NERRS SWMP Vegetation Monitoring Protocol

Long-term Monitoring of Estuarine Vegetation Communities

National Estuarine Research Reserve System Technical Report

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Introduction

Macrophyte communities including tidal wetlands, mangrove forests and submersed vegetated beds as well as the transitional ecotones between tidal and non-tidal habitats are important components of estuarine ecosystems. They form a buffer between upland systems and the sea and provide complex habitats with irreplaceable value to both coastal systems and their adjacent watersheds. They serve as important sites for education and recreation and are critical components of the National Estuarine Research Reserve System (NERRS).

Despite their currently recognized values, the distribution, abundance and diversity of these communities have been lost as a direct result of human impact. For example, nearly 50% of the coastal wetlands have been lost in the United States through dredging and filling activities (Dahl 1990). "Status and Trends of Wetlands in the Coastal Watersheds of the Eastern United States, 1998 to 2004" documents an annual loss of 59,000 acres of wetlands from 1998 to 2004 in the Great Lakes, Atlantic Ocean, and Gulf of Mexico. Changes in hydrological regimes through alterations in drainage (Niering and Warren 1980) or tidal flushing patterns (Roman et al. 1984), as well as local subsidence (Davis and Andronaco 1987) have severely impacted many others. In the Chesapeake Bay region beds of submersed aquatic vegetation are currently at their lowest levels of abundance in recorded history (Orth and Moore 1983) and these declines can be directly related to watershed inputs of sediments and nutrients accelerated by human activities (Kemp et al. 1983; Brush 2001).

Natural forces can also have significant influences on these vegetation systems. Storms can be powerful forcing functions modifying these areas over both the short and long term (Davis and Andronaco 1987; Short and Wyllie-Echeverria 1996; Roman et al. 1997). Sea level rise or other changes in water level, as well as global climate change can have a wide range of effects on submersed and emergent coastal plant communities as well as wetland-upland ecotones including: submergence and drowning, changes in salinity, temperature, runoff and water quality, as well as increasing UV radiation (Warren and Niering 1993; Watson et al. 1996; Short and Neckles 1999). Additionally, non-indigenous or invasive plant species can have significant impacts on the structure and function of these habitats (Sandlund 1999).

Restoration of emergent and submersed aquatic vegetation (SAV) communities and adjacent upland ecotones is a major management goal in most developed estuarine areas and is an important initiative of NOAA and NERRS. Further, the process of natural recovery of systems impacted by extreme events such a hurricanes can be of significant interest. Evaluating the success of restoration efforts, as well as assessing the recovery of impacted communities requires detailed, statistically rigorous, protocols that can be equally applied to both reference areas and impacted sites (Dionne et al. 2012). Periodic, consistent, long-term monitoring of un-impacted or reference sites can also provide measures of natural variability that are very useful in evaluating restoration efforts or recovery from perturbation.

Evaluating habitat change of vegetation communities can be accomplished at various levels of detail in the landscape. Comprehensive, broad evaluation of habitats typically requires the use of airborne or satellite remote sensing tools and imagery. Ground surveys, although not a replacement for remote imagery, provide a level of detail for assessing community composition and change that is complementary to broader scale remote surveys. Additionally, the implementation of statistically rigorous ground surveys can provide a foundation for other monitoring activities related to quantifying the process of change in these communities.

This monitoring protocol for emergent and submersed vegetation communities has the following objectives:

- 1. Quantify vegetation patterns and their change over space and time;
- 2. Maintain consistency with other monitoring protocols used nationally and worldwide;
- 3. Apply over a wide range of estuarine sites and habitats, and for a variety of reserve specific purposes;
- 4. Quantify relationships among the various edaphic factors and the processes that are regulating the patterns of distribution and abundance in these communities;
- 5. Support comprehensive remotely sensed mapping of vegetation communities and other NERRS System Wide Monitoring Program (SWMP) data collection protocols, as well as NERRS/NOAA education, stewardship and restoration efforts.

Methods

The general approach used here consists of fixed transects with permanent sampling plots located along transects that can be stratified or otherwise located within vegetation zones or defined segments of the ecotone, mangrove forest, marsh or SAV bed. This approach has been used in a variety of studies for assessments of vegetation communities (Doumlele 1981; Moore et al. 1995; Perry and Atkinson 1997; Perry and Hershner 1999) and has been recently adopted as a monitoring protocol by the National Park Service and others to assess and compare both reference and restoration wetland sites on local and regional scales (Neckles and Dionne 2001; Roman et al. 2001; Neckles et al. 2002). Additionally, similar protocols have been established for quantification of seagrass dynamics in a global seagrass monitoring program (http://www.SeagrassNet.org; Short et al. 2002).

Site Selection

Sites in each study area are first identified as locations that have historically not been markedly impacted by natural or anthropogenic factors. The sites should be representative of existing estuarine vegetation communities in the region. These determinations may have to be made using the best professional judgment of NERRS scientists based upon available information for each study area. As much as possible they should be associated with locations of other SWMP monitoring activities including water quality sampling and/or Sentinel Sites Monitoring. The focus of this detailed vegetation monitoring can vary with the particular circumstances or goals for each study.

For example, in the Chesapeake Bay Reserve in Virginia (CBNERRVA), a different emergent marsh community is associated with each of the four reserve components (i.e. Goodwin Islands, Catlett Islands, Taskinas Creek, and Sweet Hall Marsh) (http://www.vims.edu/cbnerr/reservesites/index.htm), and each reserve component is located within a different salinity regime of the estuary. In this case, each Reserve component could potentially serve as a study site. However, since seagrass beds are known to be associated with only the most downriver site (i.e. Goodwin Islands), the submersed macrophyte sampling would only be conducted at that reserve component.

Sites may also be selected to represent potential impacts of interest and be established as elective areas for comparative monitoring. Some of these potential impacts include:

- 1. Invasive or non-native species expansions.
- 2. Rare species declines
- 3. Changes in hydrology, geomorphic process, sea level rise or salinity intrusions.
- 4. Catastrophic impacts such as oil spills or storms.
- 5. Direct or indirect human impacts such as dredging, diking, filling or subsidence due to groundwater withdrawal or other factors.
- 6. Disease.

Emergent or submersed areas that have been or will be the focus of restoration efforts, either directly or indirectly through watershed modifications, could also be chosen for study. Other factors to consider in site selection are meeting or advancing NERRS Strategic or System-Wide goals such as the Sentinel Site Initiative or the Education TOTE (Teachers on the Estuary) Program. The objectives of monitoring for each specific site should be chosen *a priori* for each study; however, the protocols provided here can also be applied to additional sites chosen in the future. For example, if additional property were acquired and wetlands on that property were the focus of restoration efforts, the habitat change associated with those efforts could be quantitatively monitored over time or could be compared to an existing study site if appropriate.

Application to Sentinel Sites Initiative

This vegetation monitoring protocol can be used as an element within the NERRS Sentinel Sites Initiative. [Additional information may be added here, in consultation with

the Sentinel Sites workgroup, detailing how the vegetation monitoring protocol should be used at sites also implementing Sentinel Sites work. For instance, transects should perhaps be required to be located near SETs and/or water level measurement stations. This may apply primarily to emergent vegetation.]

Site Delineation

The vegetation habitat of interest for study should be delineated and the boundaries defined *a priori*. A general base map should be developed providing the fundamental features of the site. The degree of detail of this reference map will be dependent on the extent of the geographical detail of the region. Typically, topographic quadrangles, digital orthophoto quarter quadrangles, vertical or oblique photographs can be used.

Stratification of a study site into segments of similar community type based upon the dominant environmental gradient may be necessary if a significant environmental or other gradient exists (Roman et al. 2001). For example an emergent creek marsh system dominated by an upstream-downstream salinity gradient or a restriction in tidal flushing could be stratified into two or more segments (Figure 1). Similarly a submersed aquatic vegetation community affected by a gradient of exposure (fetch), sediment type or some water quality characteristic such as salinity, or tide range could be stratified into several segments.

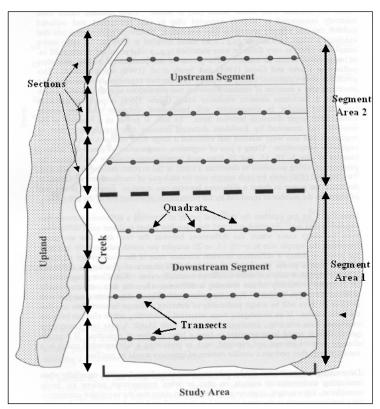


Figure 1. Emergent Marsh Sampling Design (from Roman et al. 2001).

Emergent Vegetation Sampling Design

Each study area or segment of a study area may then be systematically divided into equal sized sections, if necessary (e.g., for large study systems), to achieve good interspersion of samples throughout the area. One or two transects are then randomly located within each section so that a total of three to five transects will be located in a segment for a total of approximately 20 permanent plots (i.e., quadrats). Setting the random location of the sampling transects within a section can be accomplished by dividing the shoreline of each section into equal sized intervals, numbering each interval and then randomly choosing interval numbers for transect establishment using tables or computer random number generation. Individual transects should be no less than 10 m apart to maintain independence and should cover an area that is representative of the segment. Each transect should traverse the elevation gradient from the creek bank to the upland. A detailed description of transect establishment for an emergent wetland study site using this approach is provided in Roman et al. (2001) (www.nature.nps.gov/im/monitor/protocoldb.cfm).

The first permanent plot on each transect should be randomly located within the marsh zone adjacent to the creek bank or open water ("creek marsh zone"). For example, if the creek marsh zone is only 3 m wide then this zone is divided into five 60 cm intervals and the first permanent plot is randomly located at the mid-point of one of the five intervals. Similarly, if the creek marsh zone is 30 m wide then the first plot is located randomly within the first 10 m of the transect. Each "permanent plot" is an area immediately adjacent to a specific point along the transect that is used for repeated vegetation sampling (Figure 2).

The remaining permanent plots are then located at regular intervals along each transect. A total of approximately 20 plots per marsh segment are suggested for adequate sampling power (Roman et al. 2001), therefore the permanent plot intervals should be adjusted for the overall area of the marsh to achieve this replication. No permanent plots should be less than 10 m apart. Scaling of placement of the plots across the landscape will depend on the scale of the study area. For example, a larger marsh could have permanent plots placed at 20 m intervals along each transect with individual transects 50m apart. Alternatively, a smaller creek marsh could have permanent plots placed at 10m intervals, with transects 10 m apart. Each permanent plot is then permanently marked with labeled stakes driven into the marsh. The permanent vegetation sampling plots which are one meter on a side are offset approximately 1 m from the marking stake perpendicular to the transect line and two diagonal corners of each plot are marked with small stakes (after Roman et al. 2001).

A groundwater well may be electively established approximately 1 m from the marking stake at 180° from the permanent plot. Capped wells can be constructed from schedule 40 PVC extended at least 50 cm into the marsh surface (after Roman et al. 2001). If desired, wells can be established using other established and published methodologies provide they are appropriate and well documented in the methods and metadata (USCOE, 2000). Temporary groundwater "sippers" (e.g. plunger with long tube) can also be used to sample groundwater. Other sampling items such as a permanent

surface elevation table (SET) may be similarly arranged around the sampling location stake as appropriate (see section above on linkages to NERR Sentinel Sites Initiative).

At sites where there is a high potential for vegetative community migration due to sea level rise or other driving forces, additional elective sampling should be implemented to allow for monitoring of future shifts. In instances such as this at least one permanent plot should be assessed on either end of the current vegetation boundaries, with the same spacing between plots as is used in the rest of the transect. Appendix 4 outlines additional optional protocols that can be applied for monitoring boundary changes.

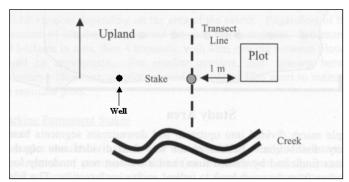


Figure 2. Sampling Plot and Well Orientation (modified from Roman et al. 2001)

Emergent Vegetation Sampling Methods

Each 1 m² vegetation sampling plot is sampled non-destructively for visual estimates of percent cover for each species and for unvegetated cover. A variety of methods for plant cover estimates are available. Here a percent cover estimate with 5% intervals is proposed. A reference cover guide such as that used for SeagrassNet monitoring (Short et al. 2003) can be developed to assist in standardization of cover estimates. Point intercept methods are also acceptable. In addition to cover estimates, shoot or stem densities and maximum canopy height is to be determined. If the vegetation is very dense, then the plot may be sub-sampled, and data on density need only be provided for the dominant or co-dominant species in each plot. It is understood that various emergent communities may require larger or smaller sampling plots than 1 m² to effectively sample the vegetation; therefore there is flexibility in how stem density and cover are measured, provided the methods are appropriate, justified, and described and committed to over the long term so that effective comparisons can be made both within and among sites in the study areas. More details on data collection and submission using a standardized spreadsheet format that is provided to CDMO are provided in Appendix 1. Groundwater may be sampled for salinity/conductivity, water level, and other ground water constituents as elective components of the study. Since groundwater measurements are many times critical to determinations of marsh function or change, inclusion of some component of groundwater sampling should be considered as part of the overall workplan.

Sampling should be conducted at least during the annual maximum biomass for the marsh plant community in the study area. All sampling should be completed within a two or three week interval, and should be conducted during low tide to minimize surface water effects if possible. In salt marsh areas this maximum biomass period may occur in late summer. In freshwater marshes the plant community may progress through several periods of changing species dominance and sampling may have to be repeated 2-3 times during the growing season (Doumlele 1981; Odum et al. 1984). If the growing season patterns are unknown for the study region, more frequent sampling should be undertaken during at least the first year of the monitoring to delineate the seasonal peak(s). Annual sampling may be initially repeated at 1-2 year intervals and subsequently at 3-5 year intervals depending on the system and the objectives of the study. If the study area is a restoration or impacted site, at a minimum the sampling should be repeated annually until the recovery period or rate of change slows to a pre-determined rate of change or some level of vegetation cover is reached (i.e., 100% or desired Restoration Performance Index relative to reference conditions). However, more frequent sampling may be conducted depending on the specific research or management questions that are being investigated. Modifications to this general protocol to make it more suitable for mangrove systems are provided in Appendix 5.

Submersed Aquatic Vegetation Sampling Design

Beds of SAV are typically found growing along an open shoreline of a bay, lagoon or tidal estuary or river. In most cases the study bed or vegetated area can be considered one segment. Transect placement for a SAV bed is similar to the emergent vegetation transect placement. It may be possible to continue the transect within the emergent wetlands into the submerged aquatic beds, however, in many cases,this might not be possible or logical if there are no adjacent emergent wetlands or only the SAV vegetation is being studied.

For the study of SAV beds alone at a particular site, individual transects are located by first dividing each study area into equal sized sections. One or two transects are randomly located within each section so that a total of three to five transects are established across the study area. Each transect should traverse the elevation gradient from the shoreline bank to the deepest edge of the bed. Transects should be located no less than 10m apart to maintain independence and should cover an area that is representative of the section. The determination of "representative sections can be done by use of aerial imagery or if this is not available, visual estimates of cover or SAV abundance from a boat using best professional judgment. The first permanent plot should be located at a random distance off a suitable benchmark on the shoreline. For example, if the vegetated zone is only 3 m wide then the zone is divided into five 60 cm intervals and the first permanent plot is randomly located at one of the five intervals. If the vegetated zone is 30 m wide then the first plot is located randomly within the first 10 m of the transect. If the transect is a continuation of the tidal marsh transect then the first plot location should be located off the initial emergent plot. Differences in scale of the width (shallow to deep, or upland to water edge) of the separate communities may

necessitate that the sampling plot intervals be different between the emergent and submersed habitats or portions of the transects.

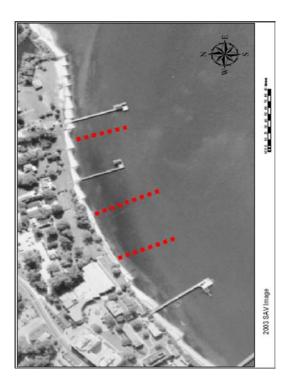


Figure 3. SAV sampling transects located across fringing SAV bed segment in the York River, Virginia.

As with the emergent community a total of at least 20 plots per submersed bed study segment are required for adequate sampling power, therefore the permanent plot intervals should be adjusted for the area of the bed to achieve this sampling replication. No permanent plots should be less than 10m apart. Because the location and depth of outer edge of a SAV bed can have important implications related to environmental conditions affecting bed dynamics, the location and depth of the outer bed edge should be delineated. As SAV abundance may gradually lessen with depth depending on the depth contour, the outer bed edge may have to be estimated. Typically, it is defined as less than 5% cover.

The permanent transects should be fixed by placing permanent PVC poles and/or stakes at intervals along each transect (see example for emergent wetland in Figure 2). A minimum of two to three poles and underwater stakes per transect should be established at appropriate intervals depending on the scale of the study area. If poles are not feasible then underwater stakes extending a suitable distance above the sediment surface should be established. In many systems stakes placed 50m to 100m apart and extending 25 cm above the sediment surface are suitable. In large systems, the transects may have to be established and marked using GPS, buoys or other appropriate techniques.

Each permanent sampling plot is offset approximately 1 m from each individual plot location along the transect. The sampling plots are located by stretching a 100 m or

longer plastic or fiberglass measuring tape or non-stretching line between the transect poles. If permanent poles cannot be established then temporary poles of PVC or other locally available material are established for the duration of the sampling.

Each plot is sampled non-destructively for percent cover by each species as well as unvegetated cover, within an approximate 0.25 m² area. (Note: SAV patchiness may require a much larger sampling area than 0.25m²). There is flexibility in the quadrat sampling size provided it is continued over time, so that effective time series comparisons can be made at the same scale in the landscape. However other larger or small scale sampling size area may be added if necessary if there are large changes in patchiness that cannot be adequately sampled using the established sampling quadrat. In addition to cover estimates, shoot or stem density and maximum canopy height should be determined within each plot for at least the more dominant or abundant species and study species of concern. Typically, canopy height is measured from where the plant emerges from the sediment surface to the top of the canopy. This is usually defined by the longest leaves of approximately 90% of the shoots if the plants are not positioned vertically. If a few shoots typically extend well above the canopy (ie. less than 10-20 % of the total in the case of reproductive or other type shoots) the canopy measurement made be measured to the height of the typical shoots (ie. approximately 90%) in the bed provided the methods are well documented and consistent. If the vegetation is very dense then the plot may be sub-sampled for density, height and leaf or shoot width as appropriate for the community (van Tussenbroek 1996; Short 1983; Phillips 1983). Macroalgae should also be included in the SAV assessments, for individuals that are attached within the plot; drift algae are considered too ephemeral to include. For species where density is not appropriate this measurement, such as some canopy forming poorly rooted freshwater SAV (e.g. Ceratophyllum demersum) can be omitted as "not applicable". There is flexibility in determining the most appropriate measure of metrics such as shoot height, provided such measurements are cited in the published scientific literature, well described in the study work plan, methods and metadata and do not preclude comparison of plant morphology in the study area with other study areas. More details on data collection and submission are provided in Appendix 1. An area reserved for elective sampling of other factors such as sediment nutrients, pore water sulfide, sediment deposition, etc. should be located at approximately 1m from the transect line point oriented 180° from the vegetation sampling plot. Voucher specimens including flowers, fruits and belowground material of each species and their various morphological variants should be sampled and appropriately preserved. Changes can be made to specific reserve sampling protocols over time if necessary, provided adequate ground truthing is provided to calibrate old vs. new methodologies. Documentation for any method change would be similar to that required for study methods that might be included in publication of the results of the study in the peer-reviewed scientific literature.

Submersed Aquatic Vegetation Transect Sampling Methods

Sampling should be conducted during at least the annual maximum biomass for the dominant SAV species in the study area although more frequent sampling can be undertaken as required or deemed necessary for the study objectives and documented in the methods and workplan. Annual sampling should be completed within a two-three

week interval if possible. In many seagrass areas this typically will occur from early to mid summer. In freshwater SAV areas this period may occur in late summer or early fall (Moore et al. 2000). In mixed species SAV communities such as those in the lower Chesapeake Bay (Orth and Moore 1986) there may be seasonal dominance of one species (*Z. marina*) in early summer and another (*R. maritima*) in the late summer and two samplings may be required. If the growing season patterns are unknown for the region of interest, initial monthly or other more frequent sampling may be required to delineate the season peak(s). Since SAV can be subject to significant year-to-year variability in abundance, at least annual sampling should be conducted. If the study area is found to be consistently stable then monitoring intervals can be extended to 2-3 years or more. More frequent sampling (annually or monthly) may be conducted, as deemed necessary, to evaluate the level of SAV abundance changes required for a particular study.

A complementary approach to SAV sampling that can optionally be implemented by the NERRS is the SeagrassNet protocol, summarized in Appendix 3.

NERRS System-wide Sampling Consistency

The objectives of the NERRS-SWMP vegetation monitoring sampling are to both evaluate reserve specific questions as well as to support regional, national, sentinel site and climate change objectives. In that regard, while some latitude in site specific monitoring is provided above, the application of slightly differing approaches should not preclude use of the individual reserve's data for comparisons across various reserves or regions. In fact, this is the strength of the program that necessitates these protocols. In that regard reserves should try to coordinate timing of sampling and their methods of assessment with other reserves with similar communities to facilitate these comparisons. One way to facilitate this is to provide opportunities for the sharing of scientific methods, results at sector and national meetings as well as providing specific time for scientific interactions relative to any questions or problems that arise among the reserves conducting biological vegetation monitoring. It is primarily through these scientific interactions and the individual and collective reserve system experiences that the optimum and most effective sampling protocols can be developed. These protocols should be regularly re-visited and modified as necessary to improve clarity and systemwide consistency. Appendix 1 clarifies required elements for system-wide consistency in data collection and submission.

Data Analyses

Repeated measures analyses or other parametric approaches are typically used to evaluate changes in plant metrics over time and among sites. Non-parametric statistical tests can be used to evaluate similarities of vegetation communities between sites or over time (Roman et al. 2001). Ordination techniques and similarity indices or other univariate, graphical and multivariate approaches (e.g., PRIMER-E, Plymouth, UK) as well as single or multiple regression techniques can also be used to develop and test hypotheses relative to community structure or relationships between vegetation communities as well as other environmental factors.

Science Implications

The NERRS vegetation sampling protocols are based upon established, peer reviewed and published protocols and they are consistent with other ongoing emergent and submersed monitoring programs. The spatial design and sampling intensity is appropriate for long term monitoring for comparing specific study areas over time. Some illustrative general questions with specific examples of studies at the reserve level that have already been and published in the peer-reviewed scientific literature using these protocols are: What is the change in non-impacted habitat, degraded or recovering habitat over time (Kennish et al. 2008)? How do impacted and un-impacted vegetation communities compare (Jarvis and Moore 2010)? What is the effect of invasive species on the native plant community (Nieder et al. 2004)? What factors are related to observed vegetation changes (Moore and Jarvis 2008; Moore et al. 2013)? How is relative sea level rise and/or other global climate change stressors affecting representative vegetation areas in the reserves (Edmiston et al. 2008)? Are there consistent changes in vegetation communities among different (i.e spatially located) study areas within the NERRS (Rumrill and Sowers 2008)?

Management Implications for NERRS

Quantification of habitat change both within and among the reserves in the reserve system is an important NERRS goal (NERRS 2002). In addition, the developing strategy and framework for NERRS restoration efforts requires consistent, "scientifically-based" monitoring studies that can be applied similarly to natural, impacted and restored sites, so that the effectiveness of habitat restoration as well as quantification of cause and effect relationships can be measured. Additionally, accurate evaluation of change in coastal vegetation systems at the national and international level requires that consistent methodologies be applied so that any broad trends can be more clearly determined. Evaluating patterns of non-native and invasive species expansion across broad regions also requires a consistency of approach. The emergent and submersed vegetation monitoring approach proposed here will address these and other management objectives.

Data Submission and Archiving

Data will be submitted to CDMO after each funding period as stipulated in the individual workplans using agreed upon NERRS system-wide spreadsheet templates. Submitted data will include site, location, and replicate identifications as well as basic parameters including species composition, cover, shoot height, and density. Metadata

should be submitted using the required template (Appendix 2). The submission templates will allow comparison across reserves. Elective elements (e.g., archival photographs; measurements of explanatory variables) will not currently be submitted to CDMO, but the potential for this will be explored if future funding becomes available to support the CDMO for biomonitoring submissions. Data will be reviewed by members of the biomonitoring workgroup, and sites will be required to revise data as needed to meet approval by the workgroup. Use of the basic information for publication beyond that undertaken by each individual reserve that generates the data should be undertaken after coordination with responsible reserve personnel. Data usage and publication should follow typical scientific standards for citation and authorship.

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Appendix 1. Guidelines for vegetation data collection and submission.

The following descriptions are provided in order to clarify requirements for consistency in data collection and submission among different monitoring sites.

PERCENT COVER CATEGORIES

<u>Vegetation percent cover</u>: percent cover of all live species found in a plot should be estimated if possible (e.g using 0.1% as an arbitrary value for rare species); sites using point intercepts may use R instead of numeric value to designate rare species seen in quadrat but not encountered with intercept, explaining this in the metadata; other descriptors for rare species cover (e.g. <3 stems) should not be used; percent cover for a single species should never be above 100%; if known, full genus and species name of each species should be listed in the database (use genus only, or "unidentified" if not known).

<u>Algae</u>: Any attached algae (attached to sediment or to emergent and submerged aquatic vegetation) should be included in assessment; naming should be as specific as possible (species level ID, genus level, or division "green algae" or even just "algae").

<u>Unvegetated cover</u>: If any unvegetated cover is present then this must be quantified as well. (Note that long dead vegetation should be considered "unvegetated" cover, but for annual plants, dead tissue that was clearly alive earlier in the current growing season can be counted as the appropriate species in the live vegetation percent cover estimate. If this is done it should be described in the metadata). Sites may further break down unvegetated cover categories for local site use (e.g., bare ground, dead vegetation, duff, wood, channel), but these subcategories should be lumped as "unvegetated" for submission to the national database (the metadata can explain what subcategories were lumped).

<u>Wrack</u>: The recommendation of the biomonitoring workgroup is to remove canopy wrack that is on top of live vegetation, so the live vegetation underneath it can be assessed. If there is no live vegetation under the wrack, this portion of the quadrat is quantified as "unvegetated cover." The wrack should then be replaced on the plot to allow for relatively natural dynamics. "Canopy wrack" can be included as a category in the database (its percent cover would of course be assessed prior to removal). In these cases total cover in the quadrat would always be >100% (total of vegetated + unvegetated cover under wrack will always total at least 100%, and canopy wrack % cover would be additional on top of that).

Some Reserves prefer not to remove wrack, so as not to interfere with natural dynamics. These Reserves may report only on the "canopy wrack" category, where it is obscuring vegetation underneath, and not provide percent cover estimates for the vegetation or unvegetated cover in the plot. This is approach is allowable but not recommended.

Other explanatory variables: Sites may wish to record data on various potential explanatory variables associated with the plots, such as porewater or sediment type. If

funding is available in the future, we will explore the possibility of entering such ancillary variables in the CDMO database as well.

PERCENT COVER METHODS

Each site should choose the most appropriate method for generating estimates of percent cover. Both, visual estimates and point-intercept measurements (converted to percent cover) are allowable, but the method must be clearly described in the metadata.

If necessary, sites can also use a combination of visual estimate and point intercept methods; e.g., Chesapeake Bay MD has found it useful to first estimate bare ground by visual estimate, then estimate vegetation cover by point intercept.

Sites that are developing vegetation monitoring protocols will confer with other NERR sites that have similar vegetation types and are already conducting vegetation monitoring at annual sector meetings, so that they can use identical or nearly identical methods. This will allow for much more accurate comparisons at a regional scale (e.g., nearby NERRS should try to standardize) or for similar vegetation types (e.g., tidal freshwater marshes should try to standardize). Eventually it would be helpful to compile a master chart summarizing methods and seasonal timing used at all sites, to allow for easy comparisons.

PERCENT COVER CALCULATIONS AND OVERLAPPING CANOPY LAYERS

For vegetation that occurs in overlapping canopy layers, percent cover values for all species summed together may add up more than 100%. For instance, Species A (low marsh plant) may have 80% cover near the ground, while Species B (tall branching forb) may have 50% cover in a higher canopy layer. For point intercept data, simply calculate percent cover as follows:

Percent cover of Species A = (# intercepts with Species A / total intercepts assessed)* 100

For example, if you assess 50 intercepts per quadrat, and 40 of them hit Species A on the way down and 25 of them hit Species B on the way down and 5 of them hit nothing at all on the way down, then you will enter 80% cover of Species A, 50% cover of species B, and 10% "unvegetated".

Reserves using visual estimates of cover may choose to "force" the total of all vegetated cover plus "unvegetated" cover to equal 100%, i.e. not to capture multiple canopy layers that can result in cover >100%. Such choices must be well-documented in the metadata.

Note that "unvegetated" is only scored if there is zero live (or recently alive, for annual plants from earlier in the season) vegetation at any canopy layer above that spot.

DATA ENTRY OF PERCENT COVER CATEGORIES

Reserves may choose to only submit data rows with non-zero cover values, e.g. reporting only species (and unvegetated cover) that occurred in each plot. Alternatively, Reserves

may provide complete species lists (e.g. of all species found at a particular site in all monitoring years to date) for each plot, e.g. including many rows with values of zero. The latter approach will facilitate analyses of Reserve datasets by end-users in their current form, and is thus recommended so long as the burden on the site is not too great (very feasible for sites with short species lists, as with most SAV monitoring). Eventually, the goal is to merge all datasets into a sophisticated database that would fill in complete regional species lists and generate those zero rows for all sites.

STEM DENSITY AND CANOPY HEIGHT MEASUREMENTS

These can be time consuming for sites with very dense or diverse vegetation. They are nevertheless a required component of the NERRS protocol. Figure out a way to make it feasible – for instance, subsample using much smaller quadrats (e.g., 10 x 10 cm) to do density counts, and/or conduct density estimates only for the dominant species in each quadrat.

It is critical to document clearly in the metadata how these measurements were made, particularly if you subsample. Some sites locate their subsamples uniformly (e.g., bottom right corner of larger quadrat each time; NBNERR), some randomly, and some place them in regions of the quadrat they consider representative for each separate species assessed. These methods would each yield very different results, so it is critical to document what you have done to allow for repeatability over time. Again, ideally please develop methods in collaboration with other regional sites or sites with similar vegetation, so that meaningful statistical comparisons can later be made.

DATA REVIEW AND POSTING

Data will be submitted to the CDMO. Data submissions will then be reviewed by at least one member of the Biomonitoring Workgroup. Both data and metadata will be reviewed. It is likely that issues will be detected with virtually all datasets, so submitting sites should allocate time for data revision and further processing in response to requests by reviewers. Data will then be re-submitted to the CDMO and reviewed again. As needed, further rounds of revision will occur, until the reviewer(s) approve the data and the submission is considered final.

Approved datasets will be made available (along with metadata) on the CDMO webpage.

NATIONAL DATABASE

To allow for meaningful regional or national syntheses of the vegetation monitoring, the individual Reserve datasets should eventually be compiled into a master database.

This will require substantial effort, to ensure consistent species names are used across sites, and that taxonomic revisions are consistently incorporated into past datasets. It will also be challenging to combine different datasets with different species listed (for instance may need to list all species found anywhere nationally for each plot, and put zeros for most of them). Ideally, the data submission templates would be designed a priori to make such later syntheses smooth and rapid. For instance, the National Park Service has an Access database for vegetation monitoring that makes data submission and

synthesis very consistent and smooth. However, setting up such a system takes resources that are not currently available to the NERRS. So we are beginning with this piecemeal approach, with datasets made available separately for each site and year.

In the future, if biomonitoring funding is available, some funding should be allocated to a one-time effort to design a workable national master database for the NERRS vegetation data, and to conduct the first synthesis of the data. This would be a large effort, requiring RC and CDMO participation as well as work by a contractor or postdoc. Subsequently, once the database is set up, much less effort would be required for annual updates (adding new species or changing names in accordance with the International Taxonomic Information System. Setting up simple macros to look for common errors would also improve reviewing and quality of the data.

Appendix 2. Metadata template

Reserve Name (include 3 letter code here) NERR Vegetation Monitoring Metadata Months and year(s) the documentation covers

Latest Update: Date that the last edits were made

I. Data Set and Research Descriptors

- 1) **Principal investigator(s) and contact persons** List the staff members responsible for the design, implementation and continuation of the data set. Include name, title, mailing address, phone number, and email address for the Research Coordinator, SWMP or field technicians, and person(s) responsible for data management.
- 2) Entry verification This section explains how the data were verified (QAQC'd) before being sent to the CDMO to be archived into the permanent database. Specifically, list how your data are acquired, validated, processed, and archived. Mention how your Reserve deals with outliers, unidentified vegetation, creating numerical data from categorical data, etc. Remember to list the person(s) responsible for data management.
- 3) **Research objectives** Describe briefly the nature of the monitoring program resulting in this data set (for example, control versus impacted site, long term monitoring, spatial or temporal coverage, etc.). Describe the goal or purpose of this research. Include the date that monitoring began.
- **4) Research methods** Detail the specifics of your experimental design and sampling methods. Be sure to include explanations detailed enough to allow your methods to be repeatable in the future, including explanations of:
 - What method was used to estimate percent cover (point intercept vs. visual estimate?)
 - Whether percent cover of all vegetation plus unvegetated cover was "forced" to total 100%, or whether multiple canopies were assessed, sometimes resulting in >100% cover
 - How rare species cover was estimated, if different from other estimates (e.g. R assigned to species seen in plot but not encountered in point intercept)
 - How maximum canopy height was determined
 - How stem density was measured; if subplots were used, how these were located (uniform, haphazard, random) and where, as well as what size?
- **5**) **Coded variable definitions** Define site IDs, transect IDs, and plot IDs. You may define your coding scheme if applicable, but where necessary, define each individual code.

SiteID TCE = Chesapeake Bay Virginia Taskinas Creek East

TransectID Fred = the easternmost transect at Site TCE. Please describe the vegetation type for each transect broadly (e.g., SAV, salt marsh, tidal freshwater marsh, mangrove), so that similar vegetation types can readily be compared in regional or national analyses.

PlotID = numbered 1-X, with plot 1 located at the beginning of the transect and each subsequent plot located 10 meters (or at other appropriate intervals) down the transect. For each plot, please describe further subdivisions of vegetation type as appropriate to guide future analysis comparisons, e.g., "muflat-marsh ecotone,"", "low marsh,"", "high marsh," "", marsh-upland ecotone,"", "upland above marsh"). We recognize that different sites may use different classification schemes, but at least having some level of categorization will allow for regional or national analyses to compare similar vegetation types.

- 6) Site location and character Describe your NERR site in general and the sampling sites associated with the vegetation monitoring effort. <u>Include the following</u> in your description for EACH sampling location. If certain characteristics apply to all sample sites or the entire Reserve they may be discussed in an overview. You may also include a map of your transect locations here (not required).
 - a) habitat type(s)
 - b) tidal range
 - b) salinity range
 - c) type and amount of freshwater input
 - d) pollutants in area
 - e) description of watershed draining to the site
- 7) **Species information** At a minimum list scientific name for each species present at your sampling sites. You may include additional information such as plant vs. algae, family, common name, native/invasive/cryptogenic status, etc.
- 8) **Distribution** This section will address data ownership and data liability by including the following excerpt from the Ocean and Coastal Resource Management Data Dissemination Policy for the NERRS System-wide Monitoring Program in the metadata.

NOAA/ERD retains the right to analyze, synthesize and publish summaries of the NERRS System-wide Monitoring Program data. The PI retains the right to be fully credited for having collected and processed the data. Following academic courtesy standards, the PI and NERR site where the data were collected will be contacted and fully acknowledged in any subsequent publications in which any part of the data are used. Manuscripts resulting from this NOAA/OCRM supported research that are produced for publication in open literature, including refereed scientific journals, will acknowledge that the research was conducted under an award from the Estuarine Reserves Division, Office of Ocean and Coastal Resource Management, National Ocean Service, National Oceanic and Atmospheric Administration. The data set

enclosed within this package/transmission is only as good as the quality assurance and quality control procedures outlined by the enclosed metadata reporting statement. The user bears all responsibility for its subsequent use/misuse in any further analyses or comparisons. The Federal government does not assume liability to the Recipient or third persons, nor will the Federal government reimburse or indemnify the Recipient for its liability due to any losses resulting in any way from the use of this data.

Also <u>include the following excerpt</u> in the metadata which will address how and where the data can be obtained.

NERR vegetation monitoring data and metadata can be obtained from the Research Coordinator at the individual NERR site (please see Principal Investigators and Contact Persons), from the Data Manager at the Centralized Data Management Office (please see personnel directory under the general information link on the CDMO home page) and online at the CDMO home page http://cdmo.baruch.sc.edu/. Data are available in comma separated value format.

- 9) Associated researchers and projects (link to other products or programs) Describe briefly other research (data collection) that correlates with or enhances the vegetation monitoring data. At a minimum, mention the SWMP abiotic data sets.
- **10**) Other remarks/notes Use this section for further documentation of the research data set. Include any additional notes regarding the data set in general, circumstances that resulted in missing data, or any deviation from the SOPs

Appendix 3. SeagrassNet based sampling: an optional complement to NERR submerged vegetation monitoring protocols.

SeagrassNet is a global monitoring program developed to investigate and document the status of seagrass and SAV resources worldwide. The SeagrassNet protocol (Short et al. 2002) may be considered as an optional additional component of NERRS submersed vegetation monitoring. This specific approach will provide additional detailed, complementary information relative to the structure of the vegetation communities at shallow-edge, mid-bed and deep-edge areas. In addition, it permits detailed comparisons of the status and trends of submersed vegetation communities in the NERRS reserves with that of other groups studying long term trends in seagrass and other SAV communities world-wide. The SeagrassNet program started with a pilot study in seven countries of the Western Pacific in 2001 and is now expanding to other countries in North America, Europe and Africa. Its purpose is to develop a network of intensive monitoring sites linked via the World Wide Web by an interactive database (www.seagrassnet.org). The monitoring component consists of a science oriented monitoring program that is based on specific standardized monitoring protocols (Short et al. 2002). The general approach of this detailed monitoring protocol is to establish three, permanent, 50-m wide, cross-transects that are oriented parallel to the shoreline near the inner edge, middle and outer edge of the SAV bed. The center of the transects would be located along a perpendicular transect located as described (Figure A1-1) in the NERRS SAV monitoring protocol.

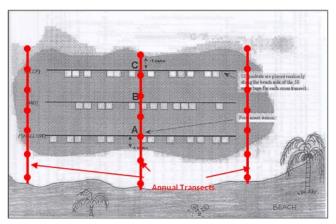


Figure A1-1. Example of Permanent Transects and SeagrassNet Transects (modified from Short et al. 2002).

The SeagrassNet sampling is generally repeated quarterly (every three months). Two weeks prior to each sampling a Hobo® (Model 8-004-02; MicroDAQ.com, Ltd) light logger or comparable remote monitor is deployed at the midpoint of each of the three cross-transects, and an additional logger is placed on shore above the high tide elevation. Six quadrats are located randomly along the 25 m cross-transects to the left and right of the centerline. Sampling of each 0.25 m² quadrat consists of: vertical photographs using disposable cameras; visual % cover; water depth and local time; canopy height for dominant species; evidence of grazing recorded; flower and fruit counts. A biomass core is sampled 0.5 m adjacent to the quadrat. The vegetation is

separated into leaves, sheaths and stems, and belowground material. The shoots are counted by species. Dry weights are then determined on all components. Triplicate, small (20 cc syringe) sediment cores are sampled from the mid-point of each cross-transect for organic content and grain size (Short and Coles 2001). The distance to channelward seagrass edge (continuous meadow) and distance to last shoot (most offshore) and distance from the shore to the shallowest edge is measured to evaluate any bed migration. Voucher specimens of each species identified including shoots, flowers and fruits (if available) and belowground material should be collected and appropriately preserved. Duplicate specimens would be sent to SeagrassNet where they are currently housed in a special collection at the Smithsonian Institution.

Currently individual SeagrassNet participants send all voucher specimens, field photographs and field data to the Jackson Estuarine Lab for data summarization, further sample processing, QA/QC and archiving. Field data are entered directly into the web. All other processing is done locally. For NERRS, one reserve could serve the archival function as well as serving as the focus of the interactive database. Data QA/QC would be done locally and then sent directly to CDMO for archiving and in a manner similar to the current nutrient monitoring data.

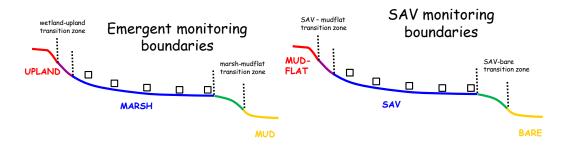
Appendix 4. Monitoring ecotone boundaries: an optional complement to NERR vegetation monitoring protocols.

Introduction

Ecotones are transition zones between adjacent ecological systems (Risser 1995). Typically, they are narrow zones with steep environmental gradients located between extensive systems with more stable environmental conditions. Ecotone boundaries may be particularly responsive to environmental changes, because species are living near the edge of their tolerances (Peters et al. 2006). Thus, ecotones may serve as sensitive indicators of global climate change (Risser 1995).

As global climate changes, and in particular, as sea level rise accelerates, the wetland-upland boundary is expected to migrate landward in many estuarine ecosystems. Likewise, other boundaries (Fig. 1), such as between submerged vegetation and intertidal mudflats, or between intertidal mudflats and emergent vegetation, may migrate landward. However, such changes are complicated by shorter-term and regional oceanographic drivers of water levels, such as ENSO. Optional elements can be added to the NERRS vegetation monitoring protocol to detect responses of the ecotone to short-term and longer-term changes in water level and other factors, such as invasions or restoration. The two elements below are simple to implement, either separately or in combination, and do not require much additional investment beyond the regular required sampling. Nevertheless, they are sufficient for basic characterization of boundary changes.

Figure 1. Types of boundaries that can be monitored to supplement the regular vegetation monitoring transects (shown in blue).



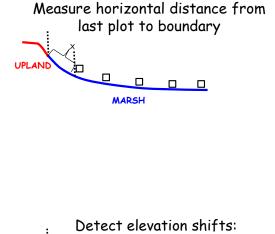
Measurements of the location of vegetation boundaries

One approach to ecotone monitoring is to quantify the location of vegetation boundaries during each sampling period, so that their migration can be tracked over time.

A straightforward approach to this at sites with relatively narrow ecotones (<100 m) is to simply take a transect-tape measurement from a permanent marker to the boundary on the landward and/or seaward edge of each permanent transect (Fig. 2, left). For instance, the PVC stake marking the most landward or seaward plot on the permanent

transect can be designated as the marker (or if that is not convenient, a separate marker can be placed). The location of the ecotone boundary can then also be marked with a PVC stake. Having at least two permanent markers allows for consistent repeat monitoring, because the angle at which the transect tape should be run is specified (start at the same permanent marker on the transect, then run tape to last year's boundary marker, then keep going as needed). New markers can be added annually to extend this transect if the ecotone is wide; at narrow ecotones, the original boundary marker may be sufficient to document the angle of measurement.

Figure 2. Measuring distances to boundaries. Left: Measurement of horizontal distance (X) from last plot on vegetation transect to vegetation boundary; in this case, boundary is wetland-upland boundary. Right: Measuring the vertical elevation of boundaries; in this case Y represents the vertical change in elevation from the originally measured and marked boundary to the current one.



measure elevation of boundary

At sites with very wide ecotones spanning many kilometers, visual sighting of markers and measurements by transect tape may not be feasible. Documentation of the location of the boundary in each monitoring year could be accomplished by GPS navigation, following the same direction of the transect line until the boundary is reached, and then marking a waypoint at that location. Such field GPS monitoring could be complemented or replaced by remote sensing at sites where boundaries can be clearly distinguished in aerial imagery.

A critical requirement for consistent documentation of boundaries is development of a repeatable definition, which will be similarly applied by different workers. At some sites, this may be very simple. At Elkhorn Slough NERR, the definition used for the

marsh/upland boundary is "the most seaward location in the transect where 100% of the vegetation cover is by upland species". However such a definition might be more complicated to apply at sites where there are multiple transition zones between different wetland types (e.g. saltwater to brackish to fresh), where the ecotone is very wide, or where the vegetative community landward of the estuary contains substantial representation by wetland plants. Different sites may thus need to develop separate definitions. These definitions should be documented in the same file as the data.

The location of the boundary may vary seasonally at some sites. To ensure consistency in monitoring, measurements should always be taken at the same time of year. Related to seasonality, it is important for the definition of the boundary to include consideration of live vs. dead plant material. Using the "100% upland cover" definition seems simple. But if there is a dead marsh plant in a plot that otherwise has only live upland vegetation, is that 100% cover? The definition must provide guidance, for instance, "vegetation that is clearly recently dead, persisting from the last growing season, should be included in the cover estimate, while vegetation that is long dead, more than 1 year old, should not be included".

In addition to measuring from a permanent plot marker to the landward and seaward vegetation boundaries, measurements to other vegetation indicators of interest can also be taken at the same time. For instance, both the seaward and the landward edge of the ecotone can be monitored to track ecotone width, at sites with a relatively homogenous and narrow ecotone. At sites with multiple transitions between different communities, the boundaries of each can be marked and monitored. Or the locations of the landward or seaward boundaries of distribution of single species of interest can be monitored. For instance, the distance to the most seaward representative of an upland weed of concern along the permanent transect can be measured, to determine whether the species is moving down into the wetland over time. Or the distance to the most landward representative of a particular threatened marsh species can be measured, to determine whether it is successfully able to track sea level rise in future years. Again, such measurements are simple, but require explicit definitions that are well-documented.

The above method of documenting the location of the boundary (and other vegetation landmarks) involves horizontal measurements of the location relative to a permanent marker. This allows of detection of lateral migration of the boundary. For instance, it may be X m from the designated permanent plot marker in Year 0, but X+Z m in Year 5. Such lateral measurements can be complemented by monitoring of vertical elevation changes (Fig. 2, right). This could be done by simple surveying of relative elevations using a theodolite and stadia rod, or with RTK GPS to obtain absolution elevations.

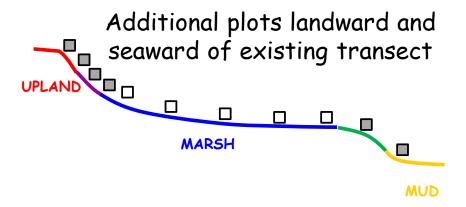
In areas with relatively homogenous or limited rates of elevation change (due to sedimentation, subsidence, or uplift), elevation measurements can also be made retrospectively. For instance, at Elkhorn Slough NERR, sedimentation rate and elevation changes are low in the high marsh – upland transition zone. The location of the wetland-upland boundary for a decade of past monitoring was reconstructed by re-measuring horizontal distances from the seaward permanent marker to find old boundary locations;

these old boundaries were then surveyed for elevation. This was helpful because the high investment of elevation monitoring was undertaken only once, after interesting lateral migration patterns of the boundary had been observed. The retrospective elevation monitoring, while perhaps not entirely accurate, enabled reconstruction of the elevation changes involved with the observed lateral migration (Wasson et al. 2013).

Additional permanent plots to characterize the ecotone and changes near boundaries

Another complementary approach to detecting and quantifying vegetation changes at boundaries is simply to add additional permanent plots on the landward and seaward portion of each permanent transect, so we are poised to detect changes to these areas over the coming years (Fig. 3). All the same parameters should be assessed as in the regular vegetation monitoring, allowing both community composition changes and changes to particular species (in biomass or density) to be detected. At the emergent-upland boundary, this will require field crews to be able to identify upland plant species as well as marsh species. If this proves challenging, one option is to limit species identifications to marsh plants, and to lump upland species into broad categories ("grass", "shrub", etc.). Since the emphasis of the monitoring program is on estuarine vegetation changes, not on upland communities, this lack of specificity may be acceptable for those sites where upland identifications would pose a barrier to implementation of the monitoring.

Figure 3. Adding plots on either end of the regular vegetation transect. In this example, just two were added at the seaward end, one in the transition zone and one beyond it. At the landward end, due to predicted changes and rapid changes over small areas, multiple, closely spaced plots were added.



The number of additional permanent plots to be added will vary based on site characteristics and management concerns. At a minimum, we recommend adding two

additional permanent plots at both the landward and seawards ends of each permanent vegetation transect. One of the additional plots should be located within the current ecotone (transition between mud/SAV, mud/emergent, or emergent/upland). The second additional plot should be located an appropriate distance beyond the first, using the judgment of the person designing the transect. The goal would be to locate this second plot in an area where the boundary might reasonably migrate within the next 10 years. For instance, for the wetland-upland boundary, additional transects might be set up within a zone extending 10 cm higher than the current boundary, to accommodate the high end of projected sea level rise combined with impacts of increasing storm surges. At sites where ecotone migration is a high scientific or management concern would benefit from including more than two additional plots to characterize boundary changes. As needed, additional plots can also be added in future years, if boundary changes occur more rapidly than anticipated. Knowledge of groundwater hydrology is helpful for establishing wetland-upland boundary sampling design, as hydrology will influence salinity patterns and migration rates.

The spacing of the additional plots may be similar to those along the rest of the transect, or may be lower or higher, as appropriate for site issues. For instance at Elkhorn Slough NERR, the marsh plain has a relatively gradual slope, while the ecotone and the uplands above it have a steep slope, such that the ecotone zone is only a few meters wide, while the marsh is hundreds of meters wide. At such a site, keeping the same spacing of plots as in the marsh would not sufficiently characterize the rapid changes occurring in this narrow boundary zone. In this sort of steep ecotone, it makes sense to add multiple closely spaced permanent plots that span the current ecotone and the zone above it. In contrast, at other sites, such as some of the Gulf Coast NERRs, the ecotone slope is very gradual, and the transition zone spans many kilometers. At such a site, additional permanent plots directed at detecting boundary shifts could be very widely spaced.

For sites where there are trees at the ecotone-upland boundary, modifications to the sampling protocol may be necessary. Quadrat sizes or distribution may need to be different to accommodate large or dense vegetation such as is found in some forested areas.

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Appendix 5. Mangrove monitoring: an optional complement to NERR vegetation monitoring protocols.

Introduction

In general, the term "mangrove" refers to an assemblage of tropical trees and shrubs that grows in the intertidal zone. Mangroves include approximately 16 families and 40 to 50 species (depending on classification). Mangroves are tropical species that are not generally tolerant of freezing temperatures. Their latitudinal limits worldwide vary depending on air and water temperatures, but the majority of populations occur between latitudes 30° North and 30° South (Waisel 1972; Sherrod & McMillan 1985; Sherrod et al. 1986; Tomlinson 1986). Mangrove communities are particularly sensitive to many of the factors that are associated with climate change and other changing coastal conditions (e.g., sea level rise, freshwater inputs, sedimentation, storms, etc.) and therefore their long-term monitoring is important to both individual reserves and the reserve system overall. This is especially important for reserves located at the mangrove - salt marsh ecotone, where mangroves may be expanding northward resulting in significant changes in dominant emergent vegetation communities.

Mangroves often exhibit complex zonation patterns in many coastal areas. For example, many areas in Florida and the Caribbean show *Rhizophora mangle* (red mangrove) occupying the seaward zone, followed by *Avicennia germinans* (black mangrove), and *Laguncularia racemosa* (white mangrove) in the most landward zone. A number of factors have been proposed to explain this zonation including: the process of land building or other geomorphological processes and plant succession; differential dispersal, sorting or predation of propagules across the intertidal gradient; differing physiological constraints affecting plant growth and survival across the gradient; and plant interspecific competition (Feller and Situik 1996). In addition to horizontal spatial patterns, mangroves also exhibit vertical stratification across the supratidal, intertidal, and subtidal. The supratidal stratum includes the arboreal portions of the forest. The intertidal stratum extends from the high to low water tidal heights and encompasses the aerial root systems of the mangroves and peat banks. The subtidal stratum occurs below the low water mark includes mangrove roots and peat banks.

Sampling Methods

General methods for measurement of mangrove ecosystem structure and function have been previously described by Lugo and Snedaker (1975), Pool et al. (1977), Snedaker and Snedaker (1984), Cintron and Novelli (1984), and Elzinga et al. (1998). Much of this work is built upon and summarized in the CARICOMP Methods Manual, Levels 1 & 2 (2001). The sampling protocol presented in this appendix follows much of that described in CARICOMP (2001), with some modifications to align mangrove sampling with that of the NERRS emergent and submersed vegetation protocols, parts of which will be followed in this appendix. The goal of sampling should be to measure community composition and abundance changes over time, including having sufficient measurements

across any appropriate gradients, as has been previously described for emergent marsh and submerged vegetation communities.

The study area of interest should be delineated and the boundaries defined a priori. Control or reference sites in each study area are first identified as areas that have historically not been markedly impacted by natural or anthropogenic factors. The areas should also be representative of natural mangrove communities in the region. The focus of this detailed vegetation monitoring can vary with the particular circumstances or goals for each study (e.g., sampling design may be different at the mangrove - salt marsh ecotone as compared to tropical mangrove forests). A general base map should be developed providing the fundamental features of the site. The degree of detail of this reference map will be dependent on the extent of the geographical detail of the region.

Within the reserve, potential study areas should be identified, preferably at least two sites, and must be of appropriate scale to address the general (system-wide) and specific (individual reserve) objectives of the study. Once sample sites are identified, at least two transects should be established per site, each spanning from the marsh edge to the upland edge (see Figure A3-1).



Figure A3-1. Two transects spanning from the marsh edge to the upland edge (approx. 570 m) established at a sample site. Five whole plots (10 x 10 m) are distributed evenly along each transect.

For each transect, five permanent 10 x 10 m "whole plots" will be established evenly along the transect (i.e., 1 on each end then 3 evenly spaced along the transect; Figure A3-2). Thus, at a minimum each sample site will have 10 whole plots. Whole plots, numbered from the marsh edge (#1) to the upland edge (#5), will be marked with PVC poles on each corner and GPS locations will also be recorded for each corner. Within each whole plot, five permanent 1 x 1 m "sub-plots" will be established at even intervals within the whole plot (Figure A3-2), thus at a minimum there will be 50 sub-plots at each sample site. Sub-plots will be demarcated with PVC poles on each corner and will be numbered starting with #1 at the corner closest to the marsh edge along the transect, then continuing clockwise ending with #5 in the middle of the whole plot.

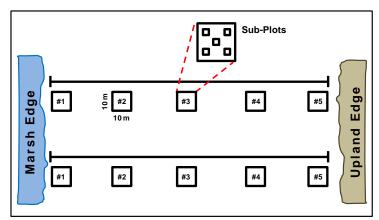


Figure A3-2. Layout of whole plots and subplots along a transect.

Mangrove community dynamics will be examined at three scales (whole plot, sub-plot, and individual trees) along each transect, with specific metrics identified for each level (Figure A3-3). Together, these multi-scale metrics will provide comprehensive information on the mangrove community, and should be attainable under various sampling resource scenarios (e.g., not too labor intensive or expensive). This division of examination also helps to ensure that the sample plots (both whole and sub) will be minimally disturbed and won't get destroyed during sampling. Most importantly, this three-pronged approach ensures that important community characteristics aren't overlooked. For example, large trees aren't often captured in the sub-plots, but the high number of shoots and small trees (< 0.5 m) in some areas prohibits examination of every individual mangrove within the entire whole plot. Thus the sub-plot measurements combined with those from individual trees more accurately reflects the mangrove community composition. Specific metrics are described for each level as follows:

Whole plot - General characteristics of each of five whole plots along the transect will be measured including percent cover (mangrove spp. vs. other) and soil porewater salinity and temperature (both measured at the time of collection in the field).

Sub-plot - For each sub-plot percent cover (mangrove spp. vs. other) will be measured. Mangroves will be identified by species and stage (shoot = no branches; tree =has branches), then trunk diameter (mm; just above the sediment, ~ 2 cm) and canopy height (cm) will be measured for each individual in the sub-plot. Thus for each sub-plot there will be information on total mangrove count, the abundance of shoots and trees, and the sizes of each.

Individual trees - To track individual trees over time within each whole plot, 10 large trees for each mangrove species present will be tagged and a suite of measurements will be taken to examine tree architecture. Ideally, the 10 largest trees for each species will be selected so as to be easiest to track over time. A suite of metrics designed to assess tree architecture will include:

- 1. Canopy Height (cm) Height of top of canopy
- 2. Trunk Formation Single or multiple trunks

- 3. Trunk Diameter (mm) Diameter just above sediment (~ 2 cm); if multi-trunk, then measure largest trunk
- 4. Clear Height (cm) Height from sediment to first branch
- 5. Canopy, Wide Axis (cm) Canopy width at the widest point
- 6. Canopy, Narrow Axis (cm) Perpendicular to the wide axis, canopy width at the widest point
- 7. Canopy Offset (cm) The horizontal distance between the trunk and the intersection of the canopy wide and narrow axes
- 8. Ground Cover Species present in the area under the tree canopy

Together, these tree architecture metrics will help assess how individual trees are changing over time. This will be especially important in ecotone areas where mangrove ranges are expanding northward (i.e., winter dieback could be detected with canopy measurements).

Sampling should be conducted at least during the annual maximum biomass for the mangrove community in the study area. All sampling should be completed within a two-three week interval, if possible, and should be conducted during low tide to minimize surface water effects. Depending on the specific study objectives, some metrics may be sampled at greater frequency throughout the year.

WHOLE PLOTS	SUB-PLOTS	TREE ARCHITECTURE
(10 x 10 m)	(1 x 1 m)	(Individual Trees)
Percent Cover Temperature (Soil Porewater) Salinity (Soil Porewater)	1. Total Count - Mangrove Spp 2. Form (shoot vs. tree) 3. Trunk Diameter 4. Canopy Height 5. Percent Cover	 Canopy Height Trunk Formation Trunk Diameter Clear Height Canopy – Wide Axis Canopy – Narrow Axis Canopy Offset Ground Cover
n = 5	n = 5	n = 10 ind / spp
per transect	per whole plot	per whole plot

Figure A3-3. Summary of metrics examined at each level.

Future improvements to this protocol

NERR implementation of vegetation monitoring in mangrove systems has only just begun. In coming years, the protocol will likely be refined and updated with lessons learned as more sites implement it.

The following considerations have been raised (Fall 2012 NERR meeting) and may be included in future versions:

--options for sites with closed, taller canopies, where it is hard to link roots all the way up to canopy

- --use standard mangrove nomenclature for size classes (seedling/sapling rather than shoot)
- --consider counting numbers within these size classes, rather than measuring each individual
- --re-consider whether it is appropriate that this protocol ignores all other vegetation and only covers mangroves; maybe have option to also include other species in same plots if desired?
- --review international blue carbon monitoring protocols and make this protocol as compatible as possible

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