FIELD PROTOCOL: MEASUREMENT OF HERBACEOUS STRUCTURE AND BIOMASS

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A_DRAFT	03/25/2011	ECO-00280	Initial Draft Release
B_DRAFT	01/13/2013	ECO-01140	Updates from 2013. Will be finalized in next rev.

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1 DESCRIPTION

1.1 Purpose

The primary purpose of this document is to provide a change-controlled version of Observatory protocols and procedures. This document provides the content for training and field-based materials for NEON staff and contractors. 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.

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

1.2 Scope

This document relates the tasks for a specific field sampling or laboratory processing activity and directly associated activities and safety practices. This document does not describe:

- general safety practices
- site-specific safety practices
- general equipment maintenance

It does identify procedure-specific safety hazards and associated safety requirements such as safe handling of small mammals or safe use of required chemicals and reagents.

1.3 Acknowledgements

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 information that shall be applied in the current document. Examples are higher level requirements documents, standards, rules and regulations.

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

2.2 Reference Documents

Reference documents contain information complementing, explaining, detailing, or otherwise supporting the information included in the current document.

RD [01]	NEON.DOC.000008	NEON Acronym List
RD [02]	NEON.DOC.000243	NEON Glossary of Terms
RD [03]	NEON.DOC.000914	NEON Science Design: Plant Biomass and Productivity
RD [04]	NEON.DOC.005003	NEON Scientific Data Products Catalog
RD [05]	NEON.DOC.014051	Field Audit Plan
RD [06]	NEON.DOC.000824	Data and Data Product Quality Assurance and Control Plan
RD [07]	NEON.DOC.005005	Level 0 Data Product Catalog
RD [08]	NEON.DOC.000987	Woody Vegetation Structure Protocol

2.3 Acronyms

LAI	Leaf Area Index
NEE	Net Ecosystem Exchange
NEP	Net Ecosystem Productivity
NPP	Net Primary Productivity

2.4 Verb Convention

“Shall” is used whenever a statement expresses a convention that is binding. The verbs “should” and “may” express non-mandatory provisions. “Will” is used to express a declaration of purpose on the part of the design activity.

2.5 Definitions

A **protocol** is a formal summary description of a procedure and its related rationale, and includes information on knowledge and resources needed to implement the procedure. A procedure is a set of prescribed actions that must take place to achieve a certain result, and can also be called a method. It differs from a science design in that science designs provide a more complete description of the rationale for selecting specific protocols. It differs from a training manual in that training manuals provide materials in support of skills acquisition in the topic areas including information on how to best train staff rather than detailing only the steps of the procedure.

3 BACKGROUND AND OBJECTIVES

3.1 Background

Herbaceous vegetation is operationally defined in this protocol as non-woody plants (i.e. grasses, sedges, forbs, and some bryophytes and low-stature vines), as well as woody-stemmed plants with height < 0.5 meters at the time of sampling. The net primary productivity (NPP) of this plant group dominates the total NPP of grassland sites, such as the NEON D10 CPER site, 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, or where grazing pressure on herbaceous plants from wild grazers is significant, it is standard practice to use small, portable grazing exclosures to estimate the productivity that is consumed by grazing. NEON will employ a standard approach where clip-harvests are performed with paired grazed/exclosed areas per plot.

3.2 NEON Science Requirements

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.

3.3 NEON Data Products

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[04]).

4 PROTOCOL

Herbaceous vegetation structure measurements and biomass clip-harvests occur in both Distributed and Tower plots. An excess number of potential clip-harvest locations within a particular plot are randomly determined *a priori* by NEON Science Operations, with the knowledge that not all potential locations will be suitable for clip-harvesting – i.e. there may be obstacles such as rocks, trees, ant nests, etc. at any given location that will prevent carrying out a clip-harvest. 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 biomass plot, harvest strips are moved each year to minimize effects of harvest on subsequent biomass data. Additional harvests are required if grazing exclosures are employed at the site, and instructions for utilizing exclosures are integrated into the field standard operating procedure.

Once field work is complete at the plot, technicians must keep harvested biomass cold until sort checking is performed in the laboratory. Best practice is to place clipped biomass into a cooler containing -20˚ C cold packs, or into a 4˚C refrigerator. Keeping clipped biomass cold is critical to prevent wilting, so that species' diagnostic features are preserved and laboratory sort-checking can be performed within 24-h of harvest. In the laboratory, 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 oven-dried 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 collecting quality data from this field work.

5 QUALITY ASSURANCE AND CONTROL

The procedures associated with this protocol will be audited according to the Field Audit Plan (RD[05]). 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 (RD[06]).

A combination of hot, cold, and blind checks may be used to ensure that equipment is used properly in the field, that plant species are identified and sorted properly into functional groups, that samples are dried properly in the lab, and that dried samples are processed according to the protocol.

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

Table 1. Decision tree associated with the plant herbaceous structure measurements and biomass clip-harvests, indicating how to respond to unanticipated delays in field or lab work, and the consequences of potential delays.

Delay	Action	Adverse outcome	Outcome for Data Product
Hours	If 1) Delay prevents completion of current clip-harvest strip: a) Ensure all small bags of sorted biomass are labeled; b) place small bags into one larger 25# bag and label; and c) resume harvest of same sub-plot ASAP.	None	None
	If 2) Delay occurs between plots: resume harvest of next clip-harvest strip ASAP.	None	None
1-7 Days	If 1) Delay prevents completion of clip-harvest strip: a) Ensure all small bags of sorted biomass are labeled; b) place small bags into one larger 25# bag and label; c) oven-dry all biomass as per protocol; d) resume harvest of same sub-plot ASAP with new labeled bags; and e) combine dried biomass per functional group for weighing when all biomass is dry.	None	None
	If 2) Delay occurs between clip-harvest strips: resume harvest of next strip ASAP.	None	None
8-13 days or longer	If 1) Delay prevents completion of clip-harvest strip: a) Ensure all small bags of sorted biomass are labeled; b) place	Aboveground biomass harvested per unit area may	Increased error in aboveground biomass and NPP

Delay	Action	Adverse outcome	Outcome for Data Product
	small bags into one larger 25# bag and label; c) oven-dry all biomass as per protocol; d) resume harvest of same clip-harvest strip ASAP with new labeled bags; and e) combine dried biomass per functional group for weighing when all biomass is dry.	change in the field over this length of time.	estimates, particularly in grazed systems.
	If 2) Delay occurs between clip-harvest strips: resume harvest of next strip ASAP.	Aboveground biomass harvested per unit area may change in the field over this length of time.	Increased error in aboveground biomass and NPP estimates, particularly in grazed systems.

For QA/QC of the weighing and data entry portion of the laboratory work, the Field Operations Manager selects 10% of the previously dried, weighed samples for QA/QC per sampling bout. Technicians re-weigh, re-record, and re-enter the data from these samples in the separate "Clip Biomass QA" datasheet (Appendix G).

6 SAFETY

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.

7 PERSONNEL REQUIREMENTS

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

- For some functional groups, ID to species is not required (e.g. non-leguminous forbs).
- For the cool-season and warm-season graminoid functional groups, identification to species is required.

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

Preferably, the technicians sorting biomass are the same technicians who harvested the biomass in the field.

8 TRAINING REQUIREMENTS

All technicians must complete required safety training as defined in the NEON Training Plan (RD[04]). Additionally technicians complete protocol specific training for safety and implementation of protocol as required in Field Operations Job Instruction Training Plan (RD[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 can accurately and quickly identify C3 and C4 graminoids at all sites, as well as identify leguminous and non-leguminous forbs at all sites.

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

9 FIELD STANDARD OPERATING PROCEDURE

Vegetation structure measurements of small-stature plants (i.e. sub-shrubs, forbs, and graminoids), and harvests of these same plants occur within randomly located clip-harvest strips in both Distributed and Tower Plots. Clip-harvest strips are only placed *outside* the nested sub-plot components of the plot that are used for % cover measurements (Figure 1). Following vegetation structure measurements, sub-shrub, forb and graminoid biomass is collected from these harvest strips. Additional strips within matched grazing exclosures are utilized when applicable, and grazing exclosures are only employed in Tower Plots.

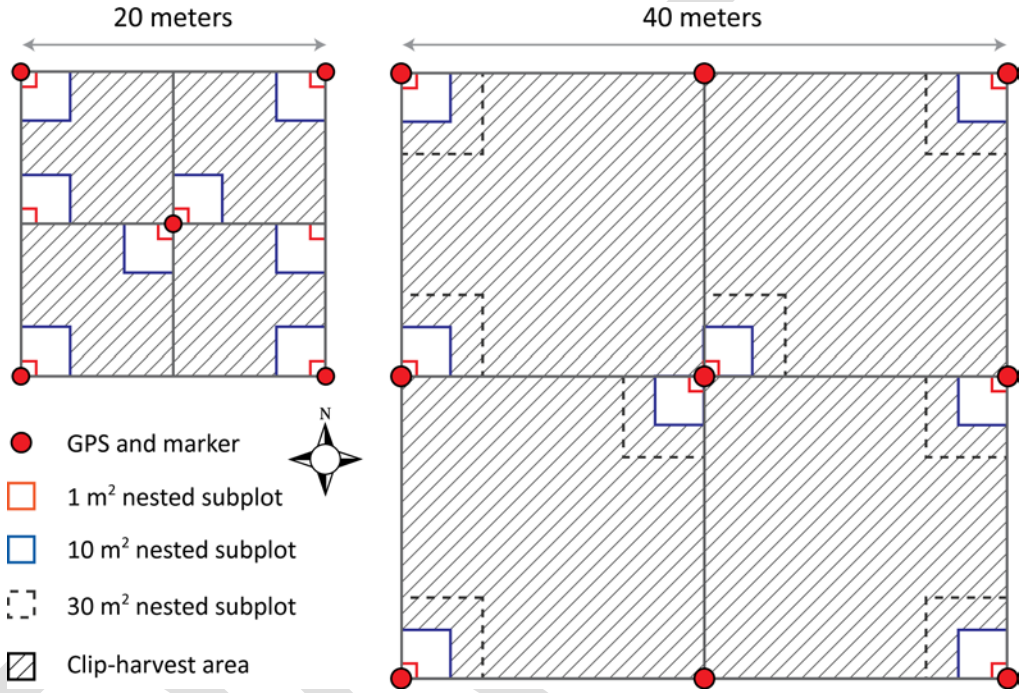

Figure 1. Illustration of two typical NEON plot sizes. Areas used for destructive clip-harvests (hashed fill) explicitly do not overlap those areas of the plot used to estimate % cover by species (white fill). The 30 m^2 subplots are not used for % cover measurements, and may contain clip-harvest strips.

9.1 Sampling Frequency and Timing

9.1.1 Criteria for Determining Sampling Dates

Sampling timing will initially be determined by Science Operations, in consultation with regional experts in order to quantify the number of temporally distinct plant communities that exist per site. For example, the D10 CPER site will require two clip-harvests per season in order to capture both the early cool-season and the late warm-season plant communities. In contrast, the D13 Niwot Ridge site will only require one clip-harvest per growing season, timed to capture peak plant biomass during late summer. Science Operations has provided the sampling onset window for clip-harvest activities per site within Appendix A, and it will be incumbent upon Field Operations to select sampling onset dates.

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.

Herbaceous clip-harvests must be performed within Tower Plots on an annual basis, and sampling these plots is a priority. Distributed Plots should be sampled once every 3 years at a given site (i.e. Distributed Plots are sampled at one site per year per Domain).

9.1.2 Sampling Frequency

Sampling of the herbaceous biomass occurs between 1 and 3 times per growing season. Appendix A lists the number of herbaceous biomass sampling bouts required per site.

9.1.3 Sampling Timing Parameters

A given sampling bout should ideally be concluded within 10-14 days of initiation, 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.

9.2 Equipment and Materials

The equipment listed here is sufficient for a team of 2 to simultaneously measure vegetation structure and carry out the aboveground biomass clip harvests in a plot. It is assumed that field technicians are working in the same plot at any given time.

Table 2. Field equipment required for a team of two people to measure herbaceous vegetation structure and perform a clip-harvest at a plot.

Maximo Item No.	Item Description	Quantity	Habitat-Specific	Special Handling
	Region-specific plant ID book	1	Yes	No
MX100703	GPS unit, pre-loaded with plot locations	1	No	No
MX100322	TruPulse 360R, with current declination entered	1	No	No
MX103238	Reflective surface (3-in bicycle reflector or reflective tape covering back of a notebook)	1	No	No
	Extra battery for TruPulse 360R (CR123A type)	2	No	No
MX100320	Mirror-site compass, declination corrected	1	No	No
MX100722, MX100318	Fiberglass meter tape	1	No	No
MX104363	4”× 5” pin flags, with PVC stakes (PVC is stakes are required to avoid magnetic interference with the TruPulse)	4	No	No
MX103533, MX103524	Pre-marked string and stake sets[1]	2 sets	No	No
MX100326	30 cm length ruler, with cm demarcations	1	No	No

Maximo Item No.	Item Description	Quantity	Habitat-Specific	Special Handling
MX103519, MX103532	Hand clippers	2	No	No
FOPS choice	Work gloves			
MX102977	Paper bags, 8# kraft	50	No	No
MX103232	Paper bags, 25# kraft	8	No	No
MX100314, MX103220	Hammer (only if grazing exclosures are present)	1	Yes	No
	Rite-in-the-Rain QC checklists	As needed	No	No
	Sharpies	2	No	No
	Mechanical pencils	2	No	No
	Large chest-style cooler, with cold packs	1	No	No

[1] Pre-marked string and stake sets are used to temporarily mark plot boundaries while carrying out field work, and require fabrication prior to field work. Each set consists of 2 tent stakes connected by fine nylon cord. Nylon cord is measured, cut, and marked such that the 2 m length of the clip-harvest strip is clearly visible on the cord.

The "Field Clip QC" datasheet is provided as Appendix C to this document, and is intended as an interim data collection medium until the NEON CI team develops a BioPDA application.

9.3 Preparation

1) For first year of field sampling at sites managed for grazing:
    a) Construct a sufficient number of grazing exclosures.
        i) Exclosures are deployed within Tower Plots only, and one exclosure should be made for each 400 m^2 plot/subplot.
    b) Place exclosures within Tower plots/subplots prior to the onset of grazing.
        i) It is incumbent on the Field Operations Manager to communicate with the site host to ascertain when grazing begins for a given growing season.
        ii) For each Tower plot/subplot, place an exclosure over the first suitable clip-strip, and stake the exclosure to the ground.
        iii) Follow steps 2 and 3 in Section 9.4 to locate potential clip-harvest strip locations and assess suitability for clip-harvest sampling.
2) Check to ensure that all consumables required for field work are available for use. Re-order items as necessary prior to field work.
3) Check that pre-marked string and stake sets are fabricated and marked.
    a) Cut a 2.5 m length of ⅛" diameter nylon cord.
    b) Mark the cord clearly with a sharpie approximately 25 cm from each end, such that the center section of the cord measures exactly 2 m long between the markings.
    c) Tie each end to a tent stake (the tent stake should be at least 8" long).

4) At least 1 day prior to field work:

   a) Pre-load the GPS unit with the Distributed and Tower Plot locations that will be sampled.

   b) Charge GPS unit batteries and CR123A type batteries (if available).

   c) Check that blades on hand clippers are clean and sharp. Sharpen if necessary.

   d) Print required datasheets and QC checklists on Rite-in-the-Rain paper.

   e) Print randomized list of clip-strip coordinates per plot/subplot.

   f) Place cold packs in the –20 ˚C freezer.

5) Check the TruPulse 360R laser rangefinder:

   a) Make sure the lenses on the TruPulse are free of dirt and debris, and clean with a lens cloth or lens tissue if necessary.

   b) Declination changes with time at each site, and should be looked up annually here: http://www.ngdc.noaa.gov/geomag-web/

   c) TruPulse Declination Offset. Check the current declination against what is entered in the TruPulse (Appendix I).

   d) TruPulse Tilt-sensor Calibration. In the rare instance that the TruPulse has suffered a severe drop shock, the tilt-sensor requires re-calibration prior to continued field work (Appendix H, Figure 4).

9.4 Sample Collection in the Field

Overview: Within each 20 m × 20 m plot (or 20 m × 20 m subplot, in larger plots), clip-harvest areas are laid out as a series of North/South facing strips with dimensions of 0.1 m × 2 m. These strips exist within 0.5 m × 3 m "cells" that are numbered and systematically gridded out across the available sampling areas within the plot (Figure 2). For each 20 m × 20 m plot (or subplot within plot), relative coordinates are assigned to each grid cell, and these coordinates correspond to the Southern end of the desired 0.1 m × 2 m clip-strip. For reference, the Southwest corner of the plot/module is (0,0), and the Northeast corner of the plot/module is (20,20)(Figure 2).

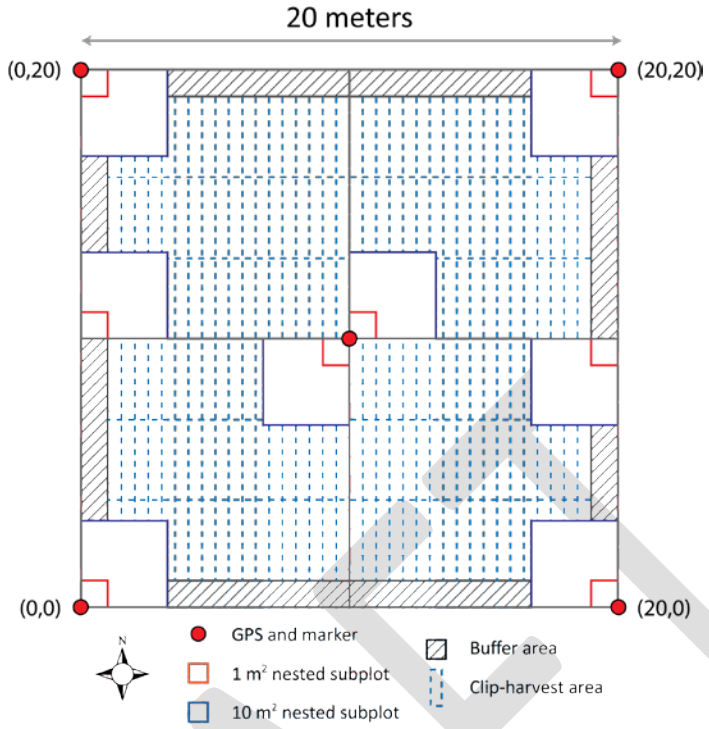

Figure 2. A Distributed Plot showing the locations of 0.5 m × 3 m clip-harvest cells (dashed blue lines) that contain potential 0.1 m × 2 m clip-harvest strips.

To determine potential harvest locations, the list of grid cell numbers is randomized, and technicians are provided with a randomized list of potential clip-strip coordinates for each 20 m × 20 m plot or subplot. Technicians should work down this list through time on a per plot/subplot basis, crossing off harvested 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.

At sites WITHOUT grazing exclosures, only one clip-strip per bout per plot/subplot is utilized. At sites WITH grazing exclosures, two clip-strips per bout per plot/subplot are utilized. Exclosures should be placed at a suitable random grid-cell location prior to grazing onset in the current season. For each sampling bout, the exclosed clip-strip is harvested along with a suitable clip-strip that is not exclosed, and the exclosure is then moved to the next suitable random location immediately after harvest.

Step-by-step procedure:

1) Confirm the **plot ID** of the plots to be harvested.

    a) The "Clip_strip_coordinates_DXX.xlsx" spreadsheet provides the randomized list of potential clip-harvest locations per plot (or per subplot for Tower Plots 40m × 40m or larger), as well as an indication of which locations have already been harvested or rejected.

    b) Identify the first potential harvest location that has not already been sampled or rejected.

    c) At sites utilizing grazing exclosures, identify the current exclosure location and the first location without an exclosure.

2) Navigate to the plot and location-within-plot where clip-harvesting will take place. See Appendix I for detailed instructions concerning use of the TruPulse 360R.

   a) If grazing exclosures are in place, use exclosure location as one clip harvest location.

   b) For the non-exclosure harvest, or If no exclosures are present, locate the relative X-coordinate within the plot/subplot:

   c) IF the relative Y-coordinate is 1, 4, or 7:

      i) Run a tape along the South edge of the plot/module from the (0,0) → (20,0) plot markers, and stretch the tape taut (Figure 2).

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

      iii) If the plot slope is > 10 degrees, or there are significant obstacles that prevent accurately stretching a tape, the TruPulse can be used in **HD** mode to place the pin flag at the correct distance and azimuth from the desired plot marker.

      iv) Be sure to calibrate the TruPulse compass in the field prior to use each day.

   d) IF the relative Y-coordinate is 10, 13, or 16:

      i) Run a tape East/West within the plot/module from the (0,10) → (20,10) positions, and stretch the tape taut (Figure 2). Use the TruPulse in **AZ** mode to guide the tape along the correct azimuth.

      ii) If the plot slope is > 10 degrees, or there are significant obstacles that prevent accurately stretching a tape, use the TruPulse as above to determine the azimuth and direction.

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

   e) Locate the relative Y-coordinate within the plot/module:

      i) Standing directly over the pin flag that was just placed at the X-coordinate, use the TruPulse in **HD** mode with a reflective surface to find the position of the Y-coordinate.

      ii) Make sure the azimuth is 0˚ (i.e. True North) when shooting the TruPulse to find the Y-coordinate.

      iii) Important: Take care not to walk on and trample the area that will be the clip-harvest strip, and similarly avoid the areas of the plot used for % cover estimation.

3) Assess whether the potential clip-strip location is suitable, and accept or reject the location:

   a) If a noticeable and significant obstacle, disturbance, or irregularity lies within the assigned clip-strip location (e.g. trees, large rocks, ant nests, etc.):

      i) The clip-strip may be moved up to 15 cm in either the East or West direction.

      ii) The location may be rejected.

   b) Obstacles, disturbances, and irregularities are considered “noticeable and significant” if:

i) They will physically prevent the delineation of the clip-strip.

ii) More than 25% of the clip-strip is affected AND the remaining area to be harvested is clearly not representative of the herbaceous vegetation throughout the rest of the plot.

c) If the clip-strip is rejected, mark it as "rejected" on the "Clip_strip_coordinates_DXX" list of locations, and assess the next locationon the list.

d) If the clip-strip is accepted, place a pin flag at the (X,Y) point associated with the South end of the clip-strip.

4) Delineate the accepted clip-strip for harvesting.

a) Using the pre-marked string and stake set, 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 in the TruPulse to orient the string in a North/South direction.

i) Keep the TruPulse at least 50 cm from non-aluminum metal plot markers.

c) Use a ruler to place the second string-and-stake set 10 cm from the first set. Check that the distance between the two strings is exactly 10 cm at both ends of the clip-strip.

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

5) Measure herbaceous vegetation structure.

a) Using a meter tape, measure the height of the tallest 5 individuals from within each of the functional groups below. Record heights to the nearest 0.01 m in the "Herbaceous Structure" datasheet (Appendix B). Functional groups are:

i) Bryophytes; code = BRY

ii) Cool-season graminoids; code = CSG

iii) Warm-season graminoids; code = WSG

iv) Leguminous forbs; code = LFB

v) Non-leguminous forbs; code = FRB

vi) Woody stemmed plants with height < 0.5 meter; code = WST

(1) Woody stemmed plants with height > 0.5 m are measured as part of the Woody Vegetation Structure Protocol (RD [08]).

vii) Old standing dead litter produced in a previous year; code = OSD

6) Harvest and sort herbaceous biomass

Most non-woody herbaceous species, as well as woody-stemmed species with height < 0.5 m, are clip-harvested within the delineated clip-strip. However, barrel-cactus species should not be clip-

harvested (they are clipped around), and *Toxicodendron spp.* must be handled according to the procedure in Appendix J. The steps below describe how to perform a typical clip-harvest.

a) Record salient observations of the clip-strip in the "Notes" column of the "Field Clip QC" checklist (Appendix C).

   i) Example: Record the estimated % area of the clip-strip disturbed by a gopher mound or the extent of minor ant nesting activity within the strip.

b) Using a sharpie, label 8# kraft paper bags for the collection of clipped biomass by functional group with the following information:

   i) **Date**; use YYYY-MM-DD format

   ii) **Plot ID**; as written on the permanent plot markers, e.g. "CPER_001".

   iii) **Subplot number**; use "0" when plot size = 20m × 20m.

   iv) **Clip ID,** as written on the "Clip_strip_coordinates" sheet.

   v) **Exclosure** presence (if applicable); use "Y" if an exclosure is present, "N" if not.

   vi) **Biomass code**; use 3-letter codes listed above.

   vii) **Bag number**; use X of Y notation, where X is the bag # and Y is the total # of bags within a given biomass code.

c) If pad-forming cacti are rooted within or that pass through the clip-strip, count and record the number of current-year pads and previous-year pads within the strip.

   i) If > 50% of a pad falls within the clip-strip, count it as a full pad. Do not count pads with < 50% of the pad in the clip-strip.

   ii) To facilitate clipping, remove the 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.

   iii) Dispose of the cactus biomass.

d) Clip and sort all aboveground biomass rooted within the clip-strip into the biomass groups listed above.

   i) Do **NOT** clip vegetation that passes through the clip-strip but is not rooted in the strip.

   ii) Technicians may split the clipping labor one of two ways:

      (1) Divide the 2 m clip-strip into 1 m sections, label two bags for each biomass code so that each technician has a set of bags, and then combine the biomass for each biomass code when clipping is finished.

      (2) Divide the clipping labor among the biomass codes. For example, one technician clips cool- and warm-season graminoids while second technician clips leguminous and non-leguminous forbs.

   iii) Clip slowly and immediately sort clipped vegetation into the appropriate coded bag.

iv) Clip vegetation as close to the ground as possible (i.e. 1-2 cm above the ground), but do not clip the crowns of perennial graminoids, as this will damage or kill the plant (Figure 3).

v) Label and use additional paper bags as necessary if you cannot fit all of the vegetation from a given category into one bag.

(1) When more than one bag per category is used, label bags within a category as "1 of X", "2 of X", etc., where X is the total # of bags ultimately used for that category.

e) Once all biomass has been harvested and sorted into bags, enter the total number of bags from each plot, as well as the total number of bags within each category into the "Field Clip QC" datasheet (Appendix C).

f) Group all of the 8# bags from a single plot into one 25# kraft paper bag.

g) IF you have just finished clipping a strip that was protected by a grazing exclosure, move the exclosure to the next suitable clip-strip location on the list of random locations for the plot/subplot.

i) Stake the exclosure into the ground.

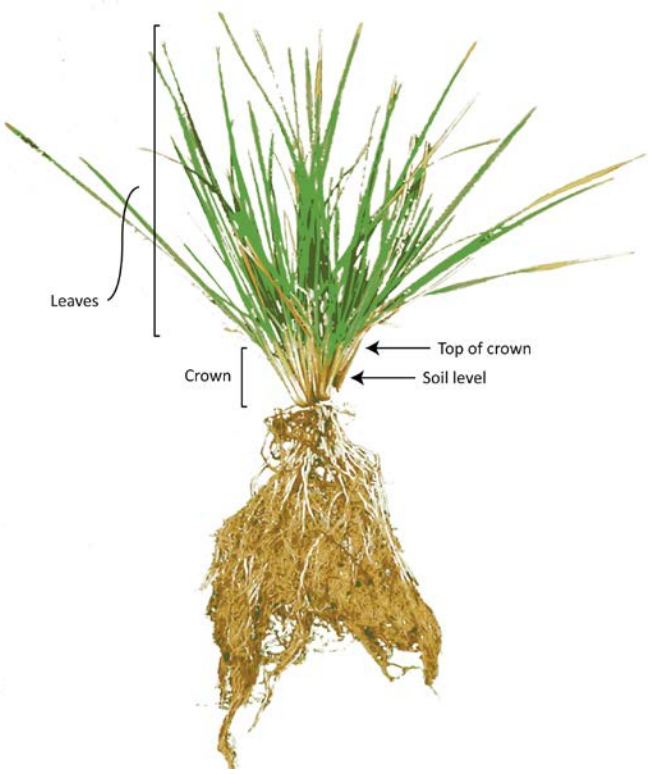

Figure 3. Illustration of a perennial grass, and the location of crown material relative to leaves and the soil surface.

9.5 Sample Preservation

1) Place all paper bags with clipped vegetation in a cooler with cold packs so that wilting is minimized and diagnostic features are preserved for the laboratory sort-check.

a) The amount of time between completion of harvest and when labeled bags should be placed in a cooler depends on many factors – e.g. humidity, temperature, wind speed, etc.

b) **Best practice:** Place bags into the chest cooler as soon as harvest of a given clip strip is complete.

2) Store clipped biomass in a refrigerator at 4˚ C for up to 24 hours.

9.6 Data Handling

1) Enter data from the "Herbaceous Structure" and "Field Clip QC" datasheets into the "Field_data_entry" and "Metadata_entry" ingest datasheets. Only transcribe those fields indicated in the datasheets.

   a) Choose the appropriate tab for the datasheets at hand.

2) Check that height data have been recorded and biomass has been clipped from all required plots.

   a) Once data have been entered from all plots, save the "csv" tab as a .csv file for CI ingest.

3) Update the clip coordinate sheets with information about "rejected/accepted" clip-strip locations. Use the following codes for "status" column:

   a) 0 = Rejected

   b) 1 = Accepted, no exclosure

   c) 2 = Accepted, exclosure

   d) 3 = Rejected temporarily, inundated

9.7 Refreshing the Sampling Kit

1) Make sure the following consumable supplies are available in sufficient quantity for the next round of clip-harvests:

   a) Paper bags, 8# and 25# kraft

   b) Rite-in-the-Rain paper

2) Return any re-usable cold packs to the -20˚ C freezer.

9.8 Equipment Maintenance, Cleaning and Storage

1) If necessary, clean the lenses on the TruPulse with a micro-fiber cloth.

2) Maintain equipment for the next sampling day/bout by doing the following:

   a) Clean blades of hand clippers, and sharpen if necessary.

      i) Blades may cleaned with either water or ethanol, whichever is most suitable.

      ii) If *Toxicodendron spp.* have been harvested, clean clipper blades with Tecnu soap or equivalent.

   b) Recharge batteries for the GPS unit.

c) Recharge batteries for the TruPulse (if applicable).

3) Return all equipment to the appropriate storage location.

10 LABORATORY STANDARD OPERATING PROCEDURE

10.1 Sample Processing Timing

1) Biomass samples that are clipped and quickly sorted to functional group in the field must be carefully checked in the lab for sorting accuracy.

   a) Bags should be checked by the same technicians who initially performed the clip-harvest of those bags in the field.

   b) Bags of clipped biomass should be checked for sorting accuracy as soon as possible and no later than 24 hours after harvest. Time is of the essence because as leaves wilt, the distinguishing features of many plants become harder to discern.

2) Clipped and sorted biomass must be placed in a drying oven within 24 h of the field harvest.

3) Following drying, biomass should be weighed, recorded, and entered into the appropriate database of Excel spreadsheet within 30 days.

10.2 Equipment and Materials

Table 3. Equipment and supplies required for processing clip-harvest samples in the laboratory.

Maximo Item No.	Item Description	Quantity	Habitat-Specific	Special Handling
	Region-specific plant ID book	1	Yes	No
	Region-specific dichotomous or polyclave key to plant ID	1	Yes	No
MX100230	Drying oven	Space in oven for samples	No	No
	Large plastic bag - black trash bag or equivalent - for temporary storage of oven dried samples (e.g. Uline # S-5111)	Box of 100 as needed	No	No
MX100265	Mass balance (0.01 g accuracy)	1	No	No
MX105897, MX105898	Desiccator (required only when mosses are present)	1	Yes	No
MX100689	Large weigh boats	100	No	No
MX100235	Grinding mill	1	No	No
MX103235	Sample microsplitter	1	No	No
MX103237	Hi-back pans for microsplitter	2	No	No
MX103219	Scintillation vial with caps	1 per biomass group per plot	No	No
MX102002	Sharpie waterproof marker, extra fine tip (or equivalent)	2	No	No
	Pencils	2	No	No
	Data sheets and QC checklists	As needed	No	No

10.3 Preparation

1) Check to ensure that all consumables required for lab work are available for use. Re-order items as necessary.

2) Clear adequate bench space for sorting, and clear required space within drying ovens.

10.4 Sample Processing in the Lab

1) Confirm that all bags are returned from the field.

    a) Count all bags and enter count into the "Lab clip QC" datasheet.

    b) Compare this independent count of the bag numbers to the field count on the "Field clip QC" datasheet to make sure that no bags have been misplaced.

2) Make sure that all bags from a given clip-strip are collated in the correct 25# kraft bag.

3) Check bags of sorted biomass for sorting accuracy *within 24 hours of harvest.* The objectives for this task are:

    a) To 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.

    b) To 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.

4) To check the sorting of biomass from a given clip-strip into functional groups:

    a) Collect all of the bags from a given clip-strip. Enter the required information on the "Lab Clip QC" datasheet. All fields except for the "Remarks" field on the "Lab Clip QC" sheet are for internal QC purposes only; "Remarks" are entered into the "Remarks" field on the "Metadata_entry" ingest sheet. "Lab Clip QC" fields include:

        i) **Name**

        ii) **Date** (YYYY-MM-DD format)

        iii) **Start** and **stop times** for sorting (24-h HHmm format)

        iv) **Plot ID** and **subplot number**

        v) **Clip ID** for clip-strip

        vi) **Remarks** – e.g. problems with species ID in the field. Record "Remarks" in the "Metadata_entry" ingest sheet.

    b) Choose a bag and empty the contents onto a clean workbench. Carefully sort the biomass that was quickly sorted in the field. *Exception:* The OSD bag is not sorted further in the lab.

    c) After checking the sorting accuracy for the contents of the bag:

i) Place the sorted, checked biomass back into the bag.

ii) Place any previous years' litter into the "OSD" bag.

iii) Set aside all other biomass that does not belong in the original bag into separate piles (i.e. one pile for each biomass category).

iv) Clean the bench of any debris, and proceed to the next bag from the same clip-strip.

d) Have the lead plant field technician perform a spot-check that all biomass that was mis-sorted in the field has now been correctly sorted.

This step is particularly important early in the year when seasonal field techs may be less familiar with local flora. As employees become well-trained, a spot-check of 5%-10% of sorted bags will suffice.

e) Place piles of resorted biomass in the appropriate bags.

f) Place all of the biomass bags from the current clip-strip into the larger 25# bag for that clip-strip, and put the biomass into the drying oven.

g) Record the ending time in the "Lab Clip QC" datasheet.

i) Knowing how much time is required to sort all the biomass from a given plot is important for improving both the workflow and the time management of biomass harvesting.

h) Clean the bench and proceed to checking bags from the next clip-strip. Sort all harvested biomass with 24 hours of collection.

5) Place sorted biomass bags into a 60˚ C drying oven for 72 h - 120 h (3 d - 5 d).

a) Critical point: In order to assess how long different batches of bags have been in the oven, particularly when harvests from multiple days occupy the same oven, label each 25# bag (with the 8# bags inside) with the date and time it was placed in the oven.

b) To ensure that plant material harvested on a given day is dry, check the weight of the same subset of n=10 bags by sequentially weighing after day 1, 2, 3, 4, etc.

i) Use the "Lab Drying QC" datasheet to record repeated sample weights during drying (Appendix E).

ii) Samples are dry when weight is constant from one day to the next (within ± 0.05 g).

c) After removing samples from the drying oven, place into a large plastic bag (e.g. a black plastic garbage bag or equivalent), and let them come to room temperature.

i) Placing samples in a bag is important because otherwise they will absorb water from the air as they cool, particularly in humid environments.

d) If mosses were harvested and dried (group = BRY), place this biomass into a desiccator before weighing. Mosses can be extremely hygroscopic and will readily pick up moisture (and thus weight) from the air if not treated this way.

6) **Once dried, samples may be stored for up to 30 days**.

7) Using a mass balance (0.01 g accuracy) and a weigh boat, weigh the contents of each bag.
   a) Record the mass to the nearest 0.01 g on the "Clip Biomass" datasheet (Appendix F).
   b) Use the pre-labeled datasheet to make sure that weights are recorded for all bags.
   c) Weigh all bags from a given plot consecutively before weighing bags from another plot.
   d) For large volumes of biomass that do not readily fit into a large weighboat, use one of the following strategies:
      i) Crush or chop the biomass to reduce the volume so it will fit into the weigh boat.
      ii) Use an empty paper lunch bag as a "weighboat".
      iii) Avoid splitting the biomass into subgroups for weighing, as uncertainty values must be added each time a subgroup is weighed.

8) QA for weighing:
   a) Select 10% of dried, previously weighed samples for re-weighing.
   b) A different technician should perform the re-weighing.
   c) Record QA data in a separate "Clip Biomass QA" datasheet (Appendix G).

9) Once all weights have been recorded, return the plant material to the appropriate bags, and place into temporary storage. Samples in temporary storage can then be prepared as time permits for bioarchive (see section 10.5).

10.5 Sample Preservation

1) Prepare samples for temporary storage in the Domain Lab facility until bioarchive contracts are established.
   a) Coarsely grind each biomass group per clip-strip with a Wiley Mill.
   b) Use a splitter to generate a representative 20 mL sub-sample that can be stored in a polypropylene scint vial.
      i) If a given biomass group is < 20 mL total volume, store the entire ground sample in the scint vial.
      ii) Discard biomass in excess of 20 mL total volume.
   b) Label the scint vial with the clip_ID, date, and biomass code.

10.6 Sample Shipping

Samples are not shipped at this time.

10.7 Data Handling

Enter all data from the "Lab Clip QC", "Clip Biomass", and "Clip Biomass QA" datasheets into the appropriate data ingest sheets.

Data from the "Clip Biomass" datasheets should be double-entered by different technicians, and any discrepancies should be resolved by examining the original paper datasheet.

1) There should be one Excel file containing herbaceous clip harvest ingest datasheets for all of the plots measured in a given year, and this file should have multiple tabs for each of the types of required data.
    a) Choose the appropriate tab for the datasheets at hand.
    b) Once data have been entered from all plots, save the "csv" tab as a .csv file for CI ingest.

10.8 Equipment Maintenance, Cleaning and Storage

1) Balances should be calibrated with a standard calibration weight set:
    a) After initial installation.
    b) Any time the balance is moved.
    c) Every 6 months.
    d) If you suspect readings are inaccurate for any reason.
2) Clean the grinding mill after grinding each biomass group, and after grinding is complete:
    a) Clean with air after grinding each biomass group.
    b) Clean with air and ethanol after grinding is complete on a given day.

11 REFERENCES

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

APPENDIX A LIST OF CLIP-HARVEST SAMPLING WINDOWS PER SITE

Domain #	Site	Bout #	Sampling Onset
1	Harvard Forest	1	7/15 – 8/1
3	Ordway-Swisher	1	7/15 – 8/1
3	Disney	1	7/15 – 8/1
10	CPER	1	5/23 – 6/7
10	CPER	2	8/15 – 8/29
10	ROMO	1	7/15 – 8/1
10	Sterling	Ag site: Depends on # crops/year	Ag site: Depends on site host harvest plans

APPENDIX B HERBACEOUS STRUCTURE DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Herbaceous Structure Datasheet Page ___ of ___

recordedBy:		# Cactus Pads (old):	
eventDate: *(YYYYMMDD)*		# Cactus Pads (new):	
Clip ID:		Exclosure (*Yes/No*):	
Subplot #:			

Biomass code	Height 1	Height 2	Height 3	Height 4	Height 5
BRY					
CSG					
WSG					
LFB					
FRB					
WST					
OSD					

recordedBy:		# Cactus Pads (old):	
eventDate: *(YYYYMMDD)*		# Cactus Pads (new):	
Clip ID:		Exclosure (*Yes/No*):	
Subplot #:			

Biomass code	Height 1	Height 2	Height 3	Height 4	Height 5
BRY					
CSG					
WSG					
LFB					
FRB					
WST					
OSD					

APPENDIX C FIELD CLIP QC DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Field Clip QC Datasheet Page ___ of ___

recordedBy:		Start time (*24h, HHmm*):	
eventDate: *(YYYYMMDD)*		Stop time (*24h, HHmm*):	
Clip ID:		Total # bags:	
Subplot #:			

Biomass code:	BRY	CSG	WSG	LFB	FRB	WST	OSD
Bag count:							

Remarks: *(e.g. clip-strip moved 15 cm W due to ant nest)*

recordedBy:		Start time (*24h, HHmm*):	
eventDate: *(YYYYMMDD)*		Stop time (*24h, HHmm*):	
Clip ID:		Total # bags:	
Subplot #:			

Biomass code:	BRY	CSG	WSG	LFB	FRB	WST	OSD
Bag count:							

Remarks:

APPENDIX D LAB CLIP QC DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Lab Clip QC Datasheet Page ___ of ___

recordedBy:		Start time (*24h, HHmm*):	
eventDate: *(YYYYMMDD)*		Stop time (*24h, HHmm*):	
Clip ID:		Total # bags:	
Subplot #:			

Biomass code:	BRY	CSG	WSG	LFB	FRB	WST	OSD
Bag count:							

Remarks: *(e.g. one bag BRY unaccounted for)*

recordedBy:		Start time (*24h, HHmm*):	
eventDate: *(YYYYMMDD)*		Stop time (*24h, HHmm*):	
Clip ID:		Total # bags:	
Subplot #:			

Biomass code:	BRY	CSG	WSG	LFB	FRB	WST	OSD
Bag count:							

Remarks:

APPENDIX E LAB DRYING QC DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Lab Drying QC Datasheet

Start date (YYYYMMDD):

Start Time (24 h, HHmm)

Page ___ of ___

Clip ID	Biomass code	t=24 h		t=48 h		t=72 h		t=96 h		t=120 h		t=144 h	
		Mass (0.01 g)	Initials	Mass (0.01 g)	Initials	Mass (0.01 g)	Initials	Mass (0.01 g)	Initials	Mass (0.01 g)	Initials	Mass (0.01 g)	Initials

APPENDIX F CLIP BIOMASS DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Clip Biomass Datasheet Page ___ of ___

recordedBy:

Clip ID	Subplot #	Clip eventDate (*YYYYMMDD*)	Biomass code	Final dry mass (0.01 g)

APPENDIX G CLIP BIOMASS QA DATASHEET

This datasheet is provided for document control purposes only. DO NOT print and use.

Herbaceous Biomass: Clip Biomass QA Datasheet Page ___ of ___

recordedBy:

Clip ID	Subplot #	Clip eventDate (*YYYYMMDD*)	Biomass code	Final dry mass (0.01 g)

APPENDIX H USING AND CALIBRATING THE TRUPULSE 360R

Setting the Declination Offset

1) Press the "Power/Fire" button to turn on the unit. The viewfinder will display the main "Measurement Mode" screen.
2) Press and hold ▼ for 4 s to enter "System Setup Mode".
3) Press ▼ until **H_Ang** is displayed in the viewfinder, then press "Power/Fire".
4) **dECLn** will be displayed in the viewfinder, press "Power/Fire".
5) **no** and **dECLn** will blink. Press ▼ until **YES** and **dECLn** blink, then press "Power/Fire" again. The current declination is shown in the viewfinder.
6) If this is the correct value, press and hold ▲ to return to the main "Measurement Mode" screen.
7) If the displayed value is incorrect for your current location:
   a) Press either ▲ or ▼ to change the tenths value, press "Power/fire".
   b) Press either ▲ or ▼ to change first integer value, press "Power/fire".
   c) Press either ▲ or ▼ to change second integer value, press "Power/fire".
   d) The value just entered will blink. Press "Power/fire" to confirm and return to the "Measurement Mode" screen.

Tilt Sensor Calibration

1) Press the "Power/Fire" button to turn on the unit. The viewfinder will display the main "Measurement Mode" screen.
2) Press and Press and hold ▼ for 4 s to enter "System Setup Mode".
3) Press ▼ until **inC** is displayed in the viewfinder, then press "Power/Fire"..
4) **no** and **CAL** will blink. Press ▼ until **yes** and **CAL** blink, then press "Power/Fire" again.
   a) Calibration can be aborted by pressing "Power/Fire" when **no** and **CAL** are alternately displayed.
5) **C1_Fd** will be displayed in the view finder.
6) Place the TruPulse on a flat, relatively flat surface (within 15deg of level). Follow the sequence outlined in the following illustration (Figure 4).
   a) At each step wait approximately 1 second before pressing "Power/fire", then wait another second before moving to the next position. It is important that the unit is held steady when you press "Power/fire".
   b) To abort and return to previous calibration at any point hold ▲ or ▼ for 4 sec.
7) After all 8 positions have been run through, look through the eyepiece. Either a **PASS** or **FAiL** message appears in the view finder.

a) **PASS**: Press the “Power/Fire” Button to return to the measurement mode.

b) **FAiL1**: Excessive motion during calibration. Unit was not held steady.

c) **FAiL2**: Magnetic saturation error. Local magnetic field too strong.

d) **FAiL3**: Mathematical fit error.

e) **FAiL4**: Calibration convergence error.

f) **FAiL6**: Orientations were wrong during the calibrations.

8) If **FAiL** appears, press the “Power/Fire” button. **No** and **CAL** will alternately blink allowing you to do a new calibration. IF the calibration fails, the unit reverts to the previous calibration.

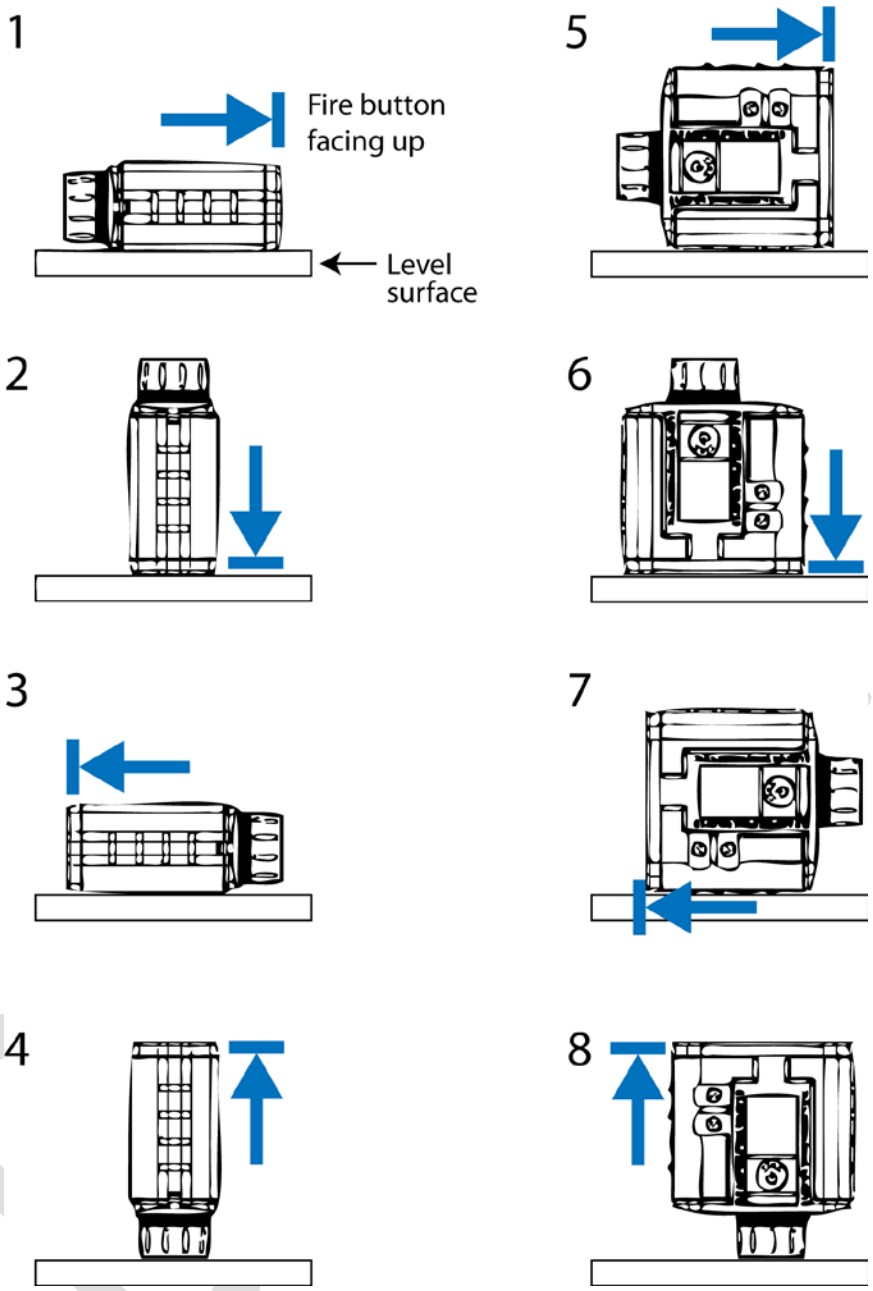

Figure 4. The tilt-sensor calibration routine for the TruPulse 360R instrument. The blue arrow and line indicate the direction of the lens at each calibration step.

Using and Calibrating the TruPulse 360R Compass

The internal compass of the TruPulse is susceptible to error and to interference with common metallic objects. The following objects may affect the compass performance, and should be kept at least 50 cm (20 in) away from the TruPulse during operation:

Batteries	Nails
Data collectors or computers	Pin flags w/ metal stakes
Portable electronics	Steel-rimmed eyeglasses
Metal watch bands	Eyeglass spring-hinges
Non-aluminum tripods	

When using the TruPulse compass, it is good practice to check the compass performance against a standard mirror-site compass or a previously established plot-line at the beginning of each day, or when beginning a new plot. In addition, ALWAYS CHECK AND RECALIBRATE THE COMPASS AFTER CHANGING THE BATTERIES. It is common for the compass calibration to be inaccurate when the low battery indicator is displayed in the viewfinder, and you should always replace the batteries when this indicator appears.

If the compass requires calibrating, you must first determine that you are in an area free from local magnetic interference. Either of the following simple tests can be used in the field to test for local magnetic interference:

1) Choose a target at least 100 m away, and shoot to it. Note the azimuth. Then step backward or forward 1 m along the sight-line to the target and shoot again. Note the second azimuth.
    a) The second azimuth should be within 1/10 to 5/10 of a degree of the first azimuth. If it is, you are likely in an anomaly-free area.
    b) For increased confidence, repeat the test with a second target at 90˚ to the azimuth of the first target.
2) Select a target at least 10 m away, shoot to it, and note the azimuth. Move to the target that was just shot, and shoot back toward the spot that was just occupied. Note the second azimuth.
    a) The two azimuths should be 180˚ different, plus or minus no more than a few tenths of a degree.

Once you have ascertained that the current location is free from local magnetic interference, complete the following steps to calibrate the TruPulse 360R compass:

1) Press the “Power/Fire” button to turn on the unit. The viewfinder will display the main “Measurement Mode” screen.
2) Press and hold ▼ for 4 s to enter “System Setup Mode”.
3) Press ▼ until **H_Ang** is displayed in the viewfinder, then press “Power/Fire”..
4) **dECLn** is displayed. Press ▼ to display the **HACAL** option, then press “Power/Fire” again.

5) **No** and **HACAL** will alternately blink. Press ▲ or ▼ to display **YES** and **CAL**, then press "Power/Fire" to begin calibration.

   a) Calibration can be aborted by pressing "Power/Fire" when **no** and **CAL** are alternately displayed.

6) **C1_Fd** will be displayed in the view finder.

7) Use a standard mirror-site compass to determine the direction of ***magnetic*** North. Holding the TruPulse 360R and facing close to ***magnetic*** North (± 15˚), the lenses should be facing as shown in (Figure 5). To complete the calibration routine, follow the sequence outlined in (Figure 5).

   a) At each step wait approximately 1 second before pressing "Power/fire", then wait another second before moving to the next position. It is important that the unit is held steady when you press "Power/fire".

   b) To abort and return to previous calibration at any point hold ▲ or ▼ for 4 sec.

8) After all 8 positions have been run through in sequence, look through the eyepiece. Either a **PASS** or **FAiL** message appears in the view finder.

   a) **PASS**: Press the "Power/Fire" Button to return to the measurement mode.

   b) **FAiL1**: Excessive motion during calibration. Unit was not held steady.

   c) **FAiL2**: Magnetic saturation error. Local magnetic field too strong.

   d) **FAiL3**: Mathematical fit error.

   e) **FAiL4**: Calibration convergence error.

   f) **FAiL6**: Orientations were wrong during the calibrations.

If **FAiL** appears, press the "Power/Fire" button. **No** and **CAL** will alternately blink allowing you to do a new calibration. IF the calibration fails, the unit reverts to the previous calibration.

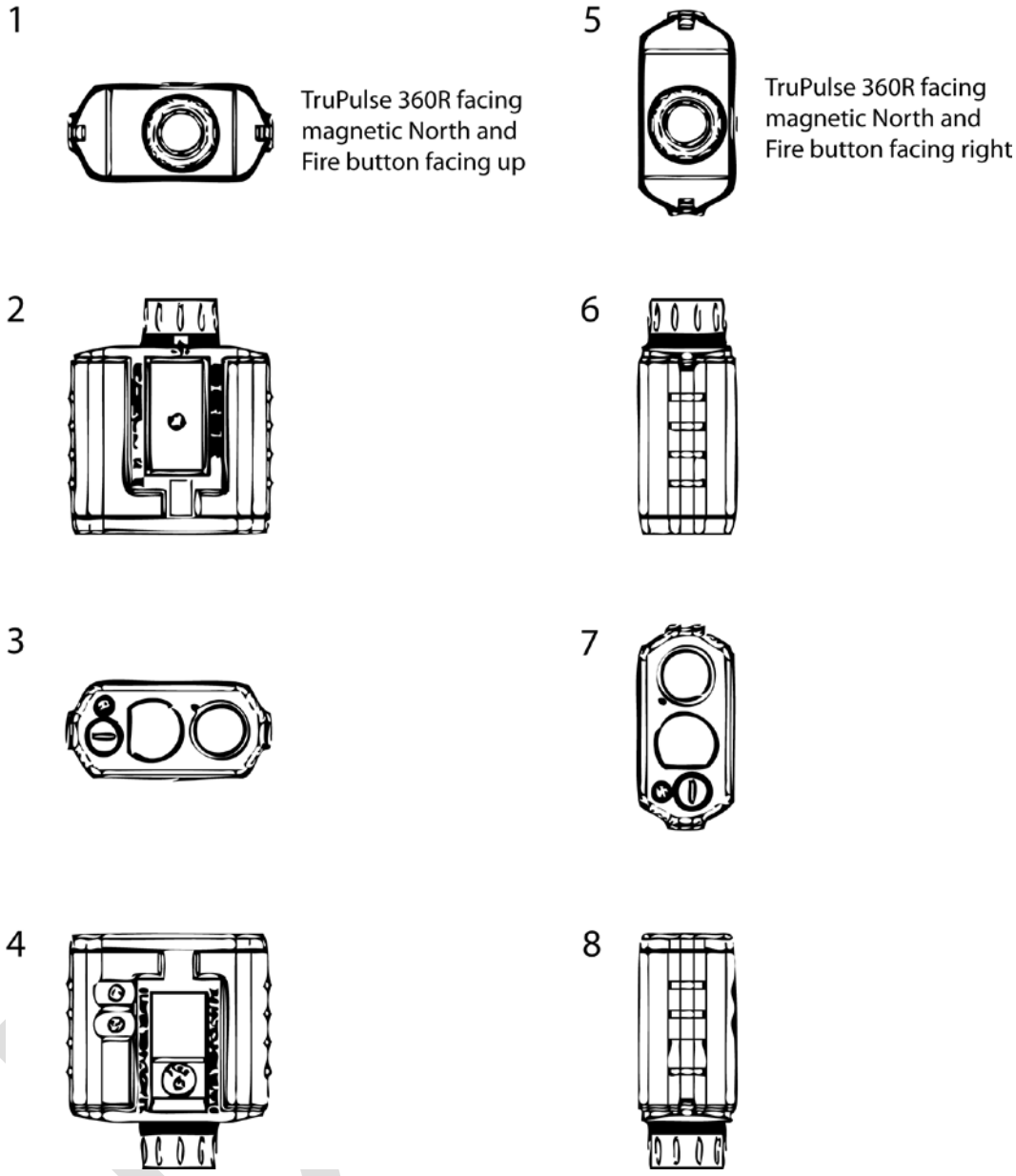

Figure 5. The internal compass calibration routine for the TruPulse 360R instrument.

Measuring Distance from a Known Point

1) Press “Power/Fire” to turn on the TruPulse.
2) Set the unit to Target Mode = Filter
    a) Press ▲ for 4 seconds. The active Target Mode appears in the viewfinder. Press ▲ or ▼ to cycle through available Target Modes.
    b) Available Target Modes are:
    c) Std = Standard; Single-shot mode. The laser will acquire data from one target.
    d) Con = Continuous; In this mode, by pressing and holding “Power/Fire” the unit will acquire a target, and will continue to acquire additional targets for a maximum of 10 s. The most recently acquired target is displayed in the viewfinder. Useful for scanning trees in order to find the highest point.
    e) CLO = Closest; Press and hold the “Power/Fire” button in this mode. Once the initial target is acquired, the unit will acquire additional targets. The **MULTI** indicator in the viewfinder denotes additional targets have been acquired. The closest acquired target is displayed in the viewfinder. Useful with narrow targets in the foreground.
    f) FAr = Farthest; Identical to CLO above, but the farthest target is displayed in the viewfinder. Useful with a target partially obscured by brush, or for finding the highest point of a tree.
    g) Flt = Filter; In this mode, the laser’s sensitivity is reduced to only detect pulses returned from a reflective surface. Useful when attempting to measure targets through thick brush. ***In very heavy brush, the optional foliage filter can be used in this mode, but is not required.*** In this mode, an ‘F’ appears at the left of the viewfinder.
    h) Choose “Flt” and press “Power/Fire” to make the chosen Target Mode active.
3) Press either the ▲ or ▼ button until **HD** (i.e. Horizontal Distance) appears in the viewfinder.
4) Person 1: Hold the reflective surface at the base of the stem so that it is visible to Person 2.
5) Person 2: Look through the TruPulse viewfinder, aim the crosshairs at the reflective surface held by Person 1, and press and hold “Power/Fire” until the distance is displayed in the viewfinder; record this distance.

Measuring Azimuth from a Known Point

1) After recording the **HD** to the stem above, press ▲ three times until **AZ** (i.e. azimuth from True North) appears in the viewfinder and the angle in degrees is displayed; record this angle.
2) The angle should be preceded by a “**d**” indicating that declination has been set for the TruPulse at your current location (as described previously).

APPENDIX I CLIP HARVESTING *TOXICODENDRON* SPECIES

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 (protocol will be updated to reflect this).
    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. If 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) Handling clipped *Toxicodendron* biomass in the field:
    a) Wear cotton gloves and dispose after single use. Toxic oils can pass through nitrile or latex gloves.
        i) For example, http://www.globalindustrial.com/p/safety/hands/cotton-canvas-gloves/anchor-4501v-8-oz-cotton-canvas-knit-wrist-1110
    b) 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.
    c) Bring a clean, new plastic bag to the field for storing and transporting contaminated gloves and clippers after use.
    d) Wear a thin outer layer of disposable PPE over clothes and shoes, such as:

        http://www.disposable-garments.com/products/Koolguard_Tyvek_Alternative/Koolguard_Coveralls_with_elastic_wrist_and_ankles_p3805.html
    e) After field work and upon returning home, wash clothing acccording to these guidelines or similar:

        http://laundry.about.com/od/removeoutdoorstains/a/poisonivylaundry.htm
3) Processing *Toxicodendron* biomass in the lab:
    a) Minimize potential spread of toxic oil by putting *Toxicodendron* biomass bags into the same drying oven every time.
    b) When drying is complete, clean drying oven shelves used for drying *Toxicodendron* biomass bags with hot water and Tecnu. Wear appropriate PPE when cleaning.
    c) 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.
    d) After weighing, dispose of all *Toxicodendron* biomass bags.

i) At this point in time, *Toxicodendron* tissue will not be specimen mounted or archived.