



Field Sampling of Vegetative Carbon Pools in Coastal Ecosystems

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GENERAL CONSIDERATIONS

The vegetative blue carbon pool consists of three different components:

- The living, aboveground biomass dominated by herbaceous (for seagrass and tidal salt marsh) and woody (for mangroves) plant mass. This can also include epiphytic organisms (e.g., algae and microbes living on the main plants) and aerial roots (pneumatophores).
- The living, belowground biomass dominated by underground roots and rhizomes.
- The non-living, aboveground biomass, including detritus consisting mainly of leaf litter (found in all three ecosystems), algae, or in mangroves dead and downed wood.

The protocols used to determine the amount of carbon in each pool—in each ecosystem—will vary depending on vegetation type and density. Allometric equations are used to describe the relationship between “measurable parameters” (height, width, circumference, etc.) and total biomass. These equations are commonly used to determine the biomass of materials where it is impractical, overly destructive, or unwise to take an entire sample back to the laboratory (e.g., trees and large shrubs). Many well-established allometric equations exist in scientific literature (many are used in this chapter), and it is recommended to use equations that have been established for similar vegetative species and locations as those found in the study site being investigated.

In all cases, the carbon pool for each vegetation type is determined by multiplying the biomass of each type of plant material (e.g., wood, leaf litter, roots, etc.) by the corresponding carbon conversion factor. The carbon conversion factor represents the fraction of vegetation that is carbon. For example, if the living aboveground wood has been determined to be 45% carbon, then the carbon conversion factor is 0.45. The carbon conversion factor is then multiplied by the total biomass of the above ground wood pool for that plot to determine the amount of carbon found in the aboveground wood pool for a certain area.

The goal of Chapter 4 is to utilize ecosystem-specific techniques to determine the biomass and the organic carbon content (% C_{org}) for each of the relevant blue carbon pools. Once the carbon content for all carbon pools has been established, they are then added together to determine the carbon content of the biomass per unit area for a specific system (MgC/hectare). This value is then added to the soil carbon pool specified for a certain soil carbon depth (Chapter 3) to determine the total carbon stock (MgC/hectare-depth) for a blue carbon ecosystem.

MANGROVES

Mangrove ecosystems are populated by halophytic trees, shrubs, and other plants that grow in brackish to saline tidal waters of tropical and subtropical coastlines (Mitsch & Gosselink 2007). In general, mangroves are restricted to the intertidal zone from approximately mean sea level to the highest high tide water level.

Mangroves are classified into four major associations based on differing vegetative structures that correspond to physical, climatic, and hydrologic features of the environment in which they exist: (1) oceanic fringing mangroves, (2) riverine or estuarine mangroves, (3) basin mangroves, and (4) dwarf or scrub (or chaparro) mangroves (Cintrón *et al.* 1978; Mitsch & Gosselink 2007) (**Fig. 4.1**). These classifications correspond to aboveground biomass ranging from more than

500 MgC/ha in riverine and fringe mangroves (such as in Asia-Pacific regions) to about 8 MgC/ha for dwarf mangroves (Kauffman & Cole 2010; Kauffman *et al.* 2011).



Figure 4.1 Classification of mangroves. (A) Oceanic fringing mangroves (© Enrico Marone, CI), (B) Riverine or estuarine mangroves (© Ginny Farmer, CI), (C) Basin mangroves (© Colin Foster, CI), and (D) Dwarf or scrub mangroves (© Catherine Lovelock, UQ)

Flooding regimes throughout the mangrove habitat cause saline or brackish coastal wetland environments that often consist of anoxic (low oxygen levels) sediments. As such, mangroves possess a number of adaptations to facilitate survival in these unique environments. Most notably, this includes aboveground roots (e.g., their characteristic stilt roots and pneumatophores), which allow gas exchange for belowground root tissues. The vegetation also varies greatly in structure (tree height, density, and species composition) and function, due to differences in temperature, rainfall, hydrology, and substrate (Saenger & Snedaker 1993). Mature mangroves may range from shrub-like stands less than 1 m in height to large trees with trunk diameters > 1 m.



Figure 4.2 Height differences among mangrove vegetation. (A) Small dwarf mangroves < 2 meter in height (© C.I. Feller, SERC), (B) Larger mangroves several meters tall (© Andreas Hutahean, KKP)

Despite several similarities, mangroves are quite different from upland forests in both composition and structure. Mangroves have stilt roots and pneumatophores, and they usually do not have significant understory vegetation or a well-developed floor litter layer as crabs are usually extremely efficient consumers of fallen leaves and litter is transported away by tides. Because of these and other differences between the structure and environment of mangroves and upland forests, approaches to quantifying their composition, structure, carbon stocks, and status differ. However, some approaches to upland forest sampling may provide guidance in project implementation and design. Notable examples can be found in the scientific literature (Pearson *et al.* 2005; Pearson *et al.* 2007; GOFC-GOLD 2009).

Field Sampling Considerations

Mangroves ecosystems are often extremely difficult environments to conduct field assessments and sampling. The trees often have extremely high stem densities with abundant stilt roots and/or pneumatophores. Project areas are frequently dissected by tidal channels which are difficult to cross. The entire ecosystem may be flooded, especially during high tides. These and a number of other hazards limit mobility and create safety concerns. Most mangroves are also subject to semi-diurnal tidal cycles and can only be sampled during low tides, limiting both the timing and duration of the sampling, especially for components on the forest floor. In the lowest elevation mangroves, sampling may be limited to low tidal periods of as little as 3 to 4 hours. This narrow time window necessitates an efficient sampling plan.

In Chapter 2, we describe approaches to determining the number and location of sampling plots within a project area or strata. Since the various carbon pools within mangroves have distinctly different scales and levels of effort required for sampling, it is usually necessary to assess different components with different size sampling areas. The biomass of trees, shrubs, herbs, lianas, and palms along with non-living vegetation like leaf litter and downed wood must be determined separately and sampled at appropriate scales (**Fig. 4.3**). For example, to get a representative sampling of trees, the sample area may need to be large (e.g., 50 m x 50 m), but if you then want to sample the leaf litter, it is not practical or necessary to collect all the leaf litter in an area that big. Thus a smaller subplot size (e.g., 2 m x 2 m) is more appropriate.

Once size and location have been determined, it is necessary to decide if the sampling plots will be permanent or temporary. Permanent plots are used if the same location will be assessed in the future to determine change. Temporary plots are used when sampling will only be done once or when permanent plots are not feasible. We give some guidance on establishing permanent sampling plots vs. temporary plots in Chapter 2. For further information on the establishment of permanent plots and sampling methods, we recommend the Amazon Forest Inventory Network (www.rainfor.org/) and the Center for Tropical Forest Science (www.ctfs.si.edu/group/Resources/Methods) websites.

With plot size, scale, and type determined, the next step is to assess the carbon content of each relevant carbon pool.

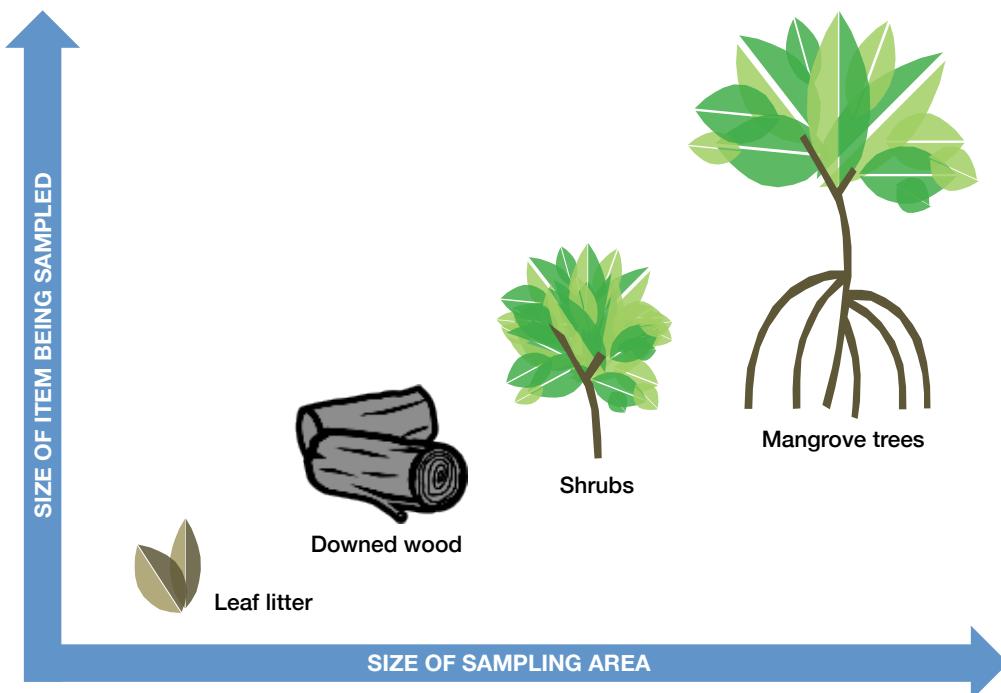


Figure 4.3 Plot scale depends on the component being assessed. Large trees require a large enough area to allow for a representative sampling, smaller trees need less area to get a representative sample, leaf litter, lianas, dead wood, and pneumatophores components are so small and numerous that a relatively small area is adequate for sampling (Kauffman & Donato 2011).

Biomass Estimates

This section provides guidance on how to measure aboveground biomass across a range of vegetation types likely to be encountered in the field. Some differences in sampling procedures are required to accommodate differences in growth forms.

LIVE TREES

Trees dominate the aboveground carbon pool in mangroves, and both their presence and condition are indicators of land-use change and ecological condition. It is essential to measure trees thoroughly and accurately. Basic data that must be recorded for all individual mangrove trees in a plot include:

- Species (there are typically few species present in mangroves, species can usually be identified with on-site training);
- Main stem diameter at breast height (dbh);
- Tree height if feasible; and
- Location and ID.

It is recommended that all live trees be sampled and recorded over the entire plot area, particularly for permanent plots where trends in carbon are being monitored. However, this is different from surveys of upland forests, where only trees greater than 10 cm dbh are measured to standardize methods (GOFC-GOLD 2009). Smaller trees are left out of the carbon calculations because they often constitute a relatively insignificant proportion of the total upland forest carbon stock (Cummings *et al.* 2002). For many mangroves, however,

smaller trees can dominate the stand composition and thus must be included (Lovelock *et al.* 2005; Kauffman & Cole 2010). A tree is included in the survey if at least 50% of the main stem is rooted inside the plot perimeter.

In the interest of efficiency, trees that are smaller in diameter may be measured in subplots to reduce the number of measurements necessary. For example, Kauffman and Cole (2010) measured all trees > 5 cm dbh over the entire plot area, while trees < 5 cm dbh were measured in smaller subareas of known size. The total number of small trees could then be estimated by assuming a constant density over the full plot area.

If numerous seedlings are present, the seedlings can be recorded as a simple count of individuals in a subarea. For our purposes, a seedling is defined as a woody plant with a height of 10–30 cm (Esquivel *et al.* 2008). To determine the carbon content associated with seedlings, a random sample should be collected from outside the plot area for analysis (only necessary to collect from outside the plot if using permanent plots, the idea is to include seedlings in the analysis but leave them in the plot so they can be monitored over time). Carbon content can be determined by drying the seedlings to determine biomass followed by laboratory analysis using an elemental analyzer (Chapter 3), but in many cases it is possible to find a published carbon conversion factor for specific tree species. The average carbon content is then multiplied by the density (seedlings per unit area) to determine their contribution to the plot and strata biomass.

To determine the biomass of mangrove trees, existing allometric equations are applied (Chave *et al.* 2005). Allometric equations give established relationships between the biomass of whole trees (or their components) and readily measured parameters. Accurate species identification is important as it allows the selection of the most appropriate allometric equation for each measured individual mangrove tree. Common parameters include tree diameter, wood density (**Table 4.2**) and tree height.

Diameter at Breast Height (dbh): The diameter of the tree is typically used to calculate the tree volume. The diameter of the tree's main stem is typically measured at 1.3 m above the ground, which is also called the dbh. These measurements are usually made with a diameter tape (if multiple measurements are needed) or tree callipers (for a single measurement for rapid assessment). This is not always a straightforward process due to anomalies in stem structure. **Fig. 4.4** gives an overview of the location for measurement for a variety of different tree configurations.

- If the tree is fairly straight with a tall trunk the dbh can be measured from the ground parallel to the trunk (**Fig 4.4A**)
- If the tree is on a slope, always measure on the uphill side (**Fig 4.4B**)
- If the tree is leaning, dbh is taken according to the trees natural height parallel to the trunk (**Fig 4.4C**)
- If the tree is forked at or below 1.3 m then measure just below the fork (**Fig 4.4D**)
- If the fork is very close to the ground measure as two trees (**Fig 4.4E**)
- For trees with tall buttresses exceeding 1.3 m above ground level, stem diameter is usually measured directly above the buttress (**Fig 4.4F**).

- For stilt rooted species (e.g., *Rhizophora* spp.), stem diameter is often measured starting above the highest stilt (**Fig 4.4G**). For some individual trees with prop roots extending well into the canopy it is not necessary, practical, or accurate to measure above the highest prop root and, typically, tree diameter is measured above the stilt roots where a true main stem exists.

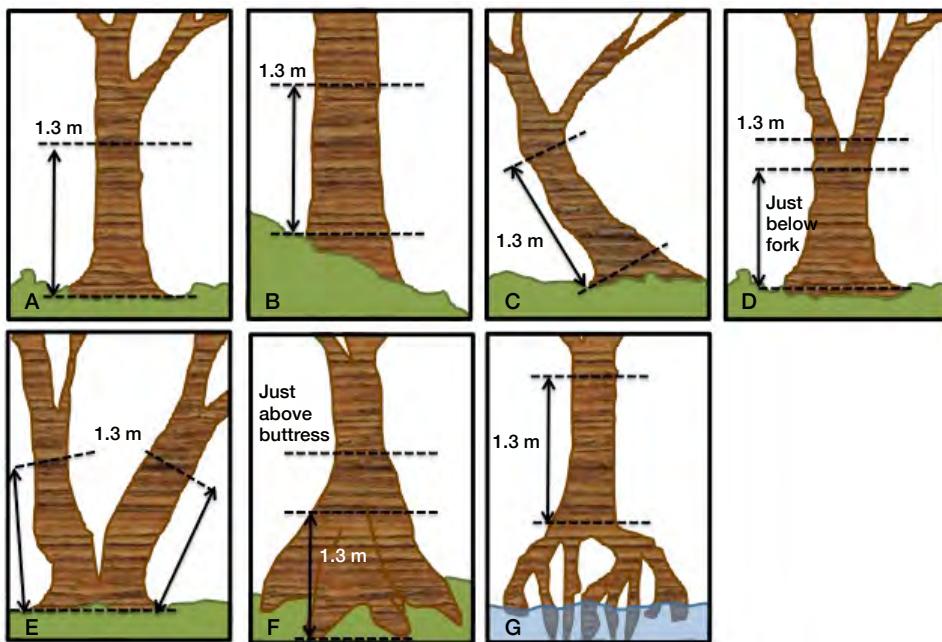


Figure 4.4 Estimating diameter at breast height for irregular mangrove trees (modified from Pearson, et al. 2005)

In permanent plots, it is quite important to mark the point of measurement when it is not at 1.3 m above ground level so that repeated measurements can be taken at the same location. This is accomplished by placing tree tags and/or by painting a ring at the exact point of measurement. In some studies, point of measurement is marked with a stainless steel nail. However, when nails are used trees tend to develop wound wood (bulges) where the wood is damaged by the nail. This can cause overestimates of tree growth and therefore is not recommended.

Wood Density: Wood density describes the relationship between the wood's dry weight (g) and the wood's volume (cm^3). The part of the plant (branches, main stem, bark, etc.) harvested for wood density depends on the practicality of obtaining samples and the level of accuracy desired. Wood density requires measuring both the volume of fresh samples and the oven-dry mass of several samples of wood (ideally $n > 25$ per sample type). Samples are commonly taken by removing small portions of the bark, cutting segments of branches (~2.5 cm), and using an increment borer on the main stem (taken from a consistent height). As a rough guideline, each piece collected for analysis should have a mass of between ~0.5 and 50 g.

Volume is obtained by determining each fresh sample's submerged mass. On a digital balance, place a container of a size sufficient to submerge each sample. Add water to the container, not to the top, but to a height that will allow the displacement of water without it spilling over the sides of the container. Each sample is attached to a needle attached to a ring stand above the scale. The sample is then submerged (without touching the bottom and sides of the container) and the change in mass is recorded. The change in mass (g) divided by

the density of water (g/cm^3) gives the volume of the sample. The density of water is $1 \text{ g}/\text{cm}^3$, thus the resulting increase in mass shown on the scale is equivalent to the volume displaced by the sample.

Dry weight is obtained by drying the wood samples in a well-ventilated oven at 100°C until constant mass is obtained (typically 24–72 hours, but the time will depend on the size of the sample). We recommend drying at 100°C because water within the cell wall can only be completely dried off at these temperatures. For each sample, calculate the wood density using the following equation and determine the mean for each sample type.

- Wood density (g/cm^3) = Dry weight (g) / volume of fresh wood (cm^3)

Wood densities for live trees (which may be different from densities of downed woody debris) are required for certain allometric biomass equations, including the general mangrove equations (see next section). Wood densities of individuals of the same species have been shown to vary greatly between sites. As such, it is desirable to use site-specific wood densities estimated by laboratory analysis of samples taken from the field. If that is not possible local forest agencies might know wood densities of specific species. Other general sources for wood density include the World Agroforestry Database (World Agroforestry Center 2001) and sources books produced by the U.S. Department of Agriculture (Hidayat & Simpson 1994). Examples of wood density for common mangrove species are given in **Table 4.1**.

Allometric equations for mangrove tree biomass: A number of references report allometric equations for mangrove biomass (Saenger 2002; Chave *et al.* 2005; Smith III & Whelan 2006; Komiya *et al.* 2008; Kauffman & Cole 2010; Kauffman & Donato 2011) and examples complied by Kauffman and Donato (2011) are found in **Table 4.2** (for equations with parameters of dbh and wood density) and **Table 4.3** (for equations with parameters of dbh, wood density, and tree height). Before deciding which allometric equation to use, consider the geographic origin and species that composed the data set from which the equation was derived. Ideally, it is best to use a species-specific equation developed in the region where the sampling is to occur. Given the differences in structure and wood density among species, species-specific equations are likely to yield greater accuracy than general equations. It is also critical to note the maximum diameter from which the equation was derived. Applying the equation to trees that exceed the maximum diameter (D_{\max}) can lead to a statistically significant overestimation of the biomass. In very data poor situations, general allometric equations for mangrove trees can be used but the uncertainty is relatively high.

Table 4.1 Wood density of mangrove species (1 ton/m³ = 1 g/cm³) (Saenger 2002; Komiyama *et al.* 2005; Donato *et al.* 2012; World Agroforestry Center 2001)

SPECIES	n	AVERAGE WOOD DENSITY (TONNES/m ³)	STANDARD ERROR
<i>Avicennia germinans</i>	5	0.72	0.04
<i>Avicennia marina</i>	6	0.62	0.06
<i>Avicennia officinalis</i>	3	0.63	0.02
<i>Bruguiera gymnorhiza</i>	8	0.81	0.07
<i>Ceriops decandra</i>	2	0.87	0.10
<i>Ceriops tagal</i>	7	0.85	0.04
<i>Excoecaria agallocha</i>	7	0.41	0.02
<i>Heritiera fomes</i>	3	0.86	0.14
<i>Heritiera littoralis</i>	6	0.84	0.05
<i>Laguncularia racemosa</i>	3	0.60	0.01
<i>Rhizophora apiculata</i>	4	0.87	0.06
<i>Rhizophora mangle</i>	7	0.87	0.02
<i>Rhizophora mucronata</i>	9	0.83	0.05
<i>Sonneratia alba</i>	6	0.47	0.12
<i>Sonneratia apetala</i>	2	0.50	0.01
<i>Xylocarpus granatum</i>	7	0.61	0.04
Average		0.71	0.02

Table 4.2 Allometric equations for computing biomass of mangrove trees where only parameters of diameter (dbh) and wood density are used. The general equations include all aboveground biomass. Individual species equations have been broken down by component. B = Biomass (kg), D = Diameter at breast height (cm), ρ = Wood density (g/cm³), D_{\max} = maximum diameter of sampled trees (cm) (Modified from Kauffman and Donato, 2011).

SPECIES GROUP	N	D _{max}	LOCATION	BIMASS EQUATION	R ²
General Equation	84	42	Americas	$B = 0.168 * \rho * (D)^{2.471}$	0.99
General Equation	104	49	Asia	$B = 0.251 * \rho * (D)^{2.46}$	0.98
<i>Avicennia germinans</i>	25	42	French Guinea	$B = 0.14 * D^{2.4}$	0.97
<i>Avicennia germinans</i>	8	21.5	Florida, USA	$B = 0.403 * D^{1.934}$	0.95
<i>Bruguiera gymnorhiza</i> (leaf)	17	24	Australia	$B = 0.0679 * D^{1.4914}$	0.85
<i>Bruguiera gymnorhiza</i> (wood)	326	132	Micronesia	$B = 0.0754 * \rho * D^{2.505}$	0.91
<i>Laguncularia racemosa</i>	70	10	French Guinea	$B = 103.3 * D^{2.5}$	0.97
<i>Laguncularia racemosa</i>	10	18	Florida, USA	$B = 0.362 * D^{1.930}$	0.98
<i>Rhizophora apiculata</i>	20	30	Malaysia	$B = 0.1709 * D^{2.516}$	0.98
<i>Rhizophora apiculata</i> (wood)	191	60	Micronesia	$B = 0.0695 * \rho * D^{2.644}$	0.89
<i>Rhizophora apiculata stylosa</i> (leaf)	23	23	Australia	$B = 0.0139 * D^{2.1072}$	0.86
<i>Rhizophora apiculata stylosa</i> (stilt roots)	23	23	Australia	$B = 0.0068 * D^{3.1353}$	0.97
<i>Rhizophora mangle</i>	14	20	Florida, USA	$B = 0.722 * D^{1.731}$	0.94
<i>Rhizophora spp</i> (racemosa and mangle)	9	32	French Guinea	$B = 0.1282 * D^{2.6}$	0.92
<i>Sonneratia alba</i> (wood)	346	323	Micronesia	$B = 0.3841 * \rho * D^{2.101}$	0.92

Table 4.3 Allometric equations for computing biomass of mangrove trees where parameters of diameter (dbh) and height are used for species specific equations, and diameter and wood density are used for general equation. The general equations include all aboveground biomass. Individual species equations represent wood mass and do not include leaves or roots. B = Biomass (kg), H = Height (m), D = Diameter at breast height (cm), ρ = Wood density (g/cm^3), D_{\max} = maximum diameter of sampled trees (cm), H_{\max} = maximum height of sampled trees (Modified from Kauffman and Donato, 2011).

SPECIES GROUP	N	D_{\max}	H_{\max}	BIMASS EQUATION	R^2
General Equation	84	42		$B = 0.0509 * \rho * (D)^2 * H$	
<i>Bruguiera gymnorhiza</i>	325	132	34	$B = 0.0464 * (D^2 H)^{0.94275} * \rho$	0.96
<i>Lumnitzera littorea</i>	20	70.6	19	$B = 0.0214 * (D^2 H)^{1.05655} * \rho$	0.93
<i>Rhizophora apiculata</i>	193	60	35	$B = 0.0444 * (D^2 H)^{0.96842} * \rho$	0.96
<i>Rhizophora mucronata</i>	73	39.5	21	$B = 0.0311 * (D^2 H)^{1.00741} * \rho$	0.95
<i>Rhizophora</i> spp.	265	60	35	$B = 0.0375 * (D^2 H)^{0.98626} * \rho$	0.95
<i>Sonneratia alba</i>	345	323	42	$B = 0.0825 * (D^2 H)^{0.89966} * \rho$	0.95
<i>Xylocarpus granatum</i>	115	128.5	31	$B = 0.0830 * (D^2 H)^{0.89806} * \rho$	0.95

Variance from Allometric equations: There is a great deal of variation in wood density, morphology, and height-diameter relationships between sites, which can affect the accuracy and utility of any given allometric equation. Different equations can yield very large differences in biomass predictions. In **Fig. 4.5**, predictions generated from different allometric equations using the same dataset from a mangrove stand in Yap, FSM, are shown (Kauffman et al. 2011). The biomass prediction of the largest *Bruguiera* tree in this mangrove forest (69 cm dbh)

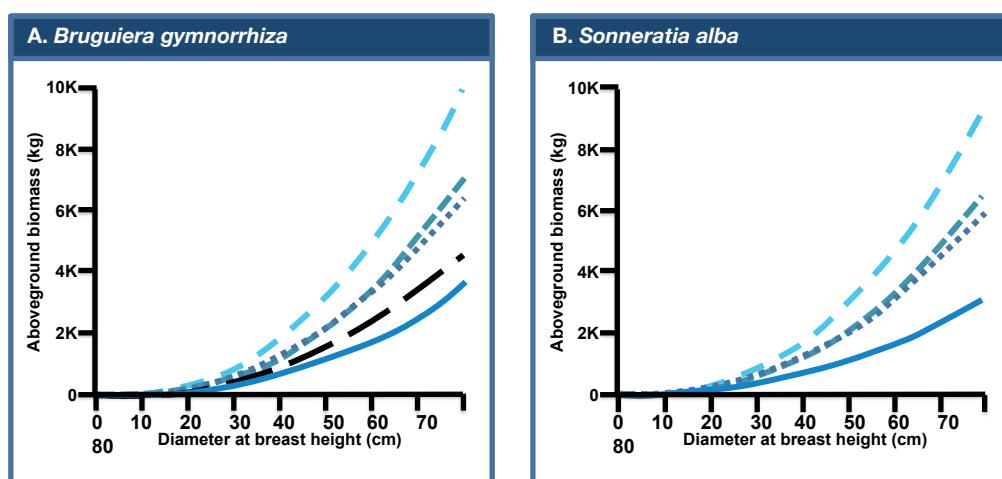
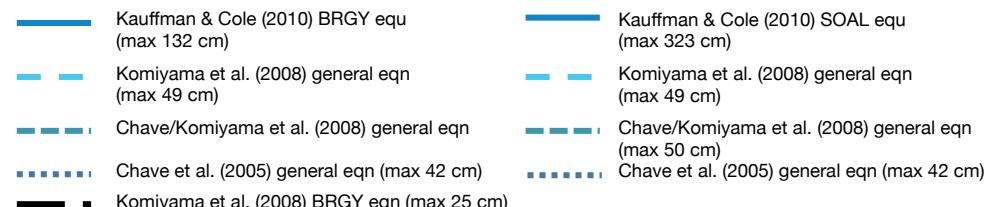


Figure 4.5 Comparison of tree biomass estimates for (A) *Bruguiera gymnorhiza* and (B) *Sonneratia alba*. The numbers in parentheses are the maximum tree diameters used to develop the equations (Chave et al. 2005; Komiyama et al. 2008; Kauffman & Cole 2010).

was 2,588 kg using the Kauffman and Cole (2010) equation and 7,014 kg using the general equation of Komiyama *et al.* (2008). Similarly, the biomass estimate for a 45 cm dbh *Sonneratia alba* tree was 873 kg using the Kauffman and Cole (2010) formula but > 1,500 kg using the other equations. For the largest trees in this stand the differences were even more dramatic. The biomass estimate for an 80 cm dbh tree was 3,034 kg using the Kauffman and Cole (2010) equation but over threefold higher (9,434 kg) using the Komiyama *et al.* (2008) general mangrove equation. It is important to note that only the equations developed by Kauffman and Cole (2010) encompassed the entire range of diameters encountered at the Yap site, and the allometric equations used only depended on diameter (dbh) and wood density for computing biomass of mangrove trees. These large differences underscore the importance of using the same equations for all trees in a project area, when comparing different mangroves, and especially for permanent mangrove plots through time.

Determining the Carbon within the Live Tree Component (kg C/m²): The live tree carbon component is determined by multiplying the biomass (kg) of each tree—as determined by an allometric equation or laboratory analysis—by the carbon conversion factor for mangrove species specific to that region. This is done for every tree sampled. Next, all the values for individual tree carbon content are added together to determine the total carbon content from the trees (kg C) for the given plot size (m²). Carbon conversion factors are based on the percent of the biomass that is made of organic carbon. This can be determined through laboratory analysis using an elemental analyzer (Chapter 3), but in many cases it is possible to find a published carbon conversion factor for specific tree species. Kauffman *et al.* (2011) reported the carbon content of *Bruguiera gymnorhiza* as 46.3%, *Rhizophora apiculata* as 45.9%, and *Sonneratia alba* as 47.1%. Thus, if local or species-specific values are not available the biomass of these and other trees can be multiplied by 0.46 to 0.5 to obtain carbon content.

EXAMPLE

- Carbon content of each tree (kg C) = tree biomass (kg) * carbon conversion factor (0.46–0.5)
- Carbon in live tree component (kg C/m²) = (carbon content of tree #1 + carbon content of tree #2 + Tree #n) / area of the plot (m²)

SCRUB MANGROVES

A great percentage of the world's mangroves have an aboveground structure of small trees less than a few meters in height, often referred to as dwarf mangrove, scrub mangrove, or “manglar chaparro.” Currently there are few allometric equations to determine aboveground biomass for these kinds of mangroves; thus, it is an important research need. The few existing equations that are most often used are from measurements of dwarf mangroves in south Florida, USA (Ross *et al.* 2001; Coronado-Molina *et al.* 2004). The equations in Ross *et al.* (2001) utilize the main stem diameter at 30 cm above the ground surface and crown volume to predict aboveground biomass of individual mangroves. Coronado-Molina *et al.* (2004) developed allometric equations utilizing crown area and the number of prop roots. The most accurate method, however, is to develop allometric equations specifically for the plants in your area of interest.

To develop an allometric equation for a new site, at least 15 to 25 trees of each species in question, encompassing the range in size from the smallest seedlings to the largest individuals, should be measured (typically crown diameter, crown volume, crown area, tree height, and/or main stem diameter at 30 cm aboveground, **Fig. 4.6**) and harvested. In the

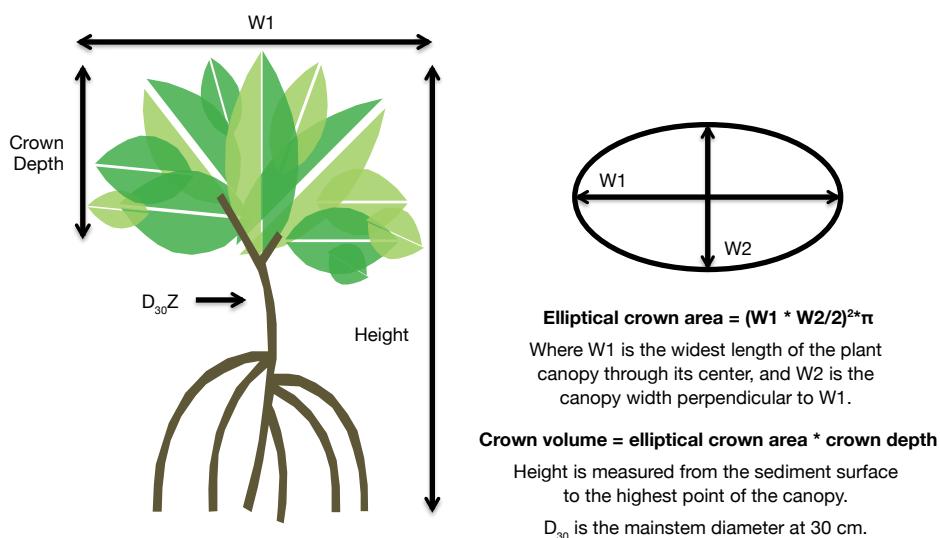


Figure 4.6 Field measurement techniques to calculate the elliptical crown area, crown depth, height, and diameter at 30 cm height (D_{30}) of dwarf mangroves. Aboveground biomass of these small trees is best predicted through allometric equations where aboveground biomass is the dependent variable and crown area, and height and/or crown volume are independent variables (Ross *et al.* 2001).

laboratory, individual trees are dried and weighed to obtain biomass. A relationship between the tree biomass and the biometric measurements (crown diameter, area and volume, main stem diameter at 30 cm) can then be developed by regression analysis.

Once allometric equations have been established, they can be applied to every scrub mangrove tree within a sample plot. The plot size is typically small; however, since scrub mangroves grow in very dense communities, field measurements can be time consuming (**Fig. 4.2A**). For example, a typical plot size and shape would be a 2 m radius half circle plot (total area 6.3 m²).

Determining the Carbon within the Scrub Mangrove Component (kgC/m²): The scrub mangrove carbon component is determined for each tree by multiplying the biomass (kg), as determined by an allometric equation, by a carbon conversion factor for scrub mangroves specific to that region. This is done for every scrub tree in the plot. Next, all the values for individual tree carbon content are added together to determine the total carbon content from the scrub trees (kg C) for the given plot size (m²).

The carbon conversion factor for scrub mangroves is not well documented in the scientific literature; therefore, it can be determined either through laboratory analysis using an elemental analyzer (Chapter 3), or it is justifiable to use the conversion factor reported for tall mangrove trees (0.46 to 0.5, see section on mangrove trees above).

EXAMPLE

- Carbon content of each tree (kg C) = tree biomass (kg) * carbon conversion factor (0.46–0.5)
- Carbon in live tree component (kg C/m²) = (carbon content of tree #1 + carbon content of tree #2 + Tree #n) / area of the plot (m²)

STANDING DEAD TREES

Within each sampling plot, all trees that are dead and standing should be recorded as such and analyzed as a separate pool. The degree to which the tree has decayed will determine how its biomass is calculated. Decay status is broken down as follows (Fig. 4.7):



Figure 4.7 Examples of dead tree decay status. 1) Recently dead trees that maintain many smaller branches and twigs, 2) Trees have lost small branches and twigs; and a portion of large branches, 3) Most branches have been lost and only the main stem remains, and is often broken (Solochin 2009).

- **Decay status 1:** Small branches and twigs are retained; resembles a live tree except for absence of leaves.
- **Decay status 2:** No twigs/small branches; may have lost a portion of large branches.
- **Decay status 3:** Few or no branches, standing stem only; main stem may be broken-topped.

The biomass of standing dead trees is then determined according to the decay status of the tree.

Decay status 1: Biomass can be estimated using live tree equations. The only difference is that leaves should be subtracted from the biomass estimate. This can be accomplished either using a leaf biomass equation to determine the quantity of leaves to be subtracted (Clough & Scott 1989; Komiyama *et al.* 2005; Smith III & Whelan 2006), or by subtracting a constant of 2.5% of the live tree biomass estimate.

Decay status 2: Biomass can be calculated by subtracting away a portion of the biomass from the live tree equations. Because they have lost some branches in addition to leaves, both leaf biomass and an estimate of branch loss must be factored in. Commonly, a total of 10–20% of biomass (accounting for both leaves and some branches) is subtracted. This amount can be adjusted and tailored to specific conditions using field observations.

Decay Status 3: Trees have often lost a significant portion of their volume due to advanced breakage; consequently, it is difficult to estimate biomass from the live-tree biomass estimates. Instead, the remaining tree's volume may be calculated using an equation for

a frustum (truncated cone). To do this, record the diameter at the base of the tree and the total tree height using a laser tool or clinometer (a tool used to measure mangrove tree height). The top diameter must be estimated using a taper equation.

Taper equation for estimating the top-diameter of a broken-topped dead tree:

- Estimating the top-diameter of a broken-topped dead tree (cm) = the measured basal diameter (cm) – [100 * tree height (m) * ((the measured basal diameter (cm) – diameter at breast height (cm) / 130)]
 - $d_{top} = d_{base} - [100 * ht * ((d_{base} - dbh) / 130)]$
 - If taper equation results in negative number, use 0 for the next equation.

Then the volume of the dead tree is determined by assuming the tree is a truncated cone:

- Dead tree volume (cm^3) = $[\pi * (100 \times \text{tree height (m)}) / 12] * [\text{base diameter (cm)}^2 + \text{top diameter (cm)}^2 + (\text{base diameter (cm)} \times \text{top diameter (cm)})]$
 - Volume (cm^3) = $(\pi * (100 * ht) / 12) * (d_{base}^2 + d_{top}^2 + (d_{base} * d_{top}))$.

Dead tree biomass (kg) is then determined by multiplying its volume (cm^3) by its wood density (g/cm^3).

- Decay Status 3 dead tree biomass (kg) = Volume of the dead tree (cm^3) * wood density (g/cm^3) * (1 kg / 1000 g).

Ideally wood density of standing dead trees would be determined in the laboratory; however, if that is not practical, a list of standard densities based on size (**Table 4.4**) can be used for this calculation. It is important to note that the density information in **Table 4.4** was derived from downed wood measured following a typhoon with little or no decay. Studies have shown that there is a broad range of densities for various components (twigs 0.628–0.350, branches 0.60–0.284, prop roots 0.276–0.511, and trunks 0.340–0.234), emphasizing the need for site specific estimations of wood density when possible (Robertson & Daniel 1989).

Table 4.4 The wood density and mean diameter of the standard wood debris size classes of downed mangrove wood (*Rhizophora apiculata*, *Sonneratia alba*, and *Bruguiera gymnorhiza*) (Kauffman & Cole 2010).

SIZE CLASS (cm DIAMETER)	DENSITY ±SE (g/cm ³)	SAMPLE SIZE (n)
< 0.64	0.48 ± 0.01	117
0.65 – 2.4	0.64 ± 0.02	31
2.54 – 7.6	0.71 ± 0.01	69
> 7.6	0.69 ± 0.02	61

Determining the Carbon within the Standing Dead Tree Component (kg C/m²): The standing dead tree carbon pool is determined by multiplying the biomass (kg), determined by decay status, of each individual standing dead tree by a carbon conversion factor. Next all the values for individual tree carbon content are added together to determine the total carbon content from the trees (kg C) for the given plot size (m²).

The carbon conversion factor for standing dead trees can be determined through laboratory analysis using an elemental analyzer (Chapter 3). If this is not practical, the carbon concentration of dead wood is usually around 50% (Kauffman *et al.* 1995). Thus, the biomass of these trees is typically multiplied by 0.5 to obtain carbon stock value.

EXAMPLE

- Carbon content of each tree (kg C) = tree biomass (kg) * carbon conversion factor (0.46–0.5)
- Carbon in standing dead tree component (kg C/m²) = (carbon content of tree #1 + carbon content of tree #2 + Tree #n) / area of the plot (m²)

LIANAS

Lianas are long-stemmed, woody vines that are rooted in the soil of the forest floor and climb up into the forest canopy on trees (**Fig 4.8**). They range from small, indiscrete vines that wind around tree trunks to giant lianas several cm in diameter that seemingly hang in the middle of the forest independent of trees. Large lianas rarely exist in mangroves, but if present they can be measured just like trees.



Figure 4.8 Lianas. (A) Close up view of a typical liana found in mangrove forests (© IITA), (B) image of the extent to which lianas can contribute to the vegetative biomass (© Mark Marathon, Wikimedia Commons)

Every liana in a sample area should be measured if they are being included in calculations. There are a number of equations to determine liana biomass (Schnitzer *et al.* 2006), but for our purposes liana biomass can be estimated using the general allometric equation:

- Biomass for lianas (kg) = (Diameter 130 cm from the soil surface (cm))^{2.657} * e^{0.968} x ln (Diameter 130 cm from the soil surface (cm))
 - $B = D^{2.657} \cdot e^{-0.968} \ln(D)$

Determining the Carbon within the Liana Component (kg C/m²): When lianas are present in mangrove forests, their carbon pool is determined by multiplying the biomass (kg) of each individual vine by a carbon conversion factor. Next, all the values for individual liana carbon content are added together to determine the total carbon content from the vines (kg C) for the given plot size (m²).

The carbon conversion factor for lianas can be determined through laboratory analysis using an elemental analyzer (Chapter 3). If this is not practical, the carbon concentration (based on dry weight) of lianas in the forest of Mexico have been reported to be ~46% (Jaramillo *et al.* 2003); therefore, a default value for the carbon conversion factor of lianas is 0.46.

EXAMPLE

- Carbon content of each liana (kg C) = liana biomass (kg) * carbon conversion factor (0.46)
- Carbon in the liana component (kg C/m²) = (carbon content of liana #1 + carbon content of liana #2 + liana #n) / area of the plot (m²)

PALMS AND OTHER NON-TREE VEGETATION

Understory vegetation (e.g., non-mangrove tree seedlings and herbs) is generally negligible in mangroves, and its measurement for ecosystem carbon pools is usually unnecessary (Snedaker & Lahmann 1988).

The most notable exception is the nypa palm (*Nypa fruticans*, **Fig. 4.9**) of the Asia-Pacific region where it may be the dominant species in some locations. Approaches to sampling these vegetation components will depend on their density, structure, and distribution (Snedaker & Lahmann 1988). The biomass of nypa palms and herbaceous vegetation may be determined either through non-destructive sampling or destructive harvests (which is most common for herbaceous vegetation, such as ferns, seagrass, grasses, sedges, rushes, and broad-leaved herbs). Non-destructive approaches are necessary for plots that will be revisited over time, especially for perennial species.



Figure 4.9 Examples of palm plants found in mangroves. (A) Small *Nypa fruticans* (© Andreas Hutahean, KKP), (B) tall woody stemmed palm plants (© Enrico Marone, CI)

To determine biomass (kg), at least 15 to 25 palm fronds from different individual plants should be collected from outside any permanent sample plots. The samples should cover the observed size distribution of the individual leaves and each sampled frond is cut at ground level. Obtain the dry mass of each frond in the laboratory and calculate the average. Count the total number of nypa leaves within a sample (or subsample) plot and then multiplying that number by the average dry mass.

Palms with woody trunks can be measured in the same manner as broad-leaved trees. Parameters of the palm that need to be measured (dbh, height, etc.) depend on the allometric equation used to determine palm biomass. The most frequently used variable to determine palm biomass is the height of the main stem from the ground to the base of leaves.

Determining the Carbon within the Palm Component (kg C/m²): The palm carbon component is determined by multiplying the biomass (kg), either the average calculated from the collected fronds or from allometric equations for larger woody palms, by a carbon conversion factor for palm species specific to that region.

If the biomass used was determined for the entire plot using the average biomass of the fronds collected multiplied by all the fronds in the plot, then simply multiply that number by the carbon conversion factor to determine the total palm carbon component for your plot.

If the biomass used was determined for larger woody palms, all the values for individual palm carbon content must be added together to determine the total carbon content from the trees (kg C) for the given plot size (m²).

The carbon conversion factor for palms can be determined either through laboratory analysis using an elemental analyzer (Chapter 3) or a conversion factor of 0.47 can be used (Kauffman *et al.* 1998).

EXAMPLE FOR SMALL PALMS

- Carbon in the small palm component (kg C/m²) = (Estimated biomass of the palm fronds * carbon conversion factor (0.47)) / area of the plot (m²).

EXAMPLE FOR LARGE WOODY PALMS

- Carbon content of each large palm (kg C) = palm biomass (kg) * carbon conversion factor (0.47);
- Carbon in large palm component (kg C/m²) = (carbon content of tree #1 + carbon content of tree #2 + Tree #n) / area of the plot (m²).

PNEUMATOPHORES

Pneumatophores of mangrove species of the genera *Avicennia*, *Bruguiera*, and *Sonneratia* can be of significant structure and biomass, and unlike the stilt roots on *Rhizophora*, these tree parts are not included in the allometric equations of biomass for live trees (**Fig. 4.10**). Pneumatophore density can be determined by counting their numbers in microplots within or immediately adjacent to, the main sampling plot.

A common-sized microplot is 50 x 50 cm² but can range from 30 x 30 cm² to 1 x 1 m². To determine biomass (kg), pneumatophores should be collected from outside any permanent sample plots. All pneumatophores within the plot should be counted and 50–100 samples should be collected. The samples should cover the observed size distribution and be cut at ground level. Obtain the dry mass of each pneumatophore in the laboratory and calculate the average.



Figure 4.10 Pneumatophores. (A) Measuring pneumatophore height, (B) Pneumatophores can be measured in or next to microplots. These microplots can be the same plots used to sample litter (described below).
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- Biomass for pneumatophores (kg) = Average dry mass of sampled pneumatophores * number of pneumatophores in the microplot.

An allometric equation using pneumatophore height to predict biomass could be developed and would be of value in permanent plots. If such an equation were developed, each pneumatophore in a microplot would need to be measured for height. This would be most relevant for those species containing large pneumatophores such as *Sonneratia alba*.

Determining the Carbon within the Pneumatophore Component (kg C/m²): The pneumatophore carbon component is determined by multiplying the average biomass (kg)—calculated from the collected pneumatophores—by a carbon conversion factor for species specific to that region.

Since the biomass used was determined for the entire microplot using the average biomass of the pneumatophores collected multiplied by all the pneumatophores in the plot, you simply need to multiply that number by the carbon conversion factor to determine the total pneumatophore carbon component for a microplot.

The carbon conversion factor for pneumatophores can either be determined through laboratory analysis using an elemental analyzer (Chapter 3), or a conversion factor for roots can be used—typically 0.39 (Kauffman & Donato 2011).

EXAMPLE

- Carbon in the pneumatophore component (kg C/m²) = (Estimated biomass of the pneumatophores * carbon conversion factor (0.39)) / area of the plot (m²).

LITTER

The litter layer is defined as the recently fallen non-woody dead organic material on the soil surface. Typically, it consists of dead leaves, flowers, fruits, seeds, and bark fragments. In most mangrove settings, the amount of this material (and therefore carbon stock) only represents a very small component of the carbon pool due to the high efficiency of detritus-consuming crabs as well as export through tides and seasonal river flooding.

If it is measured, litter in most biomass studies is destructively sampled through collection from small microplots, often 0.5×0.5 m in size. All organic surface material, excluding woody particles, is collected into a sturdy bag or container. The bags are labelled with

the location, date, plot, and sample number (**Fig. 4.11**). Given the wet nature of mangroves, pre-labelled plastic bags with permanent markers may be most efficient for litter samples.



Figure 4.11 Pre-labelled plastic bags containing litter (© Boone Kauffman, OSU)

Determining the Carbon within the Leaf Litter Component (kg C/m²): The samples should be transported to the laboratory, placed in a drying oven, and dried to constant mass. Due to constraints of carrying bulky samples from the field and the limited availability of oven drying space, especially in rural field settings, we suggest determining the wet weight of the entire sample and then extract a well-mixed representative subsample for transportation to a laboratory to be dried to a constant weight. Finally, determine the ratio between wet and dry mass of the subsample by recording the wet mass of the sample and relating it to the dry (constant) mass (Cummings *et al.* 2002).

- Biomass of litter (kg) = (dry mass of subsample (g) / wet mass of the subsample (g)) * wet mass of all the litter in the sample plot (kg)

The litter biomass (kg) can then be estimated for the given plot size (m²). Mean carbon concentrations of tropical forest leaf litter dry mass have been reported to range from 38–49% (Kauffman *et al.* 1993; Kauffman *et al.* 1995). Therefore, a carbon conversion factor of about 0.45 is recommended.

EXAMPLE

- Carbon in the leaf litter component (kg C/m²) = (Average biomass of the litter * carbon conversion factor (0.45)) / area of the plot (m²)



Figure 4.12 Downed wood. (A) Downed wood after a storm in Bangladesh, (B) Uprooted *Rhizophora* and *Sonneratia* following Typhoon Sudal, Yap, FSM (© Boone Kauffman, OSU)

DEAD AND DOWNED WOOD

There are several guides that describe methods for determining downed wood volume and mass (Harmon & Sexton 1996; Waddell 2002). Dead and downed wood material can be a significant component of aboveground biomass, particularly following natural disturbances, such as tropical cyclones (**Fig. 4.12**). Land-use and/or land-cover change may also increase the quantity of downed wood on the mangrove forest floor. To accurately assess ecosystem carbon pools and influences of natural and human disturbances, dead and downed wood is an important variable to measure. Downed wood is usually sampled either by plot-based sampling or by the line-intersect method (Waddell 2002; Baker & Chao 2009). The non-destructive line-intersect technique is recommended.

Line (or planar) intersect technique for sampling downed wood: The line (or planar) intersect technique involves counting intersections of woody pieces along a vertical sampling plane (transect). In each sampling plot, a series of transects should be established to measure this carbon pool. A transect is a straight line across the entire length of the plot. For example, four transects might be established in each of six subplots for a total of 24 transects per plot.

Important rules in measuring downed wood include:

- Dead trees that are standing are not measured in the line intersect technique;
- Downed wood must be broken from the tree where it originated; thus, dead branches and stems still attached to standing trees or shrubs do not count;
- The transect tape must intersect the central axis of a wood piece for it to be counted. This means that if the tape only intersects a corner at the end of a log, it does not count; and
- Any piece can be recorded multiple times if the tape intersects it more than once (e.g., a curved piece, or at both the branch and the stem of a fallen tree).

All downed, dead, woody material (fallen/detached trunks, branches, prop roots, or stems of trees and shrubs) that has fallen and lies on or over the transect line (within 2 m of the ground surface) is counted and classified using this technique. Woody debris can be categorized into four sizes: fine, small, medium, and large wood particles (**Table 4.5**). These size classes are regularly used in forest inventories, and convenient measurement tools exist to streamline field sampling based on these limits (**Fig. 4.13**).

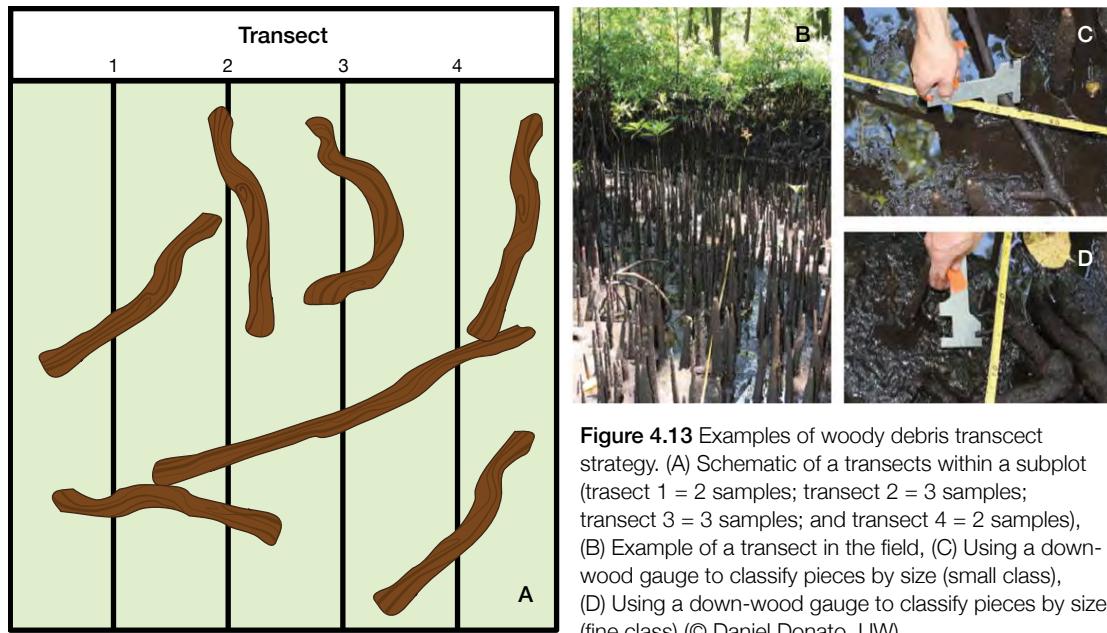


Figure 4.13 Examples of woody debris transect strategy. (A) Schematic of a transects within a subplot (transect 1 = 2 samples; transect 2 = 3 samples; transect 3 = 3 samples; and transect 4 = 2 samples), (B) Example of a transect in the field, (C) Using a down-wood gauge to classify pieces by size (small class), (D) Using a down-wood gauge to classify pieces by size (fine class) (© Daniel Donato, UW)

Table 4.5 Commonly used size classes of wood.

DESCRIPTION	DIAMETER
Fine	0 – 0.6 cm
Small	0.6 – 2.5 cm
Medium	2.5 – 7.6 cm
Large	≥ 7.6 cm

The number of fine, small, and medium pieces that cross the transect line are counted and tallied separately for each size class. Smaller pieces can be very abundant, and to save time, are only sampled along subsections of each transect (**Fig. 4.14**).

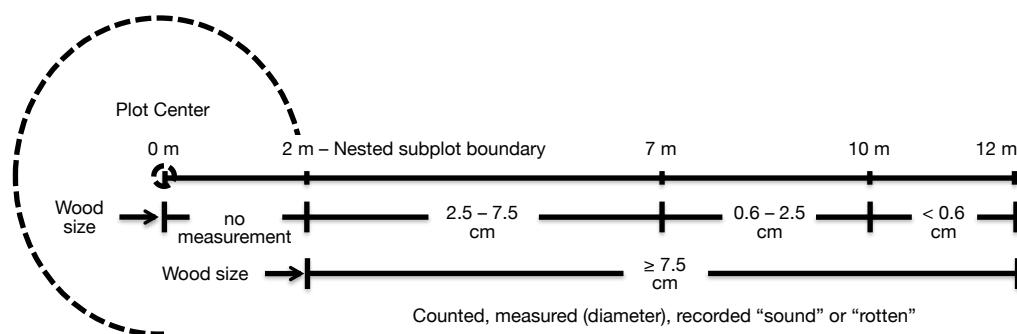


Figure 4.14 Example of a wood debris transect for sampling downed wood in mangroves using the line intersect technique. The diameter of each large wood piece encountered along the entire transect is measured. Smaller diameter pieces are counted but not measured along the designated lengths of the transect.

To convert the count for the smaller class sizes into biomass, an average diameter of wood pieces in each of these classes must be estimated. The average diameter of wood pieces can be derived from measurements of about 50–100 randomly selected pieces of each

size class (fine, small, and medium). A digital calliper is recommended for measuring the diameter of each piece. We recommend using a quadratic mean equation to calculate the average diameter of the wood pieces rather than the mean diameter of wood classes (Brown & Roussopoulos 1974). The quadratic mean diameter (QMD) is calculated as:

- Quadratic mean diameter (cm) = $\sqrt{(\sum \text{diameter of each piece of wood}^2) / \text{number of pieces sampled}}$
- $\text{QMD} = \sqrt{(\sum D_i^2) / n}$

Examples of quadratic mean diameter of down wood from mangrove forests are shown in **Table 4.6**.

In contrast to the smaller wood pieces, the diameter of each large wood piece encountered along the entire transect is recorded (**Fig. 4.14**). For each large piece crossing the transect line, record its diameter at the point where the transect line crosses the midpoint of the wood particle. Also, record whether the decay status is sound (machete bounces off or only sinks slightly when struck) or rotten (machete sinks deeply and wood is crumbly with significant loss).

In a practical sense, usually only the medium and large wood fraction (> 2.5 cm diameter) is a significant carbon pool (Kauffman & Cole 2010; Kauffman et al. 2011). If sampling the litter component, it may be simpler to include the smaller wood pieces (< 2.5 cm diameter) in the litter sample and only measure the medium and large wood pieces along the transect line.

Wood density: The density of wood debris is determined from the dry weight of the wood dried at 100 °C divided by the volume of undried wood. Density must be determined for each wood category sampled (fine, small, medium, large-sound, large-rotten) in order to determine biomass. We recommend collecting at least 20–25 pieces of wood for each species and size class, capturing a representative range of sizes within each class and the full range of species present in the sample. As a rough guideline, each piece collected for wood density determination should have a mass of between ~ 0.5 and 50 g. Randomly collect pieces to determine wood density within the project area; to avoid undue disturbance, do not collect pieces from permanent sample plots. See beginning of this chapter for a discussion on wood density calculations. Examples of wood density for downed wood from mangrove forests are shown in the table below.

Table 4.6 The wood density and mean diameter of the standard wood size classes of downed mangrove forests dominated by *Rhizophora apiculata*, *Sonneratia alba*, and *Bruguiera gymnorhiza*. Data are from downed wood following a typhoon with no decay. na = not applicable (Modified from Kauffman and Cole, 2010).

SIZE CLASS (cm diam.)	DENSITY ±SE (g/cm³)	SAMPLE SIZE	DIAMETER (cm)	QUADRATIC MEAN DIAMETER (cm)	SAMPLE SIZE
< 0.64	0.48 ± 0.01	117	0.43 ± 0.15	0.43	50
0.65 – 2.4	0.64 ± 0.02	31	1.33 ± 0.78	1.47	48
2.54 – 7.6	0.71 ± 0.01	69	4.30 ± 0.18	4.52	52
> 7.6	0.69 ± 0.02	61	na	na	na

Downed wood volume is then calculated from line intercept data using scaling equations.

- Wood volume for fine, small, and medium classes per unit of ground area (m^3/ha) = $(\pi^2 \times [\text{number of samples} \times \text{quadratic mean diameter for the size class (cm)}]^2 / (8 \times \text{transect length (m)})]$
 - Volume (m^3/ha) = $\pi^2 * (\text{Ni} * \text{QMD}_i^2 / (8 * L))$
- Wood volume of large (> 7.6 cm diameter) down wood per unit of ground area (m^3/ha) = $\pi^2 * [\sum \text{diameter of each piece of wood}^2 / (8 \times \text{transect length (m)})]$
 - Volume (m^3/ha) = $\pi^2 * (d_1^2 + d_2^2 + d_3^2 + \dots + d_n^2 / (8 * L))$
 - Each piece is individually measured

Downed wood biomass (kg/ha) is then calculated as the volume multiplied by its average wood density.

- Downed wood biomass (kg/ha) = volume (m^3/ha) * average wood density (kg/m^3)

Determining the Carbon within the Downed Wood Component ($\text{kg C}/\text{ha}$): Finally, convert downed wood biomass to carbon mass. An acceptable default value based upon carbon content of dead wood in tropical forests is 50%, corresponding to a carbon conversion factor of 0.50.

EXAMPLE

- Carbon content of downed dead wood per plot (kgC/ha) = dead wood biomass (kg/ha) * carbon conversion factor (0.5)

BELOWGROUND TREE BIOMASS

Belowground biomass (e.g., roots and the base of stems) is an important component in mangroves. The aboveground-to-belowground biomass ratio for mangroves is generally found to be between 2.0 and 3.0, whereas in upland forests the ratio is higher—between 3.96 and 4.52. Thus, in mangrove forests, a large amount of biomass tends to be allocated below ground (Komiyama *et al.* 2008). Given the extreme difficulty of collecting and measuring root systems, it is beyond the capacity of most mangrove projects to destructively harvest and measure belowground biomass or develop allometric equations. Very few allometric equations exist for belowground biomass of forests, and mangroves are among the least studied forests in this respect. Additional studies would be a valuable contribution to science. Useful belowground equations available for mangroves have been reviewed in Komiyama *et al.* (2008). The general equation reported by Komiyama *et al.* (2008) is:

- Belowground tree biomass (kgC) = $0.199 * ((\text{wood density (g/cm}^3)^{0.899}) * (\text{tree diameter at breast height (cm)})^{2.22}$

Carbon content of roots are typically lower than that of aboveground tree components. Jaramillo *et al.* (2003) reported that carbon content of roots of tropical forests range from 36–42%. A defensible default value for root carbon content would be 39% (median); making the carbon conversion factor 0.39.

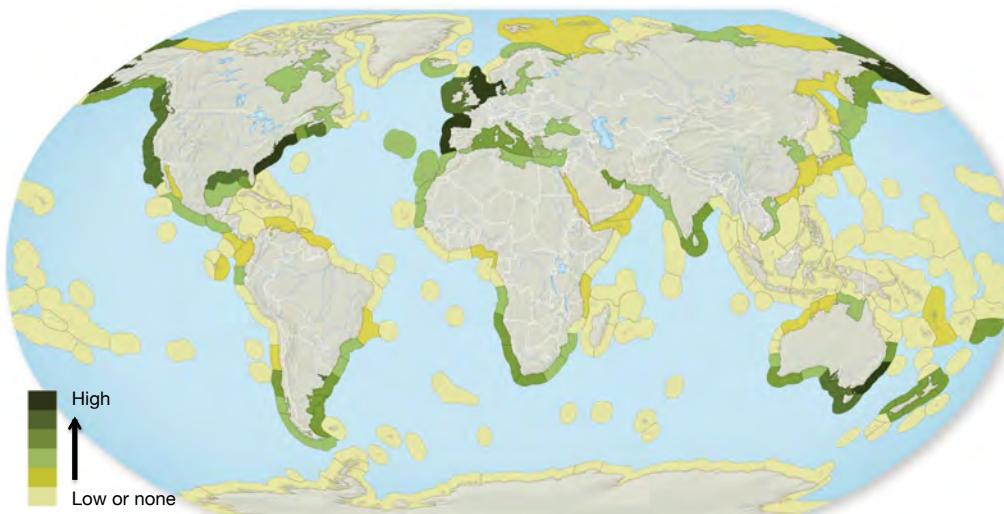
EXAMPLE

- Belowground carbon content of each tree (kg C) = Belowground tree biomass (kg C) * carbon conversion factor (0.39)
- Carbon content of belowground carbon per plot (kg C/m²) = (belowground carbon content of tree #1 + carbon content of tree #2 + Tree #n) / plot area (m²)

TIDAL SALT MARSHES

For the purposes of this manual, tidal salt marshes are defined as coastal ecosystems in the upper coastal intertidal zone between land and open salt water that is regularly flooded and covered with vegetation (excluding trees). Tidal salt marshes exclude mangroves, along with tidal freshwater/brackish forests (e.g., Swamp oak, Paperbark, Cypress swamps, and willows), and un-vegetated salt flats. The halophytic plants that inhabit coastal tidal salt marsh are not exclusively intertidal or marine—a characteristic that sets them apart from mangrove and seagrass vegetation—and are dominated by herbaceous plants including tidal grasses, sedges, and rushes. Tidal saltbushes and shrubs may also be present. The variety of growth forms is important when calculating carbon stocks because each has a different biomass.

Tidal salt marshes establish on soft substrate shores within the tidal reaches of estuaries and embayments and on some open low-wave energy coasts (**Fig 4.15**). At a global scale, tidal salt marshes establish on shorelines unsuitable for mangrove forests or where development is limited (Kangas & Lugo 1990). For this reason, these systems are most common in temperate, subarctic and arctic zones (Long & Mason 1983; Mitsch & Gosselink 2007). In tropical regions, tidal salt marshes may be replaced in the intertidal environment, either by mangroves in humid climates or saltfans in arid climates.



TNC: Conservation Maps and GIS Data

Figure 4.15 Tidal salt marsh abundance by marine ecoregion (Hoekstra *et al.* 2010)

The distribution of tidal salt marsh plants varies across the marsh and with marsh elevation, due to differences in tolerance to soil salinity, oxygen levels, sulphide levels, hydroperiod, etc. (Partridge & Wilson 1987). Along the Atlantic coast of North America, tidal salt marshes are

often divided into different zones, based on variations in plant species, and relative elevation of the ground surface with respect to the tide (Niering & Warren 1980) (**Fig. 4.16**). The low marsh area is located at the lowest elevation and is more frequently inundated by tides. The middle/high marsh is flooded less frequently and the higher high marsh is even less frequently inundated by the tide. Brackish marshes are typically located along the inland edges of the marsh area where freshwater input creates a brackish environment.

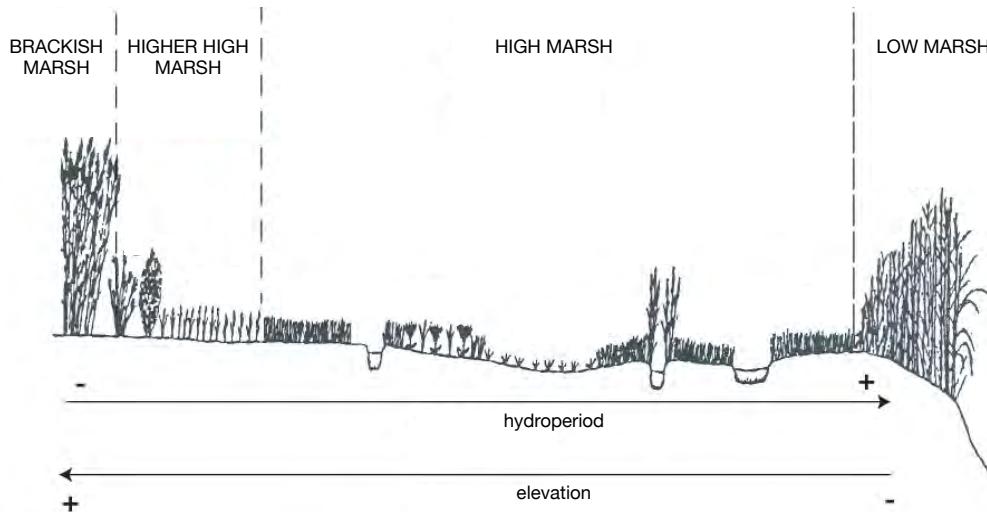


Figure 4.16 Zonation of tidal salt marsh vegetation in herb dominated systems, NE North America (modified from Niering & Warren 1980)

The different zones of the marsh may have floristic and other structural differences that influence above- and below-ground biomass of living plants. Additionally, geochemical properties, including nutrient availability and redox conditions may influence carbon retention in the soil.

Tidal salt marshes provide a host of ecosystem services, including organic matter production and export, nutrient cycling, buffering against storm activity, and carbon sequestration (Nixon 1980; Dame *et al.* 1991; Mitsch & Gosselink 2000; Chmura *et al.* 2003). Globally, tidal salt marshes have suffered from a long history of conversion into non-tidal land (termed “reclamation”), usually through the imposition of embankments or dikes, which prevent flooding of the ecosystems by tides. Other direct, human-induced causes of degradation of coastal tidal salt marshes include their conversion to salt evaporation ponds, aquaculture ponds (i.e., shrimp ponds) (Barg *et al.* 1997), and the alteration of tidal salt marsh drainage for insect control (Daiber 1982). Drainage control has also been implemented to protect low-lying land, leading to changes in the plant composition of the marsh ecosystem.

Although no global assessment of tidal salt marsh decline has been conducted, regional case studies suggest that tidal salt marsh extent has declined significantly over the past century. Wetland loss on the U.S. Mississippi Delta has been estimated to be in the vicinity of 100 km²/yr (Viles & Spencer 1995) due to the diversion of water and sediment in the Mississippi catchment and delta (Gosselink & Maltby 1990). Overall loss of North American tidal salt marshes is estimated to be 50% since European settlement (Gedan *et al.* 2009). In southeast Australia, tidal salt marsh area has declined by 30–40% since 1950, primarily due to the replacement of tidal salt marshes by mangroves (Saintilan & Williams 1999), and 50% of China’s tidal salt marshes were lost due to reclamation since the 1950’s (An *et al.* 2007).

These losses have important implications for carbon retention in the system (DeLaune & White 2012).

Tidal salt marshes sequester and store carbon efficiently in the plant biomass and soils. Belowground carbon pools usually constitute between 65 and 95% of the total ecosystem carbon stock of tidal salt marshes (Elsey-Quirk *et al.* 2011), dominating the carbon sequestration, particularly in low and middle marsh areas. Aboveground biomass becomes a more significant component of the total carbon pool in the high marsh. Thus, it is important to be able to differentiate high, middle, and low marsh when creating a sampling strategy.

Field Sampling Considerations

Tidal salt marsh vegetation will vary depending on the season as well as soil moisture, nutrient, and salinity conditions that correspond to variations in tidal flooding. Proper sampling techniques take these factors into account by subdividing areas of interest into strata of (relatively) homogeneous ecological characteristics, and sampling takes place when the biomass is at its peak growth (usually late summer). Sampling may also be limited to low tidal periods of as little as 3 to 4 hours. This narrow window necessitates an efficient sampling plan.

The strata of (relatively) homogeneous ecological characteristics within tidal salt marshes usually occur parallel to the shoreline or the tidal channel. Due to the innate structure of tidal salt marshes, strata will usually form perpendicular to the dominant tidal channel/shoreline. Sufficient sampling plots should be placed in each stratum to characterize biomass within that stratum. Plots within each stratum should be approximately 20 m x 50 m. Within these plots, a minimum of 5–6 smaller subplots (0.25 m x 0.25 m) are randomly located (**Fig. 4.17** and Chapter 2 on project planning and sampling design for more details).

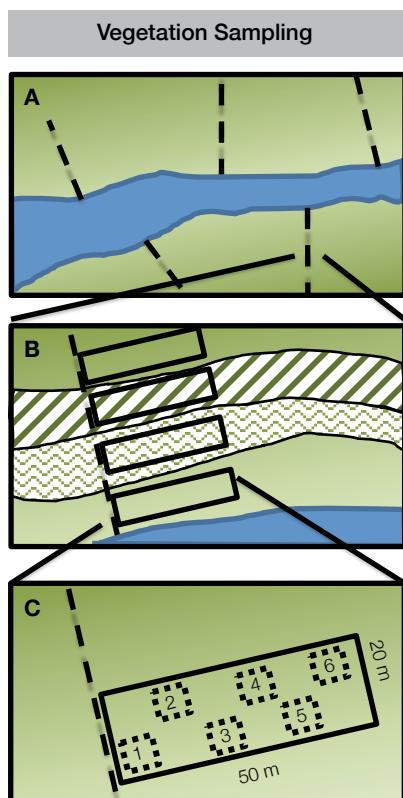


Figure 4.17 Recommended Placement of sampling plots in tidal salt marshes. (A) Transects should occur perpendicular to the dominant channels in the marsh and that cross across vegetation or other structural gradients, (B) Plots along the transects should be contained within strata, and (C) enough quadrats (minimum five) should be sampled in each plot to characterize the biomass.

Tidal salt marsh aboveground biomass can vary substantially between seasons, particularly in cool, temperate, and subarctic climates where the entire aboveground biomass of the tidal salt marsh might be lost during the winter (Darby & Turner 2008a). Other factors include elevated nutrient levels that can decrease the proportion of biomass allocated to the roots (Darby & Turner 2008b), salinity level that can increase root:stem ratio (Parrondo *et al.* 1978), grazing pressure (Giroux & Bédard 1987), and water table position (Saunders *et al.* 2006). We recommend that aboveground and belowground estimates of tidal salt marsh biomass be taken when biomass is likely to be at its greatest—usually mid-late summer. For permanent plots and/or repeated measurements, it is important sample during the same season each time so that results will be comparable.

Biomass Estimates

This section provides guidance on how to measure aboveground biomass across a range of tidal salt marsh types likely to be encountered in the field. Some differences in sampling procedures are required to accommodate differences in growth forms.

GRASSES, SEDGES AND OTHER HERBACEOUS PLANTS

Reeds, sedges and rushes are densely growing plants, with tall, straight stems and are the primary vegetation type in tidal salt marshes. The method commonly used to estimate their aboveground biomass uses a quadrat to define sampling areas within plots. Quadrats for this ecosystem are typically 30 cm x 30 cm, though a larger quadrat should be used if less than 10 individuals are likely to be counted in the 30 cm x 30 cm area. Within each quadrat, count the number of stems per species (tidal salt marshes generally contain large stands of a single dominant species but not always) and measure the total height for each species present within the quadrat. If a stem shows signs of senescence (dead material) at the tip, measure the green height (**Fig 4.18**).

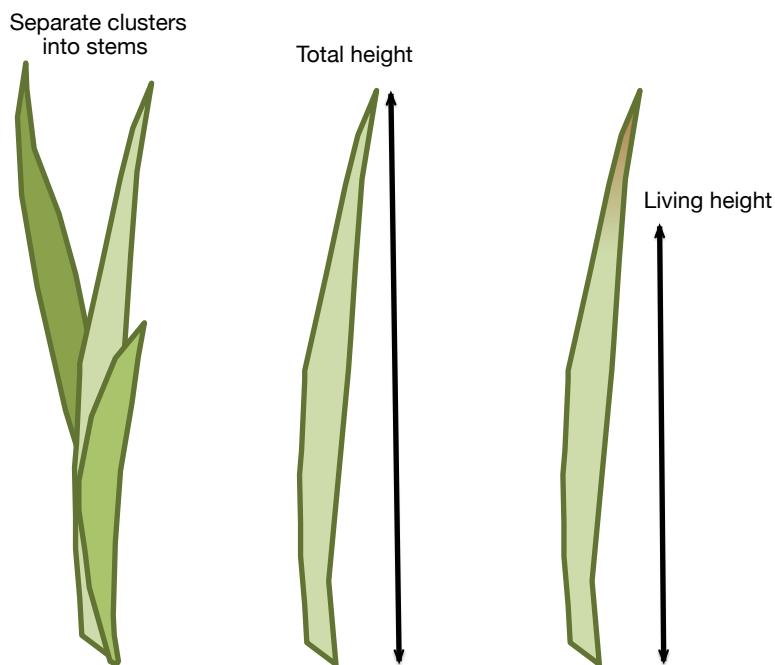


Figure 4.18 Measuring stem height and width

Developing allometric equations specific to the species and location being investigated is the most accurate way to estimate the grass carbon component. To do this, cut at least 50 individual stems over the range of observable heights for each species. Measure each individual stem for height of the green portion.

In the laboratory, determine plant biomass by oven-drying the stems to a constant weight (approximately 72 h at 60 °C, but the time will vary depending on the size of the stem). For each species, plot the results on a scatter plot representing plant biomass (y-axis) against stem green height (x-axis) (**Fig. 4.19**). Develop an allometric equation (using regression analysis) representing the relationship between plant biomass and stem height. Apply this equation to the average height of the plants sampled in the quadrat for an estimate of total mass per species within the 30 x 30 cm area.

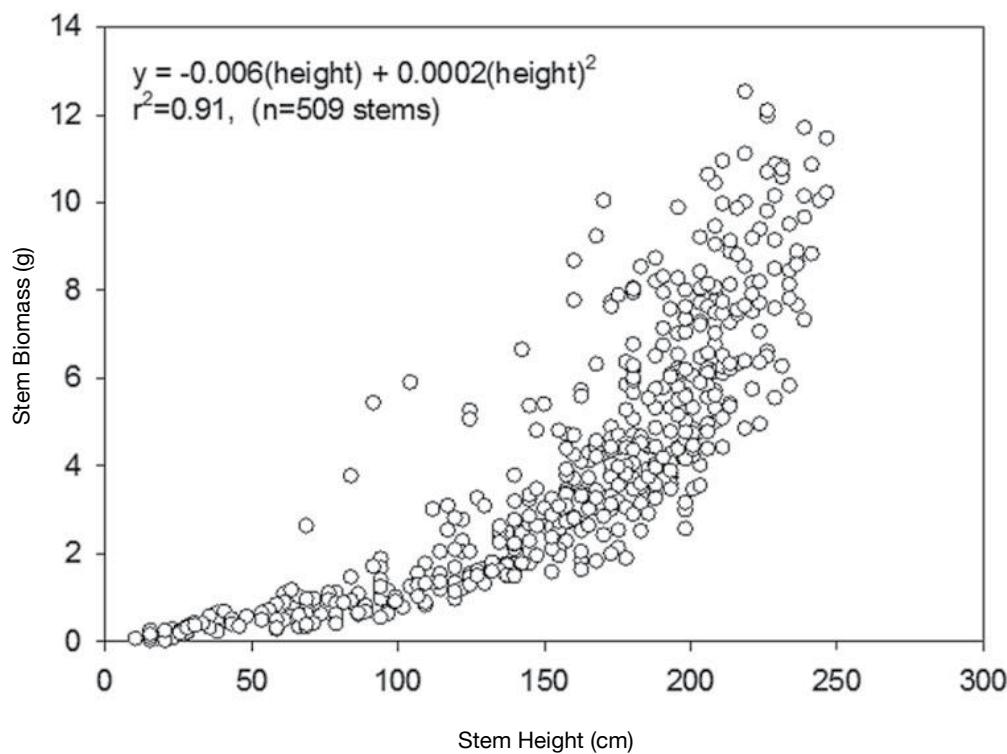


Figure 4.19 Allometric equations between stem biomass and height (Craft 2013)

Determining the carbon pool in tidal salt marsh grasses (kg C/m²): The grass carbon component is determined for each individual species by multiplying the biomass (kg)—as determined by an allometric equation—by a carbon conversion factor for the specific grass species. This is done for every stem in the quadrat. Next, all the values for individual stem carbon content are added together to determine the total carbon content from the grass (kg C) for the given quadrat size (m²).

The carbon conversion factor for grasses can either be determined through laboratory analysis using an elemental analyzer (Chapter 3) or a conversion factor of 0.45 (Fang *et al.* 1996).

EXAMPLE

- Carbon in the grass component (kg C/m²) = (Estimated biomass of the grass (kg) * carbon conversion factor (0.45)) / area of the quadrat (m²)

SHRUBS

The presence of shrubs typically varies depending on the location of the tidal salt marsh. In the northern hemisphere, shrubs make up a minor component of the marshes and are usually of the genus *Atriplex*, *Borrichia*, and *Iva* (Pennings & Moore 2001), while in the southern hemisphere—particularly in higher latitudes—shrubs of the genus *Tecticornia* may be the dominant growth form (Saintilan *et al.* 2009). The suggested approach to estimating aboveground biomass for tidal salt marsh shrubs is similar to the protocol for scrub mangroves (see section on scrub mangroves above). Currently, very few allometric equations exist for tidal salt marsh shrubs; therefore, equations should be developed on the basis of predictors including stem diameter at 30 cm above-the-ground surface, and crown volume/area or projected cover.

Crown diameter, width, volume, and area, as well as main stem diameter at 30 cm aboveground should be measured for each shrub in the plot area. To properly measure carbon content, at least 15–25 shrubs of each species, encompassing the range of sizes encountered and located outside any permanent plots that might be established, should be sampled in their entirety. In the laboratory, individual bushes are dried and weighed to obtain biomass. Biometric measurements (crown diameter, area and volume, and main stem diameter at 30 cm) can then be applied as independent variables predicting aboveground plant mass by regression analysis (**Fig. 4.20**). Once allometric equations have been established, they can be applied to every shrub within a sample plot.

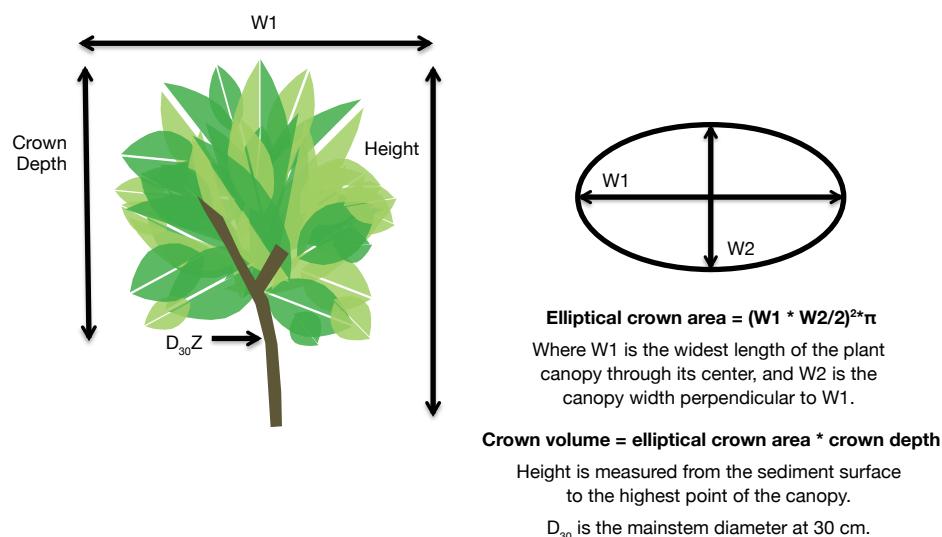


Figure 4.20 Field measurement techniques to calculate the elliptical crown area, crown depth, height, and diameter at 30 cm height (D_{30}) of shrubs (adapted from Kauffman and Donato 2012). Aboveground biomass is best predicted through allometric equations where aboveground biomass is the dependent variable and crown area, and height and/or crown volume are independent variables (Ross *et al.* 2001).

Determining the carbon pool in shrubby tidal salt marsh (kg C/m²): The shrub carbon component is determined by multiplying the biomass (kg), as determined by an allometric equation, by a carbon conversion factor for shrubs specific to that region. This is done for every shrub sampled. Next, all the values for individual shrub carbon content are added together to determine the total carbon content from the shrubs (kg C) for the given plot size (m²).

The carbon conversion factor for shrubs can either be determined through laboratory analysis using an elemental analyzer (Chapter 3), or it is justifiable to use the conversion factor reported for scrub mangrove trees (0.46 to 0.5).

EXAMPLE

- Carbon content of each shrub (kg C) = shrub biomass (kg) * carbon conversion factor (0.46–0.5);
- Carbon in shrub component (kg C/m²) = (carbon content of shrub #1 + carbon content of shrub #2 + shrub #n) / area of the plot (m²).

BELOWGROUND BIOMASS

Belowground root and rhizome biomass can contain 50–95% of the overall vegetative biomass in tidal salt marshes (Valiela *et al.* 1976; Elsey-Quirk *et al.* 2011). Belowground biomass can be estimated using allometric equations that relate belowground biomass to known aboveground biomass, or it can be determined on a site-by-site basis by direct measurements.

Few allometric equations exist that estimate belowground biomass on the basis of aboveground biomass, and those that do are based on studies conducted in North America (Valiela *et al.* 1976; Giroux & Bédard 1988; Darby & Turner 2008a). We recommend using the equations found in **Table 4.7** to estimate the total belowground biomass for a given plot size. These equations met with varying success in predicting *S. alterniflora* biomass at multiple sites, and further work is needed to derive allometric equations of belowground biomass for additional species and varying climatic and geomorphic settings.

Table 4.7 Equations to predict belowground root and rhizome biomass on the basis of aboveground biomass of *Spartina alterniflora* (Gross *et al.* 1991)

EQUATION	ABOVE-GROUND COMPONENTS INCLUDED (FOR AN ENTIRE PLOT)	r ²
$\ln(\text{Live belowground biomass, g}) = 0.718 \times \ln(\text{Live Aboveground biomass, g}) + 2.646$	Living leaves and stems	0.86
$\ln(\text{Live Belowground biomass, g}) = 0.700 \times \ln(\text{Live Aboveground biomass, g}) + 3.051$	Living leaves only	0.85
$\ln(\text{Live Belowground biomass, g}) = 0.713 \times \ln(\text{Total Aboveground biomass, g}) + 2.235$	All live and dead aboveground	0.86

Direct sampling will be the most accurate method for determining belowground biomass. To do this, samples are collected by extracting a core (see Chapter 3 for details). Most studies have taken relatively shallow cores for the estimation of belowground root biomass. However, we recommend sampling at 1 m depth because this measure correlates with established soil sampling techniques in Chapter 3, and there is evidence that salt marsh species can tap fresh water to this depth (Arp *et al.* 1993). Common protocols recommend using cores of 10 cm diameter and sampling segments 2.5 cm wide at 2.5, 5, 7.5, 12.5, 15, 22.5, and 25cm depth, and then sampling 5 cm wide segments cut at depths 35, 45, 55, and 65 cm (Saunders *et al.* 2006). we suggest adding 75 and 95 cm. Segments are washed over a 1 mm screen and visually separated into root, rhizome, and dead litter components. Living and

dead material are separated on the basis of color and texture (Saunders *et al.* 2006). Living root and rhizome materials are then oven-dried at 60 °C to constant dry weight. Belowground biomass is determined as follows:

- Belowground biomass for each sample segment (g) = dry mass of sample (g) / wet mass of sample (g);
- Belowground biomass for each sample area (entire core, g/cm²) = (Biomass of segment #1 + biomass of segment #2 + biomass of segment #n) / area sampled (based on coring diameter, cm²);
- Average belowground biomass (g/cm²) = (Biomass of sample #1 + biomass of sample #2 + biomass of sample #n) / n.

Determining the carbon within the belowground biomass (kg/m²): The belowground carbon component is determined by multiplying the biomass (kg)—either calculated from cores or from allometric equations—by a carbon conversion factor for grass species specific to that region.

If the biomass used was determined for the entire plot using allometric equations, simply multiply that number by the carbon conversion factor to determine the total belowground carbon component for your plot.

If the biomass used was determined through core sampling, the average belowground biomass for the sample area is multiplied by the carbon conversion factor and then scaled up to the plot size.

The carbon conversion factor for belowground biomass of tidal salt marsh grasses can be determined either through laboratory analysis using an elemental analyzer (Chapter 3) or by a conversion factor of 0.34 for belowground biomass of seagrasses (Duarte 1990).

EXAMPLE (BIOMASS DETERMINED BY ALLOMETRIC EQUATIONS)

- Carbon in the belowground component (kg C/m²) = (Estimated biomass of the belowground component (kg) * carbon conversion factor (0.34)) / area of the plot (m²)

EXAMPLE (BIOMASS DETERMINED BY SAMPLING)

- Belowground carbon component = Average below ground biomass (g/cm²) * carbon conversion factor (0.34)
- Because the numbers here represent the below ground carbon component found in the sample area, you can simply scale this up to your plot size

LITTER

Leaf litter is defined as recently fallen, non-wood, dead organic material on the soil surface. It may include tidal salt marsh vegetation, seagrass, or marine algal wrack. In most tidal salt marshes, the biomass of this component is relatively small due to rapid decomposition and removal by herbivores or the tide.

Litter in most studies is destructively sampled (see the section on mangrove litter above for more details). All litter easily removed from the surface is collected from a quadrat (e.g., 50 x 50 cm),

taken back to the laboratory in a sturdy bag or container, where the sample, or a representative sample thereof, is dried to a constant weight to determine biomass (kg) and converted to carbon using an estimated carbon conversion factor of 0.45.

EXAMPLE

- Carbon in the leaf litter component (kg C/m²) = (Average biomass of the litter * carbon conversion factor (0.45)) / area of the plot (m²)



Figure 4.21 Tidal salt marsh leaf litter. (© Neil Saintilan, OEH NSW)

DEAD AND DOWNED WOOD

Dead and downed wood is rarely a significant component of tidal salt marsh aboveground biomass, but it may be incorporated into a tidal salt marsh carbon stock as allochthonous material. Where this occurs in a plot, the dead wood should be sampled using the line intersect technique. Dead and downed wood biomass (kg/ha) is calculated as the volume multiplied by its mean wood density. Finally, convert downed wood biomass to carbon mass. Acceptable default values based upon carbon content (wt %) of dead wood in tropical forests are 50%, corresponding to a carbon conversion factor of 0.50. See the section above for methods of measuring dead/downed wood in mangroves.

SEAGRASS MEADOWS

Seagrass meadows are coastal marine ecosystems that are dominated by flowering plants and are completely submerged or in intertidal flood areas. There are about 60 seagrass species in four families (**Fig. 4.22**), and they are generally restricted to places with unconsolidated sediments and high light availability. Unlike mangroves, which are generally restricted to the tropics/subtropics, and tidal salt marshes, which are much more common in temperate climates, seagrass meadows are distributed globally, occurring on every continent with the

exception of Antarctica (Green & Short 2003). Seagrass meadows are valued as habitats for numerous economically and ecologically important species as well as for their high rates of primary production, enhancement of fisheries, nutrient cycling, sediment stabilization, attenuation of storm waves (Orth *et al.* 2006), and importance in the carbon cycle (Duarte *et al.* 2005; Duarte *et al.* 2010; Kennedy *et al.* 2010; Fourqurean *et al.* 2012a).

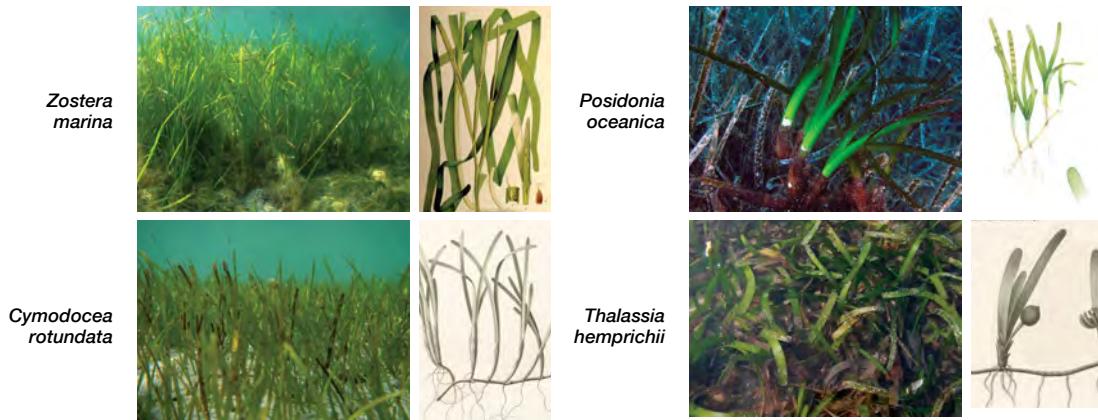


Figure 4.22 Examples of common seagrass species (© Peter Macreadie, UTS and wikicommons)

By reducing the stresses exerted by waves and currents on the soils, seagrasses reduce the amount of sediment re-suspension and enhance the trapping of suspended particles (Hendriks *et al.* 2008). They not only trap and store organic carbon generated by plants and other sources within the seagrass meadow, but also trap and bury carbon originating elsewhere (allochthonous carbon). Typically, 50% of the sediment carbon in seagrass meadows is allochthonous (Kennedy *et al.* 2010). Not all of the net production within the seagrass meadows is retained; part of seagrass ecosystem-derived carbon is also exported to other coastal and marine ecosystems where it may be consumed or buried (Heck Jr *et al.* 2008).

Saegrass meadows are also among the world's most threatened ecosystems. Rates of loss of known seagrass habitat are accelerating from a median of 0.9% per year before 1940 to 7% per year since 1990 (Waycott *et al.* 2009). Globally, the main reasons for seagrass loss are water quality degradation (e.g., eutrophication) and dredging and filling operations.

The soils that underlie seagrass meadows contain more organic carbon than terrestrial forest ecosystems (Fourqurean *et al.* 2012a). Thus the potential impact of loss of seagrass ecosystems to global CO₂ budgets is significant. In fact, the amount of organic carbon stored in seagrass meadows is roughly equal to the total amount of organic carbon stored in the world's marine tidal salt marshes and mangrove forests combined (Chmura *et al.* 2003; Donato *et al.* 2011). These estimates make seagrass meadows global hotspots for carbon storage.

Sufficient data are not available to definitively comment on regional patterns in carbon storage in seagrass ecosystems, but the data currently available are summarized by region in **Table 4.8**.

Table 4.8 Carbon stocks for a range of seagrass beds (Fourqurean *et al.* 2012b).

REGION	LIVING SEAGRASS BIOMASS (MgC/ha)		SOIL ORGANIC CARBON (MgC/ha)	
	n	Mean ± 95% CI	n	Mean ± 95% CI
Northeast Pacific	5	0.97 ± 1.02	1	64.4
Southeast Pacific	0	ND	0	ND
North Atlantic	50	0.85 ± 0.19	24	48.7 ± 14.5
Tropical Western Atlantic	44	0.84 ± 0.17	13	150.9 ± 26.3
Mediterranean	57	7.29 ± 1.52	29	372.4 ± 56.8
South Atlantic	5	1.06 ± 0.51	5	137.0 ± 56.8
Indopacific	47	0.61 ± 0.26	8	23.6 ± 8.3
Western Pacific	0	ND	0	ND
South Australia	40	2.32 ± 0.63	9	268.3 ± 101.7

*ND = no data

Field Sampling Considerations

Seagrass biomass varies seasonally, particularly in temperate and boreal climates. In some locations seagrass aboveground vegetation may be lost during winter. Therefore, we recommended measuring biomass when standing stocks are greatest and repeated sampling in subsequent years occurs at the same time of year. Seagrass can be submerged or intertidal. Sampling of intertidal seagrass can be done at low tide (as little as 3 to 4 hours), but sampling of sub-tidal seagrass meadows must be done using snorkeling or SCUBA equipment. Seagrass plants are herbaceous and relatively small; thus, small plots (0.25–1 m²) are appropriate to obtain estimates of carbon stocks. Seagrass beds vary in structure along a depth gradient due to hydrodynamic energy, reduced irradiance, and other environmental gradients such as salinity, which may affect the species composition and their ability to capture carbon (Serrano *et al.*, 2014). Thus, stratification of the study area typically aligns with depth. For example, strata can be based on intervals of known depth to obtain a reliable representation of biomass variation with water depth. At each depth interval, determine the biomass by assessing it at random positions (Chapter 2) (Short & Coles 2001).

Biomass Estimates and Carbon Content

This section provides guidance on how to measure aboveground biomass across a range of vegetation types likely to be encountered in the field. Some differences in sampling procedures are required to accommodate differences in growth forms.

LIVING BIOMASS

In most seagrass systems, the living aboveground and belowground component of biomass can be collected by inserting a large-diameter core tube (10–25 cm diameter) into the

sediment through the aboveground plant material, with care not to cut the leaves, and into the upper root dominated soils (also called the rhizosphere, typically 40 cm in depth). The tube is then capped and removed (**Fig. 4.23**). These cores are transferred to screens or mesh bags, washed free of sediment, and separated into living aboveground and belowground components (**Fig. 4.24**).



Figure 4.23 Biomass sampling/coring in seagrass meadows in Ceará, Brazil (© Cristina Rocha Barreira, UFC)



Figure 4.24 Above and belowground biomass for *Ruppia maritime* (© Margareth Copertino, FURG)

Separation of living root and rhizome material from dead structures is often ambiguous. Some of the living belowground tissues are obvious as they are typically light in color and turgid, but it can be difficult to tell if some of the older, darker-colored structures are living or dead. Local knowledge and experience are often required to ensure consistent separation of the living material from the dead. For most seagrass species, the aboveground portion of the plants are all green leaves and are easy to separate from non-green, belowground tissues. Note that in many locations, these green leaves support a considerable epiphyte load that must be removed and analyzed separately (see next section).

Determining the Carbon within the Living Vegetation Component (kg C/m²): In the laboratory, determine plant biomass by oven-drying the living biomass to a constant weight (approximately 72 h at 60 °C). The living vegetative component is determined by multiplying the biomass (kg) of a sample of plant material for a given area (m²) by a carbon conversion factor. Carbon contents are then scaled up to determine the carbon pool for the given plot size. To determine the carbon content, samples can be analyzed indirectly by determination of organic matter by loss on ignition (LOI) and use of allometric equation to estimate organic carbon or directly using an elemental analyzer (with inorganic carbon having been previously removed or accounted for by methods in Chapter 3. If this is not practical, a value of 0.34 can be used (Duarte 1990).

EXAMPLE

- Carbon in the living biomass component (kg C/m²) = (Estimated biomass of the plant * carbon conversion factor (0.34)) / area of the plot (m²)

EPIPHYTE BIOMASS

Seagrass epiphytes are organisms that grow on the blades of seagrasses, including algae, diatoms, and other encrusting organisms (Fig. 4.25). Even though epiphytes are generally a minor component of the organic carbon in a seagrass ecosystem, it is common practice to remove the epiphytes from the seagrass blades and analyze them as a separate carbon pool. Careful records must be kept regarding whether epiphytes were separated from leaf material or not to make comparison across sites possible.

There are two main techniques used for removing epiphytes from seagrass leaves: manual scraping and acid-washing (Fig. 4.26). Neither method is perfect. Scraping well-adhered epiphyte material can either result in incomplete removal of the epiphytes or it can abrade the surface of the leaves and contaminate the epiphyte component with seagrass material. Acid-washing seagrasses efficiently removes carbonate epiphytes, but fleshy epiphytes may still require scraping and the acid can leach soluble organic compounds from both the seagrass tissue and the epiphytes. If the aim is to quantify the epiphyte pool, then the scraping method is recommended.



Figure 4.25 Epiphytes. (A) example of coralline algae, (B) Example of filamentous algae (© Justin Campbell, Smithsonian Institute)



Figure 4.26 Epiphyte removal and processing. (A) Removal of epiphytes from both sides of the seagrass leaf using a blade, (B) Epiphytes (algae) and a clean seagrass sample, and (C) Dried epiphytes undergoing an acid wash (© Oscar Serrano, ECU)

Determining the Carbon within the Epiphyte Component (kg C/m^2): Epiphyte removal is conducted at the laboratory. Therefore, if using a permanent plot system the samples must be taken from an area of known size outside of the plot. Some types of epiphytes will contain high levels of calcium carbonate; thus, determining the inorganic carbon in the epiphyte component is needed. Procedures are similar as those described for soils with high levels of carbonate described in detail in Chapter 3 and briefly below.

- 1) Epiphytes are removed from both sides of the leaf by scrapping with straight edge blade (**Fig. 4.26**).
- 2) Epiphytes dried at 60°C to a constant weight.
- 3) When dry, homogenise the sample and determine the weight of sample.
- 4) Inorganic carbon is determined in one of two methods:
 - a. Acidification:
 - Soaking the sample in 1N HCL for approximately 18 hours, followed by washing the sample three times in distilled water; dry to constant weight, and analyze carbon content using an elemental analyzer or LOI Test.
 - Note that acidification can leach organic matter.
 - b. Elemental Analyzer:
 - Determine the total carbon content of a dry subsample via an elemental analyzer; ash a second subsample at 500°C ; weigh the remaining ash; determine the inorganic carbon content using an elemental analyzer; and subtract the inorganic content from the total carbon content.

Always keep track of sample masses at all stages of the procedure so you can calculate the % inorganic and organic carbon. The carbon content is corrected for the loss of inorganic carbon to produce a carbon conversion factor. The epiphyte sample biomass is multiplied by the corrected carbon conversion factor and then averaged across all samples to determine the carbon pool for the given plot size (m^2).

LITTER

The litter in seagrass meadows typically consists of dead leaves, rhizomes, fruits, and algae. In most seagrass settings, the amount of litter (and therefore the carbon stock) only constitutes a very small component of the carbon pool due to the high remineralization efficiency of leaves as well as export through hydrodynamic energy.

If it is measured, litter is destructively sampled through collection from microplots (e.g., 0.5 x 0.5 m in size). All organic surface material is collected into a bag or container. The bags are labelled with the location, date, plot, and sample number.

The samples need to be transported to the laboratory and dried to constant mass (72 h at 60 °C). Due to constraints of carrying bulky samples and the often-limited oven drying space—especially in rural field settings—we recommend measuring the wet weight of the entire sample and then extracting a well-mixed representative subsample for transportation to a laboratory, and determining the ratio between wet and dry mass of the subsample for scaling-up the biomass of the bulk sample.

EXAMPLE

- Biomass of litter (kg) = (dry mass of subsample (g) / wet mass of the subsample (g)) * wet mass of all the litter in the sample plot (kg)

The carbon content in the litter biomass (kg) can then be estimated for the given plot size (m^2). The conversion factor for living biomass (0.34) can also be used to estimate the mean carbon content of seagrass litter dry mass.

EXAMPLE

- Carbon in the litter component (kg C/ m^2) = (Average biomass of the litter * carbon conversion factor (0.34)) / area of the plot (m^2)

TOTAL CARBON STOCK

The total vegetative carbon stock within a project area is determined by adding the carbon content of each vegetative component (tree, litter, grass, shrub, etc.) within a defined area. The total carbon stock in a project area can be determined as follows:

- Step 1:** Determine the carbon content of each component for your plot size (see above).
- Step 2:** Convert the carbon content of each component for your plot size into the units commonly used in carbon stock assessment (Mg C/ha) using the following unit conversion factors (There are 1,000 kg per Mg and 10,000 m^2 per ha):

Vegetative component carbon pool (Mg C/ha) = Carbon content (kg C/ m^2) * (Mg/1,000 kg) * (10,000 m^2 /ha).

REPEAT FOR EACH VEGETATIVE TYPE MEASURED

- Step 3:** Sum all the vegetative components present within a single plot.

Total vegetative carbon in a plot (Mg C/ha) = component #1 (Mg C/ha) + component #2 (Mg C/ha) + component #3 (Mg C/ha) + ...

REPEAT FOR ALL PLOTS

Step 4: Determine the average amount of vegetative carbon in a plot and calculate the associated standard deviation to determine variability/error.

Average vegetative carbon in a plot = Total vegetative carbon for plot #1 (Mg C/ha) + Total vegetative carbon for plot #2 (Mg C/ha) + Total vegetative carbon for plot #3 (Mg C/ha) + n) / n

Standard Deviation (σ) determines how closely the data are clustered about the mean, and is calculated as follows:

$$\text{Standard Deviation } (\sigma) = \left[\frac{(X_1 - \bar{X})^2 + (X_2 - \bar{X})^2 + \dots + (X_n - \bar{X})^2}{(N-1)} \right]^{1/2}$$

- \bar{X} = average vegetative carbon in a plot
- X_1 = individual result for plot #1, in MgC/hectare; X_2 = individual result for plot #2, in MgC/hectare, etc.,
- N = number of plots.

Step 5: Obtain the total amount of carbon in the ecosystem.

Estimate of vegetative carbon in a stratum (Mg C) = Average vegetative carbon in a plot (Mg C/ha) * area of stratum (ha)

Sum the MgC values for each stratum to determine the total soil carbon stock.

Total carbon in a project area (MgC) = Estimate of vegetative carbon in stratum #1 (Mg C) + Estimate of vegetative carbon in stratum #2 (Mg C) + Estimate of vegetative carbon in stratum #3 (Mg C) + ...

Step 6: To report a value for the variability/error associated with these measurements, calculate the total uncertainty in the data. First, calculate the standard deviation of the average Mg C for each stratum. [Multiply the standard deviation carbon value (MgC/hectare) for each core determined in step 5 above by the area of each stratum (in hectares).] Then propagate the uncertainty through the calculations by combining the standard deviations of the average MgC for each stratum as follows:

$$(\sigma_T) = \sqrt{(\sigma_A)^2 + (\sigma_B)^2 + \dots + (\sigma_N)^2}$$

- Where σ_T = the total variability associated with the measurements
- σ_A = standard deviation of the core average MgC for stratum A * area of stratum
- σ_B = standard deviation of the core average MgC for stratum B * area of stratum
- σ_N = standard deviation of the core average MgC for remaining stratum * area of each individual stratum.

This approach can be used when adding average values, as is done when combining the data from the individual strata.

Step 7: The final vegetative carbon stock will be presented in an average value \pm the total uncertainty. Alternatively, a minimum and maximum carbon stock can be used by multiplying by the project area by the minimum and maximum carbon densities.

Total organic carbon in a project area (calculated in Step 5) \pm the standard deviation (calculated in Step 6)

QUICK GUIDE**Step 1: Plot design**

- Accessibility and safety are the number one priorities when choosing plot areas.
 - Remember each site may only be accessible for a few hours.
- Stratify the study site.
- Decide if the sampling plots will be permanent or temporary.
 - Permanent plots are used if the same location will be assessed in the future to determine change.
 - Temporary plots are used when sampling will only be done once or when permanent plots are not feasible.

Step 2: Measure vegetative components (above- and belowground as well as living and dead components)

- Determine the relevant components for the study site and the scale of the sampling area needed for each.
 - Mangrove forests
 - Live trees
 - Scrub mangrove trees
 - Standing dead trees
 - Lianas
 - Palms
 - Pneumatophores
 - Leaf litter
 - Dead and downed wood
 - Belowground tree biomass (if an allometric equation is available)
 - Tidal Marshes
 - Shrubs
 - Grasses and reeds
 - Leaf litter
 - Dead and downed wood
 - Seagrass meadows
 - Living plants
 - Epiphytes
 - Leaf litter
- Determine biomass either by laboratory analysis or by allometric equations.
- Determine the carbon content for a known plot size.

Step 3: Calculate Total Vegetative Carbon Stock

- Sum the relevant component carbon pools for each plot.
- Determine the average and standard deviation for plots within a stratum and multiply by the stratum area. Repeat for each stratum.
- Determine total vegetative carbon stock for the project area by summing carbon stocks within each stratum and calculating the error.
- You must include the variability associated with the measurements and the total soil depth assessed when reporting results.

