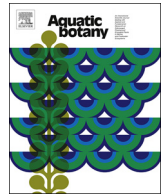




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Short communication

## Species-specific wet-dry mass calibrations for dominant Northeastern Pacific Ocean macroalgae and seagrass

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## ABSTRACT

Macroalgae and seagrasses form the base of productive ecosystems in the Northeastern Pacific Ocean. Often, ecological research on macroalgae, seagrasses, and sea wrack requires the conversion of biomass from wet to dry to create consistency across investigations. This process, however, can be destructive, impractical, time consuming, and labour intensive. We collected samples of twelve common Northeastern Pacific Ocean seaweed species (*Alaria marginata*, *Codium fragile*, *Egregia menziesii*, *Fucus distichus*, *Macrocystis pyrifera*, *Mazzaella* spp., *Nereocystis luetkeana*, *Pterygophora californica*, *Pyropia* spp., *Ulva* spp., and the seagrasses *Zostera marina* and *Phyllospadix* spp.) in two states: wet and fresh, or aged and partially desiccated. We weighed, dried, and compared samples, finding that many species displayed a strong ( $R^2 > 0.8$ ) and predictable linear relationship between wet and dried conditions. In contrast, half the aged samples did not have a significant relationship between partially desiccated and dried conditions. Our results provide practical wet weight to dry weight ratios for many species, and with further research, a reliable set of species-specific wet to dry weight ratios for all species could be established. Wet to dry weight ratios are useful for macroalgae, seagrass, and sea wrack research or commercial applications and would reduce the need to conduct extensive wet-dry calibrations in each study.

## 1. Introduction

Ecological research on macroalgae, seagrass, and sea wrack (detached, shore-cast macroalgae) has been fundamental to our understanding of ecosystem functioning (Filbee-Dexter and Scheibling, 2014), habitat connectivity (Dugan et al., 2003), community structure (Smale et al., 2016) and species distribution (Smith et al., 2018). Macroalgal research also continues to offer a key understanding of anthropogenic stressors to oceanic communities (Johnson et al., 2011; Brodie et al., 2014; Filbee-Dexter and Wernberg, 2018) and the ecosystem services provided by macroalgae (Krause-Jensen and Duarte, 2016). Additionally, the demand for macroalgae in commercial products (such as nutraceuticals and foods) has intensified and is predicted to rise (Dhargalkar and Pereira, 2005; Jiménez-Escrig et al., 2012). Obtaining accurate estimates of the biomass of macroalgae, seagrasses, and sea wrack is central to their study. However, some biomass estimation methods are destructive and can have long term ecological

consequences (Watt and Scrosati, 2013). Therefore, non-destructive methods of accurate and efficient biomass estimation of macroalgae, seagrass, and sea wrack can provide a valuable contribution to future research.

In the Northeastern Pacific Ocean, sub-tidal and intertidal biomass is dominated by various species of macroalgae (hereafter referred to as seaweeds) and seagrass. Kelps (Order Laminariales) are among the fastest growing algae in the world, and form vast and ecologically important underwater forests (Duggins et al., 1989; Steneck et al., 2002). Others, like eelgrass (*Zostera* spp.) comprise extensive mats across the shallow subtidal that provide key nursery habitats for species of ecological, cultural, and economic value, such as salmon (*Oncorhynchus* spp.) (Simenstad et al., 1982) and rockfish (*Sebastes* spp.) (Olson, 2017). Although work on seaweeds and sea wrack has well-established methods for sampling and surveying techniques and equipment in other parts of the world (e.g., Stagnol et al., 2016), a gap in knowledge remains in understanding the relationship between wet and dry biomass

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for seaweed species commonly found in the Northeast Pacific Ocean. Establishing wet-dry calibrations for dominant seaweeds of this large region would facilitate simplified future research on these ecologically and economically important species.

Dry mass measurement is considered the most reliable method for precise biomass estimates of seaweeds (Stagnol et al., 2016). This method enables consistent results across investigations (Bickel and Perrett, 2015). Dry mass can also account for variation in water content of different seaweed species. Additionally, it has the potential to account for the variation in desiccation levels of sea wrack deposits along shorelines. When feasible, seaweeds are sorted by species, functional group, or a subsample of a mixture of several seaweed species is oven dried to a constant weight at which no further mass loss is recorded (Madsen, 1993). After dry mass determination, the most commonly used approach is to regress (e.g. Orr et al., 2005; Fox et al., 2014) or use standard major axis testing (e.g. Gevaert et al., 2001, 2008; Stagnol et al., 2016) to determine the relationship between wet and dry mass. The slope is then used to create a wet weight to dry weight ratio (DW:WW) (e.g. Gevaert et al., 2001; Orr et al., 2005).

Obtaining dry mass measurements is labour intensive, time consuming, and sometimes problematic. For instance, in field studies with large amounts of macrophyte biomass, dry mass measurements may not be feasible. Performing wet-dry calibration work in remote field locations without electrical power can be impractical. Additionally, one common genus of marine macroalgae (*Desmarestia* spp.) contains sulphuric acid in its tissue that will rapidly disintegrate itself and any co-occurring specimens after collection (Druehl and Clarkston, 2016). A potential problem in beach wrack dry mass measurements is that many studies determine their dry mass estimates based on a subsample of a mixture of species, not a species-specific subsample (e.g. Mews et al., 2006; Orr et al., 2005). Lastly, dry mass determination is destructive and will render the organism unsuitable for further research and may remove large amounts of seaweeds from natural settings, which can disturb ecological processes (Dugan et al., 2003).

Despite the substantial amount of information available regarding seaweed survey and sampling techniques available from locations across the globe, we found no information on standardized regression coefficients obtained from dry mass determination for the twelve common seaweed species we sampled specific to the Northeastern Pacific Ocean. Therefore, each study that requires dry seaweed biomass measurements for these species must perform the time-consuming and labour-intensive step of dry mass determination, or resort to destructive biomass measurements. In this paper, we present the results of wet-dry calibrations for twelve common macroalgae and seagrass species collected from the Northeastern Pacific. By sharing these results, and with further research, we can provide a suite of standardized wet to dry weight ratios to use in estimating the dry biomass of macroalgae, seagrass, and wrack in western North America.

## 2. Methods

We collected 252 macroalgae and seagrass specimens from three sandy beaches and two rocky intertidal outcrops located along the northwest end of Calvert Island, British Columbia, Canada (51.6536°N, 128.1301°W) in July (n = 223) and September (n = 29) 2016. Samples contained twelve species: *Alaria marginata*, *Codium fragile*, *Egregia menziesii*, *Fucus distichus*, *Macrocystis pyrifera*, *Mazzaella* spp., *Nereocystis luetkeana*, *Pterygophora californica*, *Pyropia* spp., *Ulva* spp. (bladed form), and the seagrasses *Zostera marina* and *Phyllospadix*. Samples of various weights (27–265 g) were collected indiscriminately from shoreline wrack accumulations (n = 134) and harvested live from rocky intertidal shorelines (n = 60). Collected samples were sorted into two categories based on their moisture saturation levels: aged or wet (see Supplementary Table 1 for details of the number of samples in the aged and wet categories collected during each period). Aged samples were collected from wrack deposited in lines parallel to the water's

edge that had been stagnant on the beach for over 24 h, confirmed visually during beach surveys over a six-day period. Aged samples were collected opportunistically from these wrack lines and contained a subset of the studied species: *A. marginata*, *C. fragile*, *E. menziesii*, *F. distichus*, *M. pyrifera*, *N. luetkeana*, *Phyllospadix* spp., and *Ulva* spp., all of which were partially desiccated. Wet samples were collected opportunistically from samples that had been deposited recently on the shoreline (confirmed visually during surveys) and harvested live from rocky intertidal outcrops at low tide. All wet samples showed no signs of desiccation.

Blades, stipes, and (when applicable) pneumatocysts were collected. Samples were sorted and separated into species and for three kelp species (*M. pyrifera*, *N. luetkeana*, and *P. californica*), samples were processed as “blade only” or “stipe only” categories. However, because most researchers may not separate kelp blades from stipes, we also processed samples that were portions of entire kelp individuals: stipes with attached blades and pneumatocysts (“mixed” category). Samples were rinsed, cleaned free of sand and epiphytes, and shaken to remove surface water. Wet weights of all samples were measured on a Denver Instrument MXX-612 balance and transferred into a Fisher Scientific Isotemp drying oven within one hour of collection. Samples were dried at 80 °C until their dry weight reached a constant mass ( $\pm 0.005$  g) for three consecutive measurements (typically this took about 72 h).

All data were analyzed in R package version 3.3.3 (R Core Team, 2017). We performed standardized major axis tests (as per Warton et al., 2006) and used the slope of the linear equation to generate wet to dry weight ratios (Table 1). Using package ‘smatr’ (Warton et al. 2012) we compared the slopes of wet samples that were harvested live against wet samples that were collected from wrack lines as well as samples that were collected in July against samples that were collected in September. Additionally, we compared slopes for the three dried combinations of samples (blade only, stipe only, and mixed) of the kelps *M. pyrifera*, *N. luetkeana*, and *P. californica*. The ‘smatr’ package fits bivariate lines in allometry using standardized major axis testing to make inferences about the lines.

Using the package ‘lmodel2’ (Legendre, 2014) we plotted the confidence intervals of the wet samples for four species that had potential outliers, low sample size, and heterogeneous data.

## 3. Results

Fifteen of eighteen categories from twelve species of macroalgae and seagrass exhibited strong ( $R^2 > 0.8$ ) and significant linear relationships ( $p$ -values  $< 0.05$ ) between wet and dry mass when dried from wet samples (*C. fragile*, *E. menziesii*, *F. distichus*, *M. pyrifera* mixed, *M. pyrifera* blade, *M. pyrifera* stipe, *Mazzaella* spp., *N. luetkeana* blade,

**Table 1**

Wet to dry weight ratios (WW:DW) for the nine species across twelve categories of seaweeds that displayed strong ( $R^2 > 0.8$ ) and significant ( $p$ -value  $< 0.05$ ) relationships when dried from wet. DW stands for dry weight and WW stands for wet weight. Species with outlier driven relationships were not included.

Species	WW:DW
<i>Codium fragile</i>	DW = 0.069WW
<i>Egregia menziesii</i>	DW = 0.189WW
<i>Fucus distichus</i>	DW = 0.195WW
<i>Macrocystis pyrifera</i> mixed	DW = 0.115WW
<i>Macrocystis pyrifera</i> blade	DW = 0.168WW
<i>Macrocystis pyrifera</i> stipe	DW = 0.121WW
<i>Mazzaella</i> spp.	DW = 0.192WW
<i>Nereocystis luetkeana</i> stipe	DW = 0.045WW
<i>Pterygophora californica</i> mixed	DW = 0.151WW
<i>Pterygophora californica</i> stipe	DW = 0.245WW
<i>Pyropia</i> spp.	DW = 0.307WW
<i>Zostera marina</i>	DW = 0.224WW

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