

# **Vegetation Sampling for Wetland Delineation**

A Review and Synthesis of Methods and Sampling Issues

Edward Gage and David J. Cooper

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**Abstract:** This document reviews sampling issues and methods for characterizing vegetation, with an emphasis on delineating wetland boundaries in accordance with Section 404 of the Clean Water Act. Numerous vegetation sampling methods have been developed, but no single approach is optimal for all sampling objectives, vegetation types, and ecological settings. Methods vary in relative precision, accuracy, and efficiency, although selection of methods should also explicitly consider sampling objectives, resources, and data requirements. Key considerations include the choice of metric(s), selection of sampling units, and logistical issues such as time required for sample collection. Common metrics include frequency, cover, and biomass, with each providing different inferences to community characteristics. Vegetation may be assessed using plots, transects, or points as sampling units, and sampling may treat separate vegetation layers (i.e., strata) separately or together. Sampling units may be located subjectively or by using one of many available randomized sampling strategies, although randomization is essential for any statistical analysis. The document reviews general concepts of relative abundance and dominance and discusses the dominance ratio and prevalence index approaches, which are commonly used in wetland delineation. Lastly, sampling approaches traditionally used to assess community characteristics are contrasted with those specifically developed for boundary determination.

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# **Preface**

This report was prepared by Edward Gage and David J. Cooper, Department of Forest, Rangeland, and Watershed Stewardship, Warner College of Natural Resources, Colorado State University, Ft. Collins, Colorado, for the Remote Sensing/Geographic Information Systems and Water Resources Branch (RR-C), U.S. Army Engineer Research and Development Center — Cold Regions Research and Engineering Laboratory (ERDC-CRREL). The CRREL Principal Investigator was Robert Lichvar (RR-C).

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## 1 Introduction

#### 1.1 Background

Section 404 of the Clean Water Act is the main federal regulatory framework governing activities in wetlands. Along with the U.S. Environmental Protection Agency (EPA), the U.S. Army Corps of Engineers (USACE) has primary responsibility for developing and implementing procedures for regulation under Section 404. The USACE has established procedures for defining and delineating wetlands, which are described in the 1987 Wetlands Delineation Manual (Environmental Laboratory 1987), or simply, the 1987 Manual. This document, along with various supporting documents and supplements, is used nationwide to determine the jurisdictional status and boundaries of wetlands subject to Section 404 regulation.

While supporting the basic structure of the 1987 Manual, a National Research Council review of the technical validity of existing wetland delineation procedures concluded that regional variation among the Nation's wetlands limits the utility of a single national delineation manual and recommended that regionalized procedures be developed (National Research Council 1995; Wakeley 2002). Efforts are underway to develop region-specific guidance on wetland delineation procedures, and several interim and final regional supplements to the 1987 Manual have been produced (Wakeley 2002; USACE 2007, 2008a, b, c, d, e, f, g).

A central concept to the approach used in the 1987 Manual is that wetlands are defined by the presence of particular hydrologic, edaphic, and biotic characteristics. With the exception of atypical or problem situations, indicators of all three factors must be present for a site to be considered a wetland. The key biotic element to this "three-factor approach" is site dominance by hydrophytes. For delineation purposes, a hydrophyte is "any macrophyte that grows in the water or on a substrate that is at least periodically deficient in oxygen as a result of excessive water content" (Environmental Laboratory 1987); however, other definitions have been offered (Tiner 1991).

A variety of methods are used for characterizing vegetation for wetland delineation purposes. The 1987 Manual and the regional supplements to

the manual present specific procedures and are the only protocols accepted for regulatory use by the USACE, even though other methods have been developed, such as the Federal Interagency Manual for Identifying and Delineating Jurisdictional Wetlands and the EPA Wetland delineation manual (Sipple 1988a, b; FICWD 1989). Analysis of plant communities typically involves collecting plant cover data within sample units such as plots, with cover estimates made separately by vegetation strata. Strata are typically defined by life form (e.g., woody overstory, woody understory, woody vines, groundcover), although age and size criteria are also used (Environmental Laboratory 1987). One of the goals of using strata is to use all vegetation layers rather than just the dominant overstory species, as species in different layers may provide better indicators of hydrologic regime and wetland soil conditions (Environmental Laboratory 1987; Tiner 1991, 1999).

Specific methods for characterizing plant composition and abundance are presented in the 1987 Manual. However, in the two decades since its initial publication, significant advances in wetland science have occurred, and improved methods have been developed for assessing vegetation (Stohlgren et al. 1998; Kercher et al. 2003). In addition, there has been greater agreement that a "one-size-fits-all" approach for characterizing wetlands is limited and that region-specific methods are needed (National Research Council 1995; Wakeley 2002; USACE 2006, 2008b, c, d, f).

## 1.2 Objectives

The goal of this document is to review the relevant scientific literature on vegetation sampling and analysis, with an emphasis on applications to wetland delineation. Most relevant literature is from outside the realm of wetland science; therefore, our treatment is broad in scope. Our specific objectives are:

- To review basic vegetation sampling concepts and definitions;
- To assess the strengths and weaknesses of different sampling approaches commonly used to characterize vegetation;
- To summarize the consequences of methodological decisions such as plot size, shape, and sampling intensity on the results of vegetation analyses;
- To provide an overview of different approaches for locating sampling units in the field, including subjective and statistically based designs;

• To review the use of dominance ratios and prevalence indices, which are the two primary approaches currently used to determine whether a site supports a predominance of hydrophytes;

- To review the rationale for stratifying plants in vegetation analysis, particularly in the context of wetland delineation; and
- To review the application of vegetation data to field determination of wetland boundaries.

## 2 Literature Review and Discussion

#### 2.1 Definitions and basic concepts

Plants are a key component of most wetlands and provide many important ecological functions (Smith et al. 1995). Typically, only plants with appropriate physiological and life-history adaptations are able to persist and compete in the distinct environmental conditions characteristic of wetlands (Mitsch and Gosselink 2007). Some species occur only in wetlands; others occur in a range of wetland and non-wetland habitats. Observed differences in the relative frequency with which a species occurs in wetlands provides the basis for the National and Regional Lists of Species found in Wetlands (Reed 1988), which is used for determining if a site supports hydrophytic vegetation.

The use of plant species as indicators of wetland boundaries has important limitations. For example, genotypic variation or phenotypic plasticity can lead to considerable local or regional variation in the relative frequency with which individual plants of a particular species occur in wetlands (Tiner 1991, 1999). However, the concept of hydrophyte is integral to wetland determination and delineation, and, according to federal regulations [33 CFR 328.3(b)], a site must support "a prevalence of vegetation typically adapted for life in saturated soil conditions." It is important to note that the wetland indicator status (e.g., Obligate, Facultative Wetland, Facultative, Facultative Upland, Upland) is not well characterized for the majority of species and is often determined without reference to significant data. Regardless of the specific sampling method used to characterize vegetation, the outcome of delineations determined using methods currently accepted by regulators can change if the wetland indicator scores used in the national or regional wetland lists change.

The delineation methods currently accepted by the USACE place particular emphasis on characterizing the *prevalence* of species (and related concepts such as dominance and abundance). *Prevalence* has been defined as the number and size of spaces inhabited by an organism; *abundance* has been defined as the number of organisms in a population and combines *intensity* (density within inhabited areas) and prevalence (Begon et al. 1996). More simply, abundance has been defined as the number of individuals or shoots of a species (taxon) in a particular study area (Knapp

1984). In the 1987 Manual, prevalence refers to "...the plant community or communities that occur in an area at some point in time" (Environmental Laboratory 1987). This emphasis on prevalence has implications for assessing the relative performance of different vegetation sampling strategies. For purposes such as monitoring, functional assessment, or classification, accurate and precise estimates of broader community metrics such as species richness are important. These estimates often drive many of the critical sampling decisions such as plot size, plot shape, and minimum number of sampling units required to achieve a desired level of accuracy and precision (Barbour et al. 2000; Niemi and Mcdonald 2004; Archaux et al. 2007; Stohlgren 2007; Johnson et al. 2008; Dengler 2009).

In general terms, *dominance* can be used to describe species in the uppermost stratum¹ and having the greatest cover (e.g., *Populus deltoides* woodland), irrespective of species in lower strata (Tiner 1999). In contrast to the more general usage of the term in community ecology, a dominant species has specific meaning in the context of wetland regulation. According the 1987 Manual, dominant plant species are those that "contribute more to the character of a plant community than other species present, as estimated or measured in terms of some ecological parameter or parameters" (Environmental Laboratory 1987). Typically, assessment of dominance is made separately for each stratum present in a community on the basis of areal plant cover (herbaceous understory), greatest height (woody understory), basal area (woody overstory), and/or greatest number of stems (woody vines).

Identification and delineation of boundaries among plant communities is complicated by the fact that the very nature of plant communities remains controversial. Clements (1916) formulated an early concept of plant communities, which views communities as analogous to organisms with distinct structural and functional linkages and definable boundaries. In contrast, Gleason (1926) espoused an individualistic concept of vegetation, where communities are viewed as loose aggregations of independently distributed plant species that co-occur because of similarities in habitat requirements and environmental tolerances. The individualistic concept is favored by most ecologists (van der Valk 1981; Callaway 1997; Pickett et al. 2009).

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<sup>&</sup>lt;sup>1</sup> Strata defined by the 1987 Manual: groundcover (herbaceous species and woody plant seedling < 1-m tall), woody vines, woody understory [woody plants < 3.0 in. DBH (diameter at breast height)], and woody overstory (woody plants ≥ 3.0 in. DBH) (Environmental Laboratory 1987).

Many ecologists feel that vegetation varies continuously along environmental and geographic gradients (Curtis and McIntosh 1951; Whittaker 1953; Curtis 1955; McIntosh 1967). Depending on the specific ecosystems and the nature of underlying environmental gradients, clear boundaries between communities or plant associations often do not exist (Figure 1). From a Gleasonian perspective, a community simply consists of those species that coexist in a particular space and time (Odenbaugh 2007); from this perspective, communities lack the critical properties of selforganization and integration that would make them truly analogous to cells or organisms (Rowe 1961; Lapin and Barnes 1995). Thus, any specific decision as to how to divide continuously varying vegetation into community types is somewhat arbitrary with more than one defendable solution.

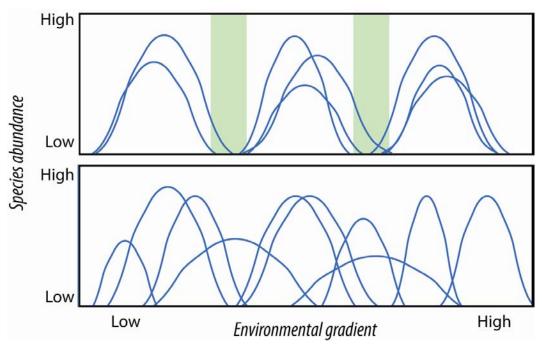


Figure 1. Hypothetical examples of different species response curves along environmental gradients. In the top panel, the species response curves are closely aligned, making objective identification of community boundaries (shaded areas) more tenable than in the bottom panel, where species response curves greatly overlap. The latter situation is consistent with a Gleasonian view of plant communities. (After Kent et al. 1997.)

A number of procedures have been developed for assessing wetland characteristics. The salient features of each procedure vary, depending on the specific purpose. For example, methods have been developed for rapidly assessing wetland condition or function in the field (Ainslie 1994; Van Dam et al. 1998; Carletti et al. 2004; Collins et al. 2006; Fennessy et al. 2007). These methods differ from those developed for broad-scale

vegetation mapping and classification (Cowardin et al. 1979; Pierce and Johnson 1986; Cooper 1998; Kittel et al. 1999; Muldavin et al. 2000; Harken and Sugumaran 2005) or for long-term monitoring or evaluation of mitigation projects (EPA 2002a; Robb 2002; Fraser et al. 2005; Brooks et al. 2006).

Methods for regulatory wetland delineation also vary for state and local governments. Different federal agencies (EPA, USACE), state governments, and even municipalities have specified methods for wetland delineation and mapping (Cooper 1988; Sipple 1988a, b; FICWD 1989). However, since the USACE is given national responsibility for wetland regulation under the Clean Water Act, most methods closely follow those mandated by the Corps in their 1987 Manual (Environmental Laboratory 1987). The 1987 Manual provides general guidance on sampling vegetation for jurisdictional determination and delineation purposes, and additional guidance is provided in regional supplements to the 1987 Manual (USACE 2007, 2008a, b, c, d, e, f, g) and in a variety of texts (Lyon 1993; Tiner 1999; Kent 2001). While we provide a review of the methods in the 1987 Manual and other methodologies below, readers are directed to the original documents and more comprehensive texts available for wetland delineation (Lyon 1993; Tiner 1999).

Clear identification of sampling objectives is critical for selecting an appropriate sampling approach. The most suitable method for a routine wetland determination will differ from that needed for long-term monitoring. Different sampling procedures are commonly followed in the scientific literature, depending on the specific discipline and sampling objectives. Statistical and computational procedures for boundary determination differ from those used to characterize mean conditions or trends and frequently differ from approaches used in traditional community ecology (Jongman et al. 1995; Fortin and Dale 2005). Examples of statistical and computational techniques are available from other disciplines such as remote sensing, landscape ecology, and medical imaging, which share a more consistent focus on data segmentation and boundary determination (Cernadas et al. 2002; Fortin and Dale 2005; Baker et al. 2006, 2007; Yu et al. 2006).

The 1987 Manual offers users flexibility in making sampling decisions, allowing methods to be tailored to specific site conditions and sampling purposes (e.g., routine versus comprehensive determinations). The

regional supplements provide additional guidance on selecting sampling methods (Wakeley 2002). In practice, the specific practices of wetland scientists, consultants, and regulators in the field vary widely. The boundary decisions typical of many routine delineations rely on the judgment of the delineator as much as on any quantitatively analyzed vegetation data, because a limited number of plots or transects, often purposely located interior or exterior to perceived boundaries, are used.

Data requirements for reliable sampling in research applications exceed those in most jurisdictional delineations. The literature on quantitative vegetation sampling theory and practice is extensive; likewise, methods for classification, mapping, and boundary determination are common. However, in general, the quantitative rigor of such studies—necessary to appreciably advance the science and pass peer review—greatly exceeds that typically performed for delineation of a wetland.

Billing rates for consultants are high, and regulators as a rule are overextended. As a consequence, any sampling procedure specified for regulatory use must allow for the very real constraints on acceptable time for sampling and analysis. Anecdotally, the comprehensive method presented in the 1987 Manual is rarely used by practicing delineators (W. Sipple, personal communication). Although rigorous sampling procedures will provide more accurate estimates of vegetation characteristics, such rigorous procedures will rarely be used if a less intensive alternative procedure is available.

# 2.2 Sampling variables and metrics

The two most common metrics used to evaluate vegetation are frequency and cover. Frequency refers to the probability of detecting a species in a randomly placed sampling unit (e.g. point, transect) in a particular area (Bonham 1989). Frequency is typically expressed as a percent of the total number of observations, calculated either within a sampling unit such as a plot or among sampling units. An advantage of frequency data is that it can generally be collected relatively easily and efficiently. One of the primary disadvantages is that values are highly sensitive to differences in the size and shape of sample units (Bonham 1989).

Plant cover refers to the vertical projection of vegetation, or some other cover type such as litter, onto the ground when viewed from above (Bonham 1989). Total cover refers to the relative percentage of a plot area

covered by individuals of a particular species and can sum to more than 100% when plant canopies overlap. Alternatively, one can calculate relative cover, where the total cover for all species in a particular stratum is fixed at 100%. There can be significant differences in cover estimates from different observers (Smith 1944; Sykes et al. 1983; Milberg et al. 2008), but errors can be minimized by ensuring that a single trained observer is responsible for measurements (Mitchell et al. 1988). A third option, commonly used to improve precision among observers, is to estimate cover using cover classes (Table 2). When estimates are made using cover classes, there is often a tendency to overestimate of low mean cover (<15%) and underestimate high mean cover. Estimates for small or rare individuals tend to be higher than estimates from other techniques, a consequence of the assumption that cover values are uniformly distributed about the mid-points of the cover classes (Floyd and Anderson 1987).

Variation among observers tends to be smaller at the high and low ends of the range of cover values; thus, most cover class schemes use smaller ranges at these values (Daubenmire 1959; Hatton et al. 1986). Species with the lowest cover values tend to have the largest measurement errors (Kennedy and Addison 1987), although these errors are likely of small significance to wetland delineation. Cover-class data from a few large-plot observations are relatively sensitive to qualitative changes in the understory cover of dominants but do not provide reliable quantitative estimates of cover dynamics (Mitchell et al. 1988). Anecdotes related from many years of teaching wetland delineation suggest that students often have difficulty with the 50% breakpoint used in many cover class schemes (Tiner 1999).

Basal area represents an alternative metric for characterizing the abundance of trees. Direct measurements are typically made using diameter tapes. Basal area can also be used indirectly to determine which trees are sampled via use of an angle gauge or basal area prism (Husch et al. 2003; Packard and Radtke 2007). Basal area data can be used to test for the presence of hydrophytic vegetation using the dominance ratio approach, but it cannot be used to calculate a prevalence index, which requires absolute cover data for all species in each stratum (USACE 2008b).

Presence/absence techniques simply note whether a species is present at a particular site, with no assessment of relative abundance (Elzinga et al.

1998). Given that the wetland determination is defined, in part, by the prevalence of hydrophytes (Environmental Laboratory 1987), presence/absence data cannot be used directly in making wetland determinations or delineations. Similarly, presence/absence methods by themselves are of little value for monitoring purposes because, other than complete disappearance of a species, they provide no information on trends (Elzinga et al. 1998). However, because traditional plot or transect-based approaches commonly fail to effectively capture species diversity, presence/absence data, which can be collected more rapidly than species abundance data, can be used as part of a multi-scale sampling design to efficiently note uncommon or rare species not represented in plots or transects (Ramsay et al. 2006). Such information has little bearing on wetland determination or boundary delineation, but it can be used to evaluate differences in diversity among sites; for example, comparing biodiversity in natural versus man-made wetlands (Magee et al. 1999).

Other sampling attributes include density and structural characteristics such as plant height (Elzinga et al. 1998). Stem density is used to characterize the abundance of plants in the woody vine stratum. Plant height can be used to characterize plants in the woody understory stratum (Environmental Laboratory 1987) and is an element in several forest inventory programs. These alternative metrics to cover or frequency are not regularly used in the majority of vegetation studies, nor are they regularly measured as part of wetland delineations.

Table 1. Suggested cover classes in the 1987 Manual (Environmental Laboratory 1987), originally presented by Daubenmire (1959), 1988 EPA Manual (Sipple 1988), 1989 Interagency manual (FICWD 1989), and Tiner (1999).

| Class | 1987 Manual | Sipple (1988) | 1989 Manual | Tiner (1999) |
|-------|-------------|---------------|-------------|--------------|
| Trace |             | <1%           | <1%         | <1%          |
| 1     | 0-5%        | 1-5%          | 1-5%        | 1-4%         |
| 2     | 6-25%       | 6-15%         | 6-15%       | 5-15%        |
| 3     | 26-50%      | 16-25%        | 16-25%      | 16-25%       |
| 4     | 51-75%      | 26-50%        | 26-50%      | 26-39%       |
| 5     | 76-95%      | 51-75%        | 51-75%      | 40-60%       |
| 6     | 96-100%     | 76-95%        | 76-95%      | 61-74%       |
| 7     |             | 96-100%       | 96-100%     | 75-84%       |
| 8     |             |               |             | 85-95%       |
| 9     |             |               |             | 96-99%       |
| 10    |             |               |             | 100%         |

#### 2.3 Defining and locating sampling units in the field

A key sampling decision is to determine the most suitable method for locating sampling units in the assessment area. There are two general approaches: locating sampling units subjectively in representative stands or using some sort of randomization. The former is commonly used in wetland determination and delineation assessments but is discouraged where more rigorous statistical analyses are desired. Examples of different methods for randomly locating sampling units include simple random sampling, stratified random sampling, and systematic sampling with a random start (Bonham 1989, Elzinga et al. 1998). Each has advantages and disadvantages, depending on the underlying distribution of plants in the environment and the sampling objectives. Stratification of an area or landscape *a priori* into units, within which the sampling units are randomly located, represents a hybrid approach and is often the preferred method when the goal is description and classification (Jennings et al. 2004).

A variety of sampling units can be used to assess plant abundance and community composition. Most estimates of cover or frequency use plots as the primary sampling unit. The choice of specific sampling unit determines the appropriate statistical treatment for data. Transects are the primary sampling unit when the line-intercept method is used. If plots (or points, if point-intercept methods are used) are placed in a contiguous fashion or are placed only a short distance apart along a transect line, adjacent sampling units will likely be correlated, resulting in an underestimation of the standard error (Elzinga et al. 1998). For distance methods (i.e., plotless methods), which are sometimes used to estimate tree abundance, the sampling unit is usually the individual distance between a randomly selected point and the nearest plant (Elzinga et al. 1998).

The size and shape of sampling units are important considerations in sampling design. From a statistical perspective, the ideal design is one that produces the highest statistical precision for a given unit of sampling effort (Elzinga et al. 1998); however, other considerations like ease of layout and sampling time are also important. A variety of factors can influence the relative performance of different plot shapes in accurately capturing the vegetation characteristics of interest, including vegetation type, vertical structure, and the spatial pattern of plants in the assessment area.

Published studies vary widely in the recommended size for sampling units, and it is not always clear how much of this variation can be explained by the different objectives and/or criteria being used to evaluate the performance of a particular sampling approach. In general, the size of sampling units should be a function of the size of the plants being sampled. Intuitively, larger sampling units are appropriate where plants are larger—trees versus herbs, for example. Additional considerations, however, can come into play. For example, the effectiveness of different sampling units for capturing uncommon or rare species varies. If this is an important sampling objective, sampling units will need to be larger than if identification of dominant species were the goal.

Edge effects can occur when there is uncertainty about whether plants near the plot edge are in or out of the plot. The perimeter-to-area ratios of different plot shapes differ, with rectangular plots having more edge per unit area than circular or square plots (Bormann 1953). Because of this, some advocate the use of circular plots, which minimize errors introduced by edge effects by minimizing the perimeter-to-area ratio. Circular plots are commonly used in vegetation studies and, with the exception of small plots, are recommended by the 1987 Manual (Environmental Laboratory 1987) and other delineation manuals. Depending on cover type, circular plots may be easier to create in the field, requiring only a center stake and tape (Tiner 1999).

Other authors have concluded that rectangular plots are more precise than square plots (Bormann 1953; Elzinga et al. 1998). When plant populations exhibit a clumped distribution, which is often the case in natural ecosystems, rectangular plots will yield more precise estimates than square or circular plots of the same size because rectangular plots are more apt to include some of the clumps inside of them and thereby reduce the variation among plots and increase the precision of estimates (Elzinga et al. 1998). Some comparative studies, however, have found no significant difference among plot shapes (Keeley and Fotheringham 2005).

The 1987 Manual has allowed considerable flexibility in plot shape. The choice of shape should be based on the characteristics of the communities being sampled. Long and narrow communities (e.g., riparian fringe, dune swale) are more appropriately sampled using rectangular plots of suitable dimensions. In addition, where a particular vegetation unit is too small or is irregularly shaped and cannot accommodate standard plots, USACE

methods allow workers to make percent cover estimates by walking the unit and visually estimating the coverage. Intuitively, it is appropriate to use larger plots to collect data for larger life forms such as mature trees; small organisms, such as herbaceous plants or bryophytes, can be effectively sampled using much smaller plots (Bonham 1989). The most appropriate size for a sample plot depends on the type of vegetation (i.e., trees, shrubs, herbaceous plants), the spatial extent and shape of the plant community, and the specific sampling objectives.

General suggested plot sizes are presented in several basic vegetation sampling manuals (Table 2). From a historical perspective, the relevé method of community classification (Mueller-Dombois and Ellenberg 1974) has strongly influenced notions of plot size, and variations on this approach are still widely used (Peet 1981; Wells 1996; Dierssen and Dierssen 2005; Hajkova et al. 2006). Mueller-Dombois and Ellenberg (1974) indicated that the sample area should be large enough to contain all of the species belonging to a plant community and should be relatively uniform in total plant cover. These authors suggested a range of plot sizes depending on the stature of the plants in question. In his influential work in conifer forests, Daubenmire (1968) recognized a wide variation in plant sizes and proposed using nested plots. All trees greater than 1 m (3.28 ft) tall were recorded in macro plots measuring  $5 \times 25$  m (16.4  $\times$  65.6 ft), whereas trees less than 1 m (3.28 ft) tall were recorded in separate 1-m- (3.28-ft-) wide belt transects. The cover of shrubs and herbaceous species were evaluated in sets of 20- $\times$ 50-cm  $(0.66-\times 1.64$ -ft) quadrats spaced 1 m (3.28 ft) apart along the inside boundaries of macroplots (Daubenmire 1968).

In general, plots should be large enough to adequately sample populations but not so large that sampling cannot be accomplished in a timely fashion. Kenkel and Podani (1991) evaluated the effects of changing plot size on parameter estimation efficiency, which they defined in terms of the statistical precision of estimates of all variables (e.g., species) in a data set. The authors developed three "efficiency criteria" and compared estimation efficiency and plot size using data from three community types: deciduous understory, coniferous understory, and peatland. Estimation efficiency increased with increasing plot size for all three communities, but efficiency gains at larger plot sizes were offset by increases in sampling effort. The authors concluded that the largest plot size possible should be used, given time constraints, and that plots larger than the mean patch size should be used when sampling heterogeneous vegetation (Kenkel and Podani 1991).

Quantitative methods for determining optimal plot size, incorporating the variation among plots, the per-plot sampling costs, and the cost of locating additional plots, are available (Krebs 1989; Elzinga et al. 1998) but are not widely used in wetland identification and delineation because guidance on plot size is provided in federal delineation manuals, regional supplements, and various texts (Tiner 1999). These methods require considerably more sampling effort than is typical for routine delineations. In addition, such approaches generally evaluate necessary sampling efforts in light of the capture of less common species, which, depending on the criteria used for making boundary determinations, may not be as important for routine delineation purposes.

Federal wetland delineation manuals recommend different plot sizes depending on the vegetation stratum and whether sampling is part of the routine or comprehensive method for delineation (Environmental Laboratory 1987). According to the 1987 Manual guidelines for routine determinations, a 5-ft- (152-m-) radius plot is suggested for plants in the herbaceous and woody understory strata, whereas a 30-ft- (9.14-m-) radius plot is recommended for trees and woody vines. For comprehensive determinations, the 1987 Manual specifies 30-ft- (9.14-m-) radius plots for trees, 10-ft- (3.05-m-) radius plots for saplings and shrubs, and either a 1.64-ft- (0.5-m-) radius plot or a 3.28-  $\times$  3.28-ft (1-  $\times$  1-m) plot for herbs. These guidelines are largely arbitrary and have not received extensive review under various field conditions.

Table 2. Suggested plot sizes for use in routine vegetation sampling (reproduced from Stohlgren 2007).

| Vegetation                    | Daubenmire (1968)                          | Mueller-Dombois and Ellenberg (1974)                  |
|-------------------------------|--|---|
| Mosses and lichens            | 25 m <sup>2</sup> (269.1 ft <sup>2</sup> ) |   |
| Mosses                        |  | 1-4 m <sup>2</sup> (10.8-43.1 ft <sup>2</sup> )       |
| Lichens                       |  | 0.1-1 m <sup>2</sup> (1.08-10.8 ft <sup>2</sup> )     |
| Herbs                         |  | 1-2 m² (10.8- 21.5 ft²)                               |
| Herbs and shrubs              | 0.1 m <sup>2</sup> (1.08 ft <sup>2</sup> ) |   |
| Dry grassland                 |  | 50-100 m <sup>2</sup> (538.2-1076.4 ft <sup>2</sup> ) |
| Tall herbs/low shrubs         |  | 10-25 m <sup>2</sup> (107.6-269.1 ft <sup>2</sup> )   |
| Tall shrubs                   |  | 16 m <sup>2</sup> (172.2 ft <sup>2</sup> )            |
| Large plants/<br>trees/forest | 4-100 m² (43.1-1076.4 ft²)                 | 200-500 m² (2152.7-5382 ft²)                          |
| Forest understory             |  | 50-200 m <sup>2</sup> (538.2-2152.7 ft <sup>2</sup> ) |

Guidelines have been modified in some regional supplements. For example, the use of a 15-ft- (4.57-m-) radius plot is suggested for saplings and shrubs and a plot up to a 35- or 40-ft- (10.7–12.2-ft-) radius, as needed, for trees in the Northeast and Northcentral regional supplement (USACE 2008b). The 1987 Manual and regional supplements allow for the modification of plot sizes to better characterize site conditions. Plot size and shape are typically modified to avoid crossing boundaries between adjacent communities or in more diverse communities. Sampling areas should have relatively uniform soils and hydrologic regimes (USACE 2008c, d, f). Changes from standard plot sizes or shapes are important to note on data forms because these factors will influence the repeatability of results (Elzinga et al. 1998). The use of large plots [e.g., 40 ft (12.2 m) radius] are appropriate for community characterization, but how information collected at such a broad spatial extent can be used to refine the placement of a specific boundary line is not clear. For delineation purposes, data from such large plots are likely to obfuscate boundary delineation, because the scale of measurement is so mismatched to the goal—the determination of a single line in the field.

Ideally, vegetation data are collected using plots of multiple sizes to provide a direct means of evaluating the effect of plot size on vegetation patterns (Stohlgren 2007). A variety of multi-scale plot designs are available, such as variants of the relevé (Peet 1974; Barbour et al. 1998), nested pixel plot (Kalkhan et al. 2007), and modified-Whittaker plot (Stohlgren et al. 1995; Barnett and Stohlgren 2003). These methods are particularly useful for characterizing plant species diversity and are used in wetland functional and condition assessment techniques (EPA 2002b; Rocchio 2006), although they have rarely been used for wetland delineation, where greater emphasis is placed on ease of use and speed of methods in the field.

The point-intercept method, recommended by the FICWD (1989) manual and presented as an alternative sampling approach in regional supplements to the 1987 Manual (USACE 2008a, b, c, d, e, f, g), can be viewed as a plot-based method where the point represents the smallest possible plot (Tiner 1999). Cover for any particular species is estimated by determining the total number of points that intersect the canopy divided by the total number of points evaluated. Points are systematically sampled along transect lines and are typically analyzed using a transect-based prevalence index to evaluate whether a predominance of hydrophytic

vegetation is present. The approach can improve quantification of species abundance and reduce bias compared with visual cover estimates. Repeatability among observers is generally higher using point-intercept methods compared to visual cover estimates made on plots (Symstad et al. 2008).

Point-intercept sampling is an appropriate alternative to plot-based sampling in communities that have high species diversity or patchy species distribution, where identification of representative locations for plot sampling is difficult. For point cover data, it is desirable to be able to treat points as the sampling units rather than transects along which points are located, because the estimates will be more precise because of larger sample sizes (Elzinga et al. 1998). However, where points are closely spaced along transect lines, points are unlikely to be truly independent. A disadvantage is that the approach can require more time for sampling and requires areas large enough to allow establishment of transect lines. Typically, a transect-based prevalence index is used to determine whether there is a prevalence of hydrophytic vegetation. More detailed procedures for point-intercept sampling are presented in the various regional supplements to the 1987 Manual (USACE 2007, 2008b, c, d, f).

The line-intercept method, developed for and commonly applied in grassland and rangeland studies, estimates the cover of a particular species by noting the points at which the canopy begins and ends along a transact line (Canfield 1941; Lucas and Seber 1977; Floyd and Anderson 1987; Elzinga et al. 1998; Jorgensen et al. 2000). Intercepts are added and then divided by the total line length to generate a cover estimate for a particular species. The approach can be effective for species with dense canopies such as some shrubs, but it is difficult to apply in cover types such as grasses or sedges, where a large number of small interceptions require evaluation (Elzinga et al. 1998). Disadvantages of this method include the introduction of observer bias, which is created by failure to establish a true perpendicular sighting line on the tape and imprecision created when winds blow foliage. This is also a disadvantage for the point-intercept method, particularly in forested ecosystems.

In a comparison of point-intercept and line-intercept methods in sagebrush steppe ecosystems, the degree of precision was similar; however, the line-intercept method required nearly three times more sampling effort (Floyd and Anderson 1987). Neither method appeared to

bias cover estimates for any particular taxon or growth form (Floyd and Anderson 1987). Because of the drawbacks, the line-intercept method is rarely used in wetlands studies and is not suggested in the key documents guiding sampling for wetland delineation.

Plotless methods can be effectively used to characterize the abundance of trees and, less commonly, herbaceous species. A variety of plotless approaches have been developed, although only the plotless comprehensive method or Bitterlich variable plot method (Tiner 1999) is commonly used for wetland delineation purposes. The approach involves estimating basal area using a basal area factor (BAF) prism or angle gauge to identify individual trees for diameter measurement ("in" trees). Trees are included in the sample as a function of both their DBH and distance from the sampling point; inclusion is not based on location within a fixed area, thus the designation of the method as "plotless" (Schreuder et al. 1993). The inclusion probability for each tree depends on the tree diameter; larger trees have larger inclusion probabilities than small ones (Kangas and Maltamo 2006). In the method described in the FICWD manual (1989), all trees visible using the prism or angle gauge are tallied by species and total basal area. Basal area values are summed for individual trees by species, and each species is then ranked by total basal area. A dominance threshold number is determined by summing the basal areas of all tree species and multiplying by 50%. Next, basal area values for ranked tree species, beginning with the largest value, are summed until the dominance threshold number is exceeded; all species that contributed to surpassing the threshold number are considered dominants for determining whether hydrophytic vegetation is present (FICWD 1989). If trees are randomly distributed, distance approaches can provide unbiased estimates of tree density and basal area. However, this is rarely the case. When trees are clumped or over dispersed, systematic errors are introduced (Cottam and Curtis 1956; Johnson et al. 2008). Density and basal area will be underestimated when species are aggregated and overestimated when uniform (Bryant et al. 2005; Johnson et al. 2008).

Image-based analyses have been used successfully to estimate plant cover, although these methods have primarily been used in rangelands, and their applicability to wetlands is largely unexplored (Booth et al. 2005; Booth and Cox 2008). Image-based methods take advantage of the relative ease and low cost of acquiring digital pictures and allow more extensive sampling than traditional ground-based methods. Cover estimates are

created from images using one of two approaches. Image spectral analysis and classification can be used to identify and segregate different cover types in the image. Another approach is to select points within the image and have the operator manually classify sample points, analogous to point-intercept methods in the field (Booth et al. 2006a). Studies comparing cover estimates derived from digital images to traditional methods have generally found good agreement (Booth et al. 2005, 2006b). Because these methods are largely untested for wetland determination and delineation purposes, they do not provide a suitable alternative to traditional sampling approaches. However, they can provide supporting evidence for conclusions drawn from traditional approaches and, with further validation, may prove useful for making more general assessments.

Some non-vascular plant species are strongly associated with wetlands and, when abundant, provide strong indicators of hydrophytic vegetation (Seppelt et al. 2008; Lichvar et al. 2009). Sampling bryophytes presents particular challenges. Identifying taxa is difficult, and species can vary widely among microhabitats over short spatial scales (Eldridge et al. 2003; Newmaster et al. 2005). Methods used to evaluate non-vascular species should therefore incorporate an assessment of the microhabitats present at a site. The Alaska regional supplement to the 1987 Manual presents a procedure for identifying hydrophytic vegetation based on the presence and abundance of wetland specialist bryophytes (USACE 2007). In some wetland types, bryophytes may contribute significant floristic diversity and canopy coverage to wetlands. If this is a characteristic of interest (for example, as a means of evaluating the success of wetland mitigation), then the value of sampling will be greatly reduced by ignoring the bryophyte component.

## 2.4 Vegetation stratification for sampling

Plants in wetlands exhibit a wide variety in physical forms and basic life-history attributes. Broadly, these characteristics help shape the nature of different wetland types and contribute to their basic ecological functioning (Keddy 2000; Keddy et al. 2002; Mitsch and Gosselink 2007). Wetlands range from forested ecosystems with complex, multi-layered canopies to monospecific communities dominated by low-lying annual herbaceous species or bryophytes (Mitsch and Gosselink 2007). These differences, in part, drive functional differences among wetland types. Plants are generally fixed in space; as sessile organisms, they must acquire the necessary resources for growth and reproduction in situ. The result is

significant competition with other plants for resources such as light and nutrients, influencing many of the basic processes governing the spatial and temporal dynamics of plant communities (Barnes et al. 1998; Keddy 2000).

The use of strata is an integral part of existing wetland delineation methods. Vegetation sampling procedures described in the 1987 Manual are focused on determining dominance within separate vegetation strata. One rationale for this approach is that different life forms may lead to different inferences about underlying hydrologic and soil characteristics. For example, phreatophytic trees and shrubs such as willows (*Salix* spp.) and cottonwoods (*Populus* spp.) may be able to persist even if hydrologic regimes change; in these cases, understory species may provide a more useful indicator of wetland status (Tiner 1999; USACE 2008g). Dewey et al. (2006) suggested that, in their study area, the tree stratum may provide information on long-term wetland status, whereas the herbaceous layer may provide only short-term data, especially in sites with long, dry summers and where the vine stratum is dominated by annuals and perennials inhabiting dry microclimates created by other vegetation.

An additional rationale for sampling strata is to provide inferences on historical processes such as stand establishment and potential future changes in community composition and characteristics (i.e., succession) (Turner et al. 1999; Barbour et al. 2002). The composition of different layers, particularly when interpreted in light of species life-history characteristics and physiological characteristics such as shade tolerance, can provide useful information from which to infer future changes in composition and structure. This is one reason that data regarding different layers such as seedlings, saplings, and overstory vegetation are routinely collected in many forest inventory methods (Schreuder et al. 1993; McRoberts and Hansen 1999; McRoberts et al. 2005; Kangas and Maltamo 2006). However, given the specific objectives of regulatory wetland delineation procedures, these may not be particularly compelling reasons to include separate sampling by vegetation strata.

The 1987 Manual identifies four strata: woody overstory, woody understory, woody vines, and groundcover (Table 3) (Environmental Laboratory 1987). The FICWD (1989) manual identified five strata, using different criteria than the 1987 Manual. Some regional supplements to the 1987 Manual have made modifications of the original guidelines (USACE

2008) (Table 3). A variety of other studies also employ the use of strata in vegetation sampling. For example, the U.S. Forest Service collects vegetation data using strata from long-term monitoring plots as part of its Forest Inventory and Analysis (FIA) and National Forest Health Monitoring (FHM) programs (Tallent-Halsell 1995; McRoberts et al. 2005). In the FIA methodology, separate canopy cover estimates are made using cover classes for plants in the following layers: 0-2 ft, >2-6 ft, >6-16 ft, and >16 ft (0-0.61 m, 0.61-1.83 m, 1.83-4.9 m). Vertical structure is a metric included in some wetland condition assessment approaches. Depending on wetland type, users of the California Rapid Assessment Method place vegetation into one of three height categories (Collins et al. 2006).

Sampling guidelines for the National Vegetation Classification System (NVCS) recognize six basic vegetation strata: tree, shrub, field, ground, floating aquatic, and submerged aquatic (Jennings et al. 2004, 2009). The tree stratum is defined as the layer of vegetation where woody plants are typically more than 5 m (16.4 ft) in height, including mature trees, shrubs over 5 m (16.4 ft) tall, and lianas, as well as epiphytes growing on these woody plants. The shrub stratum is where woody plants are typically more than 0.5 m (1.64 ft) tall but less than 5 m (16.4 ft) in height and can include shrubs, tree saplings, and lianas. Rooted herbs are excluded, even if they are over 0.5 m (1.64 ft) in height, with the rationale that, because their stems may die back annually, they do not provide consistent structure. The field stratum includes herbs as well as woody plants less than 0.5 m (1.64 ft) in height, whereas the ground stratum includes nonvascular mosses, liverworts, lichens, and algae growing on soil or rock surfaces. The floating aquatic stratum is the layer of vegetation consisting of rooted or drifting plants that float on the water surface, whereas the submerged aquatic stratum consists of rooted or drifting plants that remain submerged in the water column or on the aquatic bottom. Guidance provided in the core NVCS documents states that emergent plant growth forms in a wetland should be placed in the appropriate strata listed above (Jennings et al. 2004, 2009).

Table 3. Examples of strata definitions from the 1987 and 1989 wetland delineation manuals and an example from the Arid West regional supplements to the 1987 manual.

| Stratum                                 | Definition   |  |  |
|---|--|--|--|
| Environmental Laboratory (1987)         |  |  |  |
| Woody overstory                         | Woody plants with DBH ≥3.0 in. (0.08 m)  |  |  |
| Woody understory                        | Woody plants with DBH <3.0 in. (0.08 m)  |  |  |
| Woody vines                             | Climbing woody vines   |  |  |
| Groundcover                             | Herbaceous species, including seedlings of woody plants <1 m (3.28 ft) tall, plus woody trailing species |  |  |
| FICWD Manual (1989)                     |  |  |  |
| Tree                                    | Woody plants ≥5.0 in. (0.127 m) DBH and ≥20 ft (6.1 m) tall  |  |  |
| Sapling                                 | Woody plants <5.0 in. (0.127 m) DBH and ≥20 ft (6.1 m) tall  |  |  |
| Shrub                                   | Woody plants <20 ft (6.1 m) and generally ≥3.0 ft (0.91 m) tall  |  |  |
| Woody vines                             | Climbing woody vines   |  |  |
| Herb                                    | Nonwoody plants, including seedlings of woody species, plus woody trailing species                       |  |  |
| Arid West Supplement to the 1987 Manual |  |  |  |
| Tree                                    | Woody plants ≥3 in. (0.08 m) DBH, regardless of height   |  |  |
| Sapling/shrub                           | Woody plants <3 in. (0.08 m) DBH, regardless of height   |  |  |
| Herb stratum                            | All herbaceous (non-woody) plants, including herbaceous vines, regardless of size                        |  |  |
| Woody vines                             | All woody vines, regardless of height  |  |  |

The use of vegetation strata in sampling is not universal among researchers and delineators. For example, some do not differentiate cover measurements among strata, particularly within a single species; the rationale is that such an approach overcounts the actual cover for that species. For instance, in forested wetlands, the same species may be present as seedlings, saplings, and overstory trees; summing these individual cover estimates may result in the double or triple counting of cover. An additional argument against the use of strata in sampling is the largely arbitrary nature of divisions among different strata.

If strata are used in sampling, several factors should influence the choice of specific strata. The age and size structure of the vegetation communities being sampled should be explicitly considered; these relate to the most appropriate breakpoints for defining different strata, and they help in deciding the appropriate number of classes. For example, forested wetlands in the Pacific Northwest tend to have both larger trees and a broader distribution of size classes then wetlands in the arid Southwest. It is appropriate to define more strata in the more structurally diverse environments.

If sampled at all, bryophytes (e.g., mosses, liverworts) should generally be sampled as a separate stratum (FICWD 1989). Breakpoints for strata should be made at meaningful levels for the communities being sampled. For example, the shrub component of many arctic and subalpine wetlands may only reach a few feet in height, whereas at lower elevations, the shrub component may reach 20–30 ft (6.1–9.14 m) high (Windell et al. 1986; Schickhoff et al. 2002; Carsey et al. 2003). It would therefore be appropriate to use different strata to characterize the vegetation in these communities.

Regionally based criteria for defining strata have been incorporated into regional supplements to the 1987 Manual. For example, throughout much of the Arid West, short-statured "sub-shrubs" are a common growth form (USACE 2008g). The 1987 Manual combines these plants with herbaceous species into a single stratum for sampling, and the Arid West regional supplement places sub-shrubs into the sapling/shrub stratum. The regional supplement also specifies that if either the tree or woody vine stratum has less than 5% cover during the peak of the growing season, then any trees or vines may be combined with the sapling/shrub stratum.

Most vegetation assessments are focused on living plants because these contribute the most to the ecological characteristics of a particular site. However, ecological functions and characteristics can be strongly shaped by the presence of dead plant materials such as snags and downed wood (Harmon et al. 1986). Dead trees provide critical habitat for a wide range of fauna (George and Zack 2001; Risenhoover and Murden 2007) and provide favorable microclimates conducive to plant regeneration (Harmon et al. 1986; O'Hanlon-Manners and Kotanen 2004). Coarse woody debris is a key structural component of many forested wetlands, riparian areas, and aquatic ecosystems (Bragg et al. 2000; Wohl 2000).

Most methods for assessing vegetation do not characterize dead plant materials in cover estimates, although there are exceptions. For example, the U.S. Forest Service collects data on downed woody materials as part of the Forest Inventory and Analysis (FIA) program (Woodall and Williams 2005). A variety of methods have been developed for inventorying snags as part of habitat assessments for wildlife (Ducey et al. 2002; Kenning et al. 2005; Zielinski et al. 2006; Taylor and MacLean 2007; Temesgen et al. 2008). In general, it is not appropriate to include dead plants in wetlands delineations, as these plants may reflect past environmental conditions.

However, such information can be useful when the goal of the analysis is to characterize environmental changes on site; for example, alterations to the hydrologic regime.

Strata may facilitate the efficient sampling of the diverse range of life forms that occur in wetlands by allowing the selection of different sampling techniques and sampling units appropriate for a specific life form. In addition, because trees, shrubs, and herbaceous species typically differ in basic physiology and life history, separating them in an analysis of vegetation can provide important insights on underlying physical characteristics such as soil type and hydrologic regime, although such differences may also be a result of anthropogenic influences. To maximize their usefulness, strata should be modified as necessary on a regional basis to more closely reflect the characteristics of the communities being sampled.

#### 2.5 Methods for determining prevalence

Several procedures for selecting dominants species have been developed. In many vegetation studies, prevalence is assessed by reference to relative cover, with dominant species assumed to be those with the greatest relative cover. There is no standard definition for what constitutes a dominant species because such a distinction (if made at all) is made in the context of specific sampling and analysis objectives. For example, in many vegetation classifications and mapping efforts, constituent vegetation associations are named on the basis of the most common overstory and understory species, even if the information used to construct the classification itself is not restricted to information from the most abundant species (Peet 1978; Padgett et al. 1989; Jennings et al. 2004). In the 1987 Manual, dominants are specified as the three most abundant plant species in each stratum, unless only one or two strata are present, in which case the five most abundant species are selected (Environmental Laboratory 1987). Since 1992, an additional option for selecting dominance is application of the "50/20" rule (USACE 1992), which provides greater flexibility than the specific quotas specified in the 1987 Manual (Wakeley 2002). The dominance ratio is simply the percentage of dominant species that are rated as obligate (OBL), facultative wetland (FACW), and facultative (FAC).

The 50/20 rule states: "For each stratum in the plant community, dominant species are the most abundant plant species (when ranked in

descending order of abundance and cumulatively totaled) that immediately exceed 50% of the total dominance measure for the stratum, plus any additional species that individually comprise 20% or more of the total dominance measure for the stratum" (FICWD 1989). After the rule is applied, dominant species are combined across strata. Vegetation in a plot is deemed to be hydrophytic by the dominance ratio if >50% of the dominant species across all strata combined are rated obligate (OBL), facultative wetland (FACW), or facultative (FAC) on the appropriate regional list of plant species that occur in wetlands (Reed 1988). An advantage of dominance ratios is that they are generally easy to implement in the field. However, because they ignore non-dominant species and the relative abundance of dominant species, dominance ratios are often biased towards positive wetland determination, particularly at sites with many FAC species (Wakeley and Lichvar 1997; Dewey et al. 2006).

An alternative approached for determining whether a plant community is hydrophytic is the "weighted average" method, also called the "prevalence index" (Carter et al. 1988; Wentworth et al. 1988). All species in a community (not just a subset of dominants) are used to calculate a weighted-average wetland indicator status by multiplying indicator status category ratings (OBL = 1, FACW = 2, FAC = 3, FACU = 4, and UPL = 5) by the relative abundance of species in the sample. Typically, a plant community is considered to be hydrophytic if the prevalence index is less than or equal to 3.0 (FICWD 1989; SCS 1994). The prevalence index approach can be applied to community data collected from both plotbased and transect-based (point-intercept) methods using variants of the same general approach.

The results from dominance ratio and prevalence index approaches often disagree. For example, Wakeley and Lichvar (1997) found that outcomes based on the two methods disagreed nearly 20% of the time. When quantitatively compared, hydrophytic vegetation decisions based on the prevalence index tend to agree more closely with hydric soil and hydrology indicators than decisions based on dominance ratios in several wetland types (Best et al. 1990; Atkinson et al. 1993; Davis et al. 1996; Wakeley et al. 1996).

Dewey et al. (2006) examined the results of several approaches for determining the prevalence of hydrophytes for delineation purposes in a bottomland hardwood forest in Texas. The authors evaluated outcomes for

different variants of the dominance ratio method. In addition to the standard 50/20 rule, they evaluated their community data using the FAC neutral option whereby FAC species were removed from consideration, and a weighted FAC neutral test in which species with OBL and UPL indicator values were weighted twice that of FACW and FACU. The purpose was to reduce the effect of the ubiquity of FAC species on the determination (Dewey et al. 2006). They also examined the effect of using a 10% threshold for determining whether individual species should be considered dominants (50/10 rule) to evaluate whether such a change significantly altered the outcome. Outcomes from the dominance ratio methods were compared with those from an analysis of prevalence index. The authors concluded that the prevalence index was the most stringent method of assessment and the method that provided the best agreement with soil and hydrology parameters at their site.

#### 2.6 Boundary determination

Ecological boundaries, such as those separating uplands and wetlands, vary in spatial structure and function. Boundaries may be defined as places where rates of spatial change are greatest; however, in practice, the challenge is to define how rapid a rate of change is necessary to constitute a boundary (Jacquez et al. 2000; Kent et al. 2006). Quantitative approaches for boundary determination can include univariate or multivariate techniques (Cornelius and Reynolds 1991; Choesin 1997; Kirkman et al. 1998; Attrill and Rundle 2002; Choesin and Boerner 2002; Kent 2006).

A variety of factors can influence the characteristics of ecological boundaries such as the one between wetlands and adjacent uplands. Relatively distinct and narrow boundaries occur where sharp environmental changes exist; for example, where there are rapid changes in topography or geomorphology (Johnston and Naiman 1987; Kirkman et al. 1998; Choesin and Boerner 2002). Where changes in key environmental drivers such as hydrologic regime are more gradual, boundaries can be blurred and may be relatively transient. Spatial heterogeneity in underlying environmental drivers can create background noise, making detection of boundaries more difficult (Kent et al. 1997). For example, pit and mound topography created by tree-fall events can create greatly contrasting microsites that support upland to obligate wetland species over short spatial scales. In these situations, the choice of sampling unit size and shape may strongly influence the outcome of analyses.

Under current regulatory practices, methods for determining wetland boundaries are based on the three-factor approach presented in the 1987 Manual. The particular reliance on these specific wetland indicators differentiates these methods from those used in many research studies, which may rely on a single factor such as vegetation or a different combination of characteristics. Wetlands may exhibit sharp boundaries between plant communities (ecotones), a gradual boundary (ecocline), or some combination of the two.

Many scientific studies that have evaluated the effectiveness and efficiency of different vegetation sampling designs emphasize sampling areas of heterogeneous vegetation and specifically avoid sampling boundaries (Segadas-Vianna 1951; Glaser et al. 1981; Benedict 1983; Lenssen et al. 2000). Ecological boundaries have been defined in a variety of ways and can refer to real or abstract conceptual features (Strayer et al. 2003). Boundaries may or may not correspond with obvious physical changes in nature and may be qualitatively or quantitatively defined and delineated.

The specific choice of metrics and sampling unit characteristics can influence the outcome of boundary determinations. For example, using woodland data, Fortin (1997) found that ecological boundaries differed depending on whether determinations were based on density, percent coverage, or presence—absence data. Whereas boundaries showed significant overlap, a spatial lag was found between the boundaries based only on trees and those based on shrubs alone (Fortin 1997). Traditional approaches used to describe vegetation change across boundaries include transects or plots and similarity coefficients (Kent et al. 1997, 2006).

The emphasis on the three-factor approach in delineation procedures as prescribed in the 1987 Manual both simplifies and complicates decisions regarding boundary determinations. Hydric soil indicators can be determined at a finer scale than vegetation, which is explicitly defined as a community metric. The concept of "a prevalence of hydrophytes" logically requires multiple plants and some minimum area. Unfortunately, this two-dimensional community concept cannot be completely reduced to the one-dimensional realm of boundary lines.

Anecdotally, many delineators describe a process where initial boundaries are at first conceptualized based on soils information, with vegetation serving a more confirmatory role. Others report using the opposite

approach: observations of vegetation changes lead to a boundary that is sharpened by soil investigations. A downside of the three-factor approach is that outcomes are not comparable to those from most methods used in community and landscape ecology for boundary determination. This is a result of using three specific variables, each of which varies in the relative scale that it is assessed. Wetland hydrology, for example, is typically inferred for relatively broad spatial scales. Depending on the particular vegetation type present (e.g., forest versus herb-dominated), vegetation may be seen at a range of spatial scales, whereas soils are assessed at point locations (i.e., a soil pit).

A transect-based approach has been used for characterizing ecosystem boundaries, but problems may include failure to adequately account for heterogeneity in vegetation and environmental variables (Carter et al. 1994; Kent et al. 1997; Kirkman et al. 1998). Typically, transects are placed perpendicular to dominant environmental gradients; however, these gradients are rarely defined *a priori* and may be highly subjective, particularly when no obvious topographic gradients are present. Additionally, variation in transect length can make comparisons among transects difficult.

Several authors have advocated the use of two-dimensional sampling with wide, rectangular sampling grids (Fortin and Drapeau 1995; Fortin and Dale 2005). Various sampling designs may be adopted within the rectangle, but a key element is the possibility of sampling lateral variation across transitions. If the entire rectangular area is sampled using contiguous quadrats, the sampling area may be described as a "tranome" (Kent et al. 1997). As with other methods, results may vary if the scales of observation or analysis vary due to changes in sampling unit size, shape, spacing or extent (Dungan et al. 2002).

Alternative methods have been developed for quantitative boundary determination. Examples include moving split window (MSW) analysis, the Mantel test, pattern analysis, semi-variograms, spectral analysis, and analysis for spatial autocorrelation (Kent et al. 1997; Kirkman et al. 1998; Choesin and Boerner 2002; Dale et al. 2002; Husch 2003; Borcard et al. 2004). Typically, these methods first involve description of spatial rates of change in measured variables and then mapping or thresholding to determine the zones or locations of greatest change (Barbujani et al. 1989; Kent et al. 2006).

The MSW approach, in particular, shows promise for boundary delineation in wetlands. In the MSW approach, multiple plots consisting of double windows are sampled and statistically compared using attribute values such as percent of cover in each window. Similarities are typically calculated using the squared Euclidean distance (SED), calculated as the squared difference between the means of each variable in adjacent windows, and summed across all variables (Figure 2) (Choesin and Boerner 2002; Fortin and Dale 2005). This general technique has been successfully used to delineate boundaries in a variety of systems—not just vegetation (Panis and Verheyen 1995).

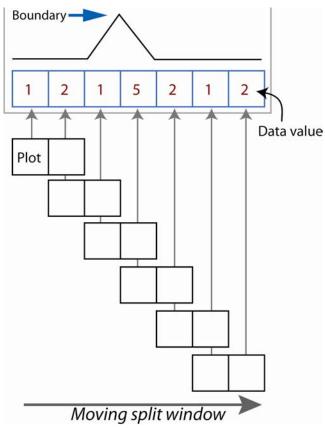


Figure 2. Hypothetical data illustrating a split moving window analysis along a transect of seven contiguous sampling units and a split moving size of two sampling units. The strongest boundary peak is located between sampling locations D and E, with a secondary peak located between each H and I. (After Fortin and Dale 2005.)

The MSW method is best used on data from contiguous sample plots along belt transects. One disadvantage of using transects is that their placement, spacing, and sample intervals are generally subjectively determined, potentially introducing bias to the data (Choesin and Boerner 2002). Commonly, transects are positioned perpendicular to perceived boundaries, which may or may not be the true gradients that drive

vegetation patterns. However, compared with traditional multivariate analysis methods used in vegetation classification, MSW is capable of measuring the strength, width, and abruptness of boundaries and requires less effort for data analysis and interpretation (Choesin and Boerner 2002). Additionally, MSW is not restricted to vegetation data. Environmental data can be directly incorporated into analyses, and the approach can be used to detect changes in vegetation without identifying plants to the species level (Choesin 1997; Choesin and Boerner 2002).

Quantitative approaches for boundary determination, such as MSW analysis, could provide much greater rigor to the key task of delineators—fixing the spatial location of the wetland—upland boundary. Under current methods, this decision is largely subjective and based on the experience of the delineator as much as on any specific data collected on site. MSW analysis could conceivably be used in instances where objective and more rigorous methods are required; for example, where specific boundary determinations are part of litigation. However, given their greater complexity and analysis requirements, quantitative approaches may not be feasible for routine delineations unless they are extensively modified.

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# 3 Synthesis

The utility of a particular method for characterizing vegetation varies based on several criteria. In general, the choice of sampling methodology should be made following careful consideration of the specific sampling objectives, the necessary accuracy and precision of the data, and the available time and resources for sampling. More robust methods should be selected when the accuracy and precision of the data are critical, such as for long-term monitoring or detailed functional assessment. Part of defining sampling objectives and comparing different sampling methodologies is explicitly identifying the most important performance metrics (accuracy, sampling efficiency, repeatability), as well as the resources that will typically be available for sampling.

The choice of criteria for determining whether a site supports a predominance of hydrophytes is an important decision. The dominance ratio method for determining whether vegetation on the site is dominated by hydrophytes is a commonly used approach and is based on information from dominants only. An advantage of this approach is that, by excluding less common species from consideration, methodological decisions such as plot size and number are less important because simply detecting dominants is easier than developing accurate estimates of species richness.

Prevalence indices are another accepted method for determining whether a site is dominated by hydrophytes. This approach has several advantages. The prevalence index is on more sound conceptual footing and leverages information from the full complement of species detected in a survey. This approach is in greater congruence with the larger sampling literature, where approaches such as weighted averaging have a usage history. In contrast, the 50/20 rule is restricted to the realm of wetland delineation. The two methods often disagree, with the tendency of dominance ratios to more often result in a positive determination of hydrophytic vegetation. Both methods are only as reliable as the underlying data.

In general, the choice of specific method for sampling vegetation should be based on the necessary accuracy, precision, and sampling efficiency, although, importantly, these cannot be simultaneously optimized. The "best" method, then, is the one that optimizes these attributes in light of the specific sampling goals. For example, absolute accuracy is of less importance for delineation than it is for monitoring because the primary

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concern is relative abundance (e.g., what is the relative abundance of upland species versus obligate wetland species). When the goal is to determine the dominant species, and determination of the full complement of species on a site is less important—typically the case for delineation when using the dominance ratio—the consequences of different "tactical" choices such plot size are minor.

Traditional approaches for sampling vegetation have focused on identifying and characterizing areas with relatively uniform species composition and structure. This represents an important limitation when the goal of the analysis is to characterize the boundaries between such units (Risser 1995; Choesin and Boerner 2002). By focusing on interior structure and composition, these approaches generally fail to provide direct information to guide delineation of boundaries.

Guidelines presented in the 1987 Manual provide considerable latitude in selecting specific methods. However, the flip side of this flexibility is a lack of clarity about the most appropriate sampling strategy for a given situation. Regionalization of methods in the 1987 Manual is a step towards providing more specific guidance for delineation, but future efforts should provide guidance tailored to the particular wetland types within a region.

In evaluating potential changes to existing sampling procedures, it is important to ensure that the procedures can be effectively applied by practitioners in the field. The botanical experience of many regulators and consultants is limited, and delineations are often conducted under less-than-ideal conditions (e.g., outside of the growing season). The procedures used for routine delineations must be implemented in a reasonable time in the field. They cannot generally match the rigor of methods used in more detailed scientific studies. However, to serve as a sound basis for regulatory decisions, methods must provide adequate precision and accuracy.

Methods chosen for widespread application should be standardized to the greatest degree feasible, although ecological realities demand that regional and/or ecosystem-specific sampling strategies should be pursued. In general, methods should be carefully crafted to address the primary sampling objectives. Given the particular emphasis on boundary detection and delineation in wetland regulation, sampling methods should reflect this focus.

## 4 Recommendations

Wetland delineation protocols used for regulation have been used for more than 20 years. During this period, significant research on vegetation sampling and wetland science has taken place and should be addressed in revisions to federal wetland delineation procedures. However, in many instances, the fundamental purpose of wetland delineation procedures—objective and repeatable determination of a fixed boundary in space—is contrary to the scientific reality that clear boundaries often do not exist. Plant communities are, by their very nature, abstract concepts. Species shift locations in space and time, and the environmental drivers of plant distribution are highly variable.

Fundamentally, plant communities lack the same structural and functional integration that characterizes cells or individual organisms (Rowe 1961). Decades of research in plant ecology have largely rejected the concept espoused by Clements (1916) that plant communities are analogous to organisms, with clear boundaries and developmental pathways. Consequently, the location of a specific boundary in the field must often be ultimately acknowledged as arbitrary relative to actual structure or ecological function (Fitzsimmons 1999).

What a delineation protocol can and must do is provide a fair and objective standard that consistently results in the same outcome. All mapping and delineation procedures are only meaningful in the context of a specifically defined classification. For wetlands, the key elements of this classification are those factors defined by statute or regulation. Fidelity to these defining factors is the ultimate standard by which any sampling procedure must be judged. Of course, these factors should be based on ecological principles, particularly because the underlying rationale for regulating wetlands in the first place is derived from their important ecological functions and values provided to society.

For all of these reasons, formulating firm rules for sampling that apply to all of the diverse wetland types that occur nationally is difficult. Some general recommendations for consideration in revising existing vegetation sampling procedures for wetland delineation for regulatory purposes are as follows:

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1. Explicitly define the vegetation criteria necessary for determining wetland status and boundaries. Will information regarding dominant species alone be sufficient to make determinations, or is a more complete understanding of species composition, including the presence and abundance of less common species, necessary? In some regards, dominant species are the worst indicators, as they typically have broad niches. However, sampling techniques for characterizing dominant species are generally robust and forgiving with regard to performance and sampling characteristics such as plot size, plot shape, and minimum plot number. Performance differences among methods, whether evaluated in reference to accuracy, precision, or efficiency (or some combination), are generally more important when the goal is to characterize diversity or to allow for the accurate assessment of trends in community characteristics (McCune and Grace 2002; Stohlgren 2007).

- 2. Explicitly identify and rank the relative importance of different performance measures when evaluating possible sampling strategies. Absolute accuracy, while always a desirable characteristic, must be realistically viewed in light of other performance criteria such as precision and efficiency. Marginal increases in accuracy of a particular sampling method may require significantly greater sampling effort; the benefits of such accuracy gains must be subjected to some sort of cost/benefit analysis. For regulatory purposes, the precision of sampling may be more important than absolute accuracy. Sampling requirements, such as the minimum number of plots necessary to achieve an acceptable level of accuracy or precision, will vary depending on whether dominant species alone are being evaluated, or whether information regarding composition of the broader plant community is necessary.
- 3. Tailor the specific methods to the finest level possible. The current trend towards presenting regional guidance for delineation procedures is a positive step towards improving the accuracy and precision of delineation methods. However, within currently defined regions, there is considerable variation in composition and structure of wetlands. This variation significantly affects the relative performance of any particular sampling procedure; therefore, guidance regarding key sampling decisions such as the number of plots should be provided at the finest feasible level of ecological classification.
- 4. Conduct pilot testing in a wide range of wetland types for any proposed revisions to existing methods. Approaches such as

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MSW analysis show promise for use in boundary determination. This technique has been successfully used but for a very limited range of wetland types. To be recommended for more broad usage, extensive testing is necessary.

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## 13. SUPPLEMENTARY NOTES

### 14. ABSTRACT

15. SUBJECT TERMS

This document reviews sampling issues and methods for characterizing vegetation, with an emphasis on delineating wetland boundaries under Section 404 of the Clean Water Act. Numerous vegetation sampling methods have been developed, but no single approach is optimal for all sampling objectives, vegetation types, and ecological settings. Methods vary in relative precision, accuracy, and efficiency, although selection of methods should also explicitly consider sampling objectives, resources, and data requirements. Key considerations include the choice of metric(s), selection of sampling units, and logistical issues such as time required for sample collection. Common metrics include frequency, cover, and biomass, with each providing different inferences to community characteristics. Vegetation may be assessed using plots, transects, or points as sampling units, and sampling may treat separate vegetation layers (i.e. strata) separately or together. Sampling units may be located subjectively or by using one of many available randomized sampling strategies, although randomization is essential for any statistical analysis. The document reviews general concepts of relative abundance and dominance and discusses the dominance ratio and prevalence index approaches, which are commonly used in wetland delineation. Lastly, sampling approaches traditionally used to assess community characteristics are contrasted with those specifically developed for boundary determination.

| Sampling<br>Vegetation          |             | Wetlands     |                               |                        |                                 |
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