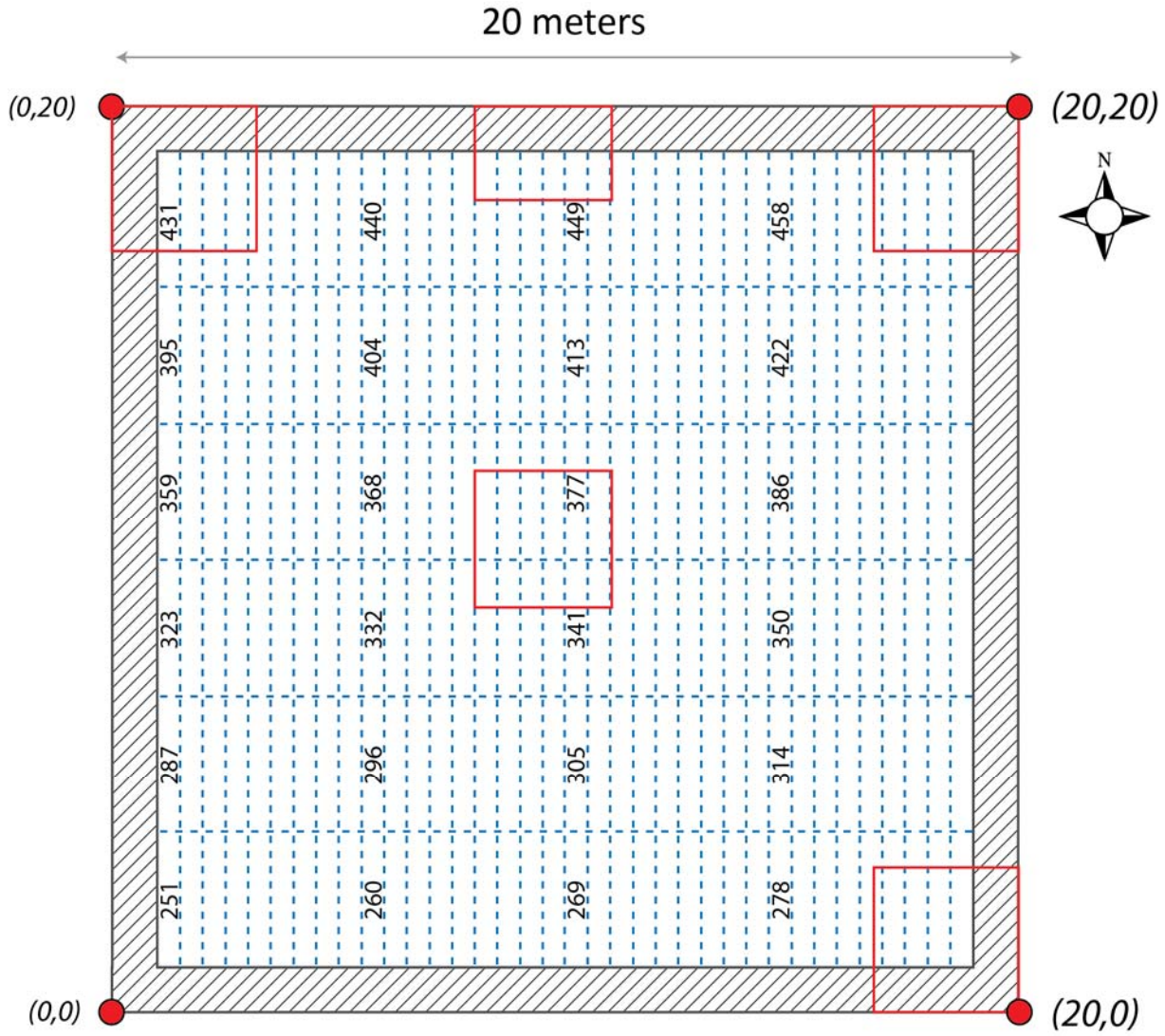

Figure 7. Map of clipCellNumbers for **subplotID = 23** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for clip sampling.

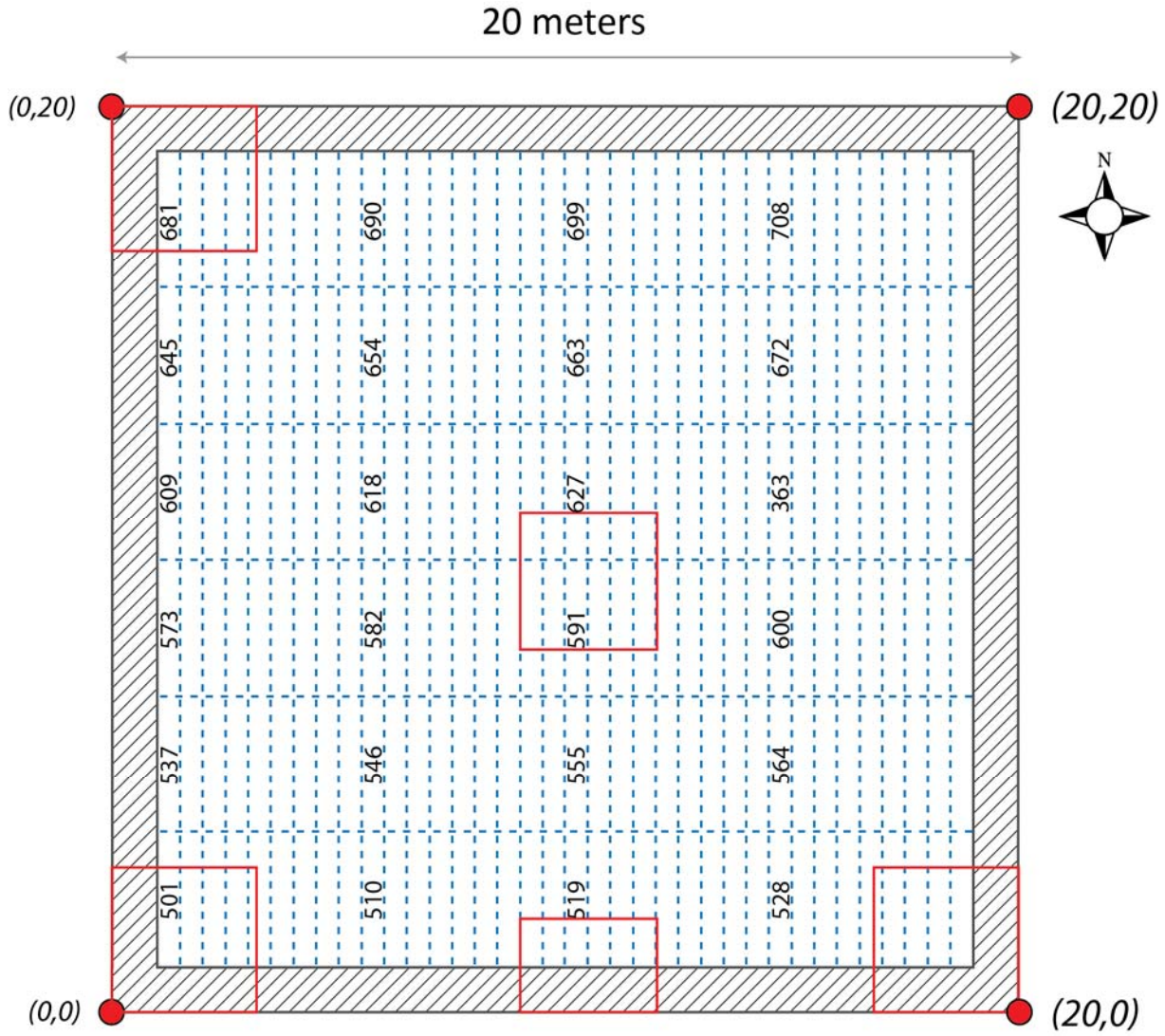

Figure 8. Map of clipCellNumbers for **subplotID = 39** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for clip sampling.

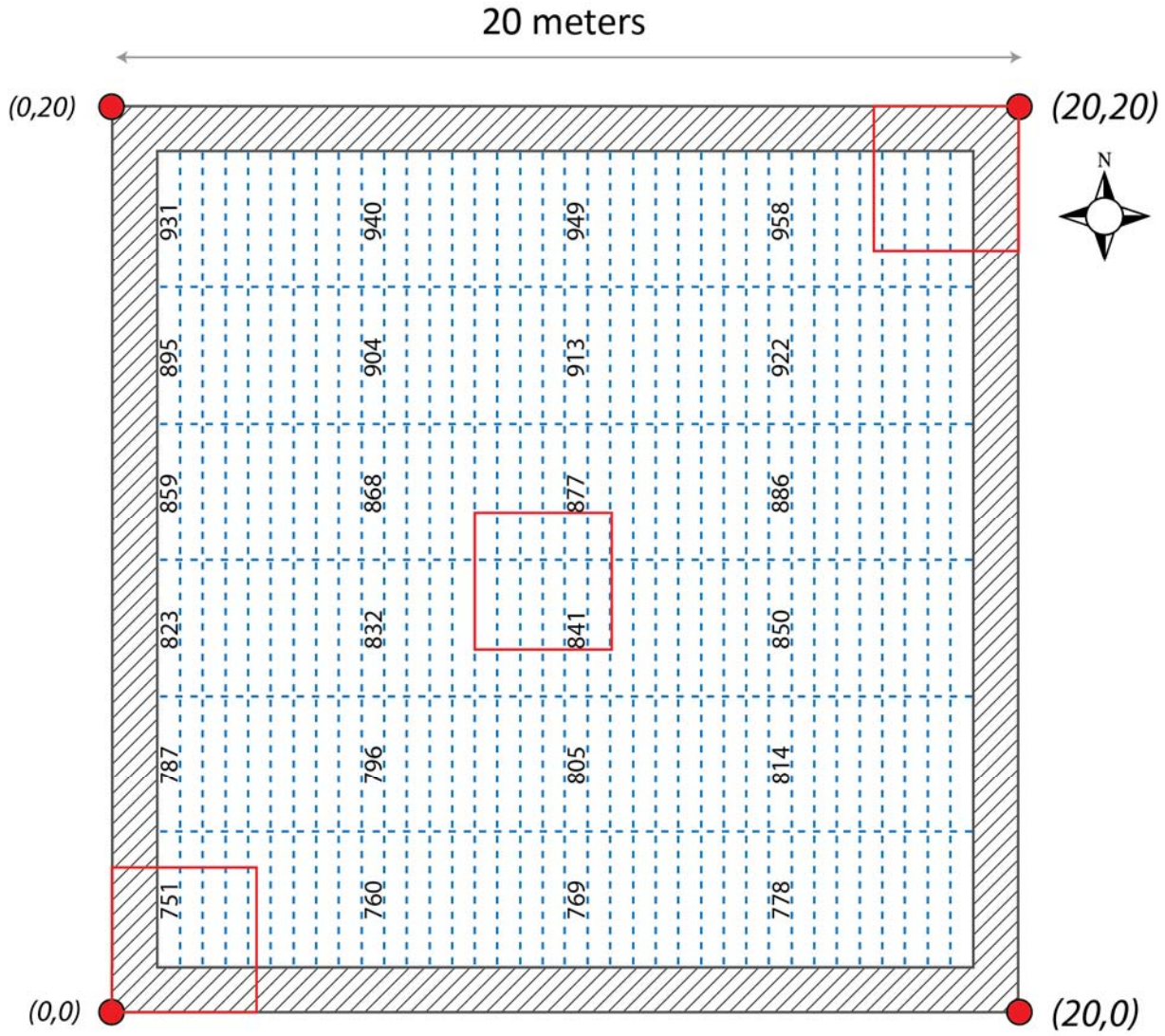

Figure 9. Map of clipCellNumbers for **subplotID = 41** in a 40m x 40m Tower base plot. Clip cells that overlap nested subplots indicated by red squares are not used for clip sampling.

F.2 Coordinates for clipCellNumbers by subplotID

Table 14. List of clipCellNumbers by subplotID and associated easting and northing coordinates. Coordinates correspond to the SW corner of a 0.1m x 2m Clip Strip, and indicate the distance in meters relative to the SW corner of the plot (subplotID = 31) or subplot (subplotID = 21, 23, 39, 41).

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

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

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

clipCellNumber subplotID = 31	clipCellNumber subplotID = 21	clipCellNumber subplotID = 23	clipCellNumber subplotID = 39	clipCellNumber subplotID = 41	easting offset	northing offset
122	122	372	622	872	7.7	10.5
123	123	373	623	873	8.2	10.5
124	124	374	624	874	8.7	10.5
125	125	375	625	875	9.2	10.5
126	126	376	626	876	9.7	10.5
127	127	377	627	877	10.2	10.5
128	128	378	628	878	10.7	10.5
129	129	379	629	879	11.2	10.5
130	130	380	630	880	11.7	10.5
131	131	381	631	881	12.2	10.5
132	132	382	632	882	12.7	10.5
133	133	383	633	883	13.2	10.5
134	134	384	634	884	13.7	10.5
135	135	385	635	885	14.2	10.5
136	136	386	636	886	14.7	10.5
137	137	387	637	887	15.2	10.5
138	138	388	638	888	15.7	10.5
139	139	389	639	889	16.2	10.5
140	140	390	640	890	16.7	10.5
141	141	391	641	891	17.2	10.5
142	142	392	642	892	17.7	10.5
143	143	393	643	893	18.2	10.5
144	144	394	644	894	18.7	10.5
145	145	395	645	895	1.2	13.5
146	146	396	646	896	1.7	13.5
147	147	397	647	897	2.2	13.5
148	148	398	648	898	2.7	13.5
149	149	399	649	899	3.2	13.5
150	150	400	650	900	3.7	13.5
151	151	401	651	901	4.2	13.5
152	152	402	652	902	4.7	13.5
153	153	403	653	903	5.2	13.5
154	154	404	654	904	5.7	13.5
155	155	405	655	905	6.2	13.5
156	156	406	656	906	6.7	13.5
157	157	407	657	907	7.2	13.5
158	158	408	658	908	7.7	13.5
159	159	409	659	909	8.2	13.5
160	160	410	660	910	8.7	13.5
161	161	411	661	911	9.2	13.5
162	162	412	662	912	9.7	13.5
163	163	413	663	913	10.2	13.5

clipCellNumber subplotID = 31	clipCellNumber subplotID = 21	clipCellNumber subplotID = 23	clipCellNumber subplotID = 39	clipCellNumber subplotID = 41	easting offset	northing offset
164	164	414	664	914	10.7	13.5
165	165	415	665	915	11.2	13.5
166	166	416	666	916	11.7	13.5
167	167	417	667	917	12.2	13.5
168	168	418	668	918	12.7	13.5
169	169	419	669	919	13.2	13.5
170	170	420	670	920	13.7	13.5
171	171	421	671	921	14.2	13.5
172	172	422	672	922	14.7	13.5
173	173	423	673	923	15.2	13.5
174	174	424	674	924	15.7	13.5
175	175	425	675	925	16.2	13.5
176	176	426	676	926	16.7	13.5
177	177	427	677	927	17.2	13.5
178	178	428	678	928	17.7	13.5
179	179	429	679	929	18.2	13.5
180	180	430	680	930	18.7	13.5
181	181	431	681	931	1.2	16.5
182	182	432	682	932	1.7	16.5
183	183	433	683	933	2.2	16.5
184	184	434	684	934	2.7	16.5
185	185	435	685	935	3.2	16.5
186	186	436	686	936	3.7	16.5
187	187	437	687	937	4.2	16.5
188	188	438	688	938	4.7	16.5
189	189	439	689	939	5.2	16.5
190	190	440	690	940	5.7	16.5
191	191	441	691	941	6.2	16.5
192	192	442	692	942	6.7	16.5
193	193	443	693	943	7.2	16.5
194	194	444	694	944	7.7	16.5
195	195	445	695	945	8.2	16.5
196	196	446	696	946	8.7	16.5
197	197	447	697	947	9.2	16.5
198	198	448	698	948	9.7	16.5
199	199	449	699	949	10.2	16.5
200	200	450	700	950	10.7	16.5
201	201	451	701	951	11.2	16.5
202	202	452	702	952	11.7	16.5
203	203	453	703	953	12.2	16.5
204	204	454	704	954	12.7	16.5
205	205	455	705	955	13.2	16.5

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