

Variation in Carbon Sediment Storage across Salish Sea Eelgrass Habitats

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**Abstract**

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Seagrasses are marine flowering plants that are found globally in coastal habitats. Seagrass meadows serve as valuable carbon sink habitats by trapping and storing carbon within the sediment. The ability of these habitats to store carbon is variable region to region. It is unclear what biotic or abiotic factors contribute to the success of carbon burial within these habitats in the Salish Sea. This study assesses sediments stored in *Z. marina* meadows and unvegetated sites within two distinct estuaries located in the Salish Sea. The two study sites represent estuaries located within an active delta and an inactive delta. I evaluated the carbon stock and isotopic signatures of these sites to understand how vegetation and location can affect the origin and amount of carbon stored in these habitats. I found that carbon stock did not significantly differ between sites regardless of estuary type or presence of *Z. marina*. This may indicate that physical processes may be the primary driver of carbon storage instead of the presence of *Z. marina* within these sites. The total carbon stock integrated over the top 50cm of sediment was lower than other published estimates with an average of 8.30 Mg ha<sup>-1</sup> within an active delta and 11.62 Mg ha<sup>-1</sup> within an inactive delta. Isotopic signatures of carbon and nitrogen were statistically significantly depending on site vegetation and location. At the current Californian market price of carbon, the mean value of all sites in this study equates to \$229,214 per square mile within the

top 50cm of sediment. I conclude that there is a need for more information to determine the abiotic influences on carbon sediment storage variability across the Salish Sea.

## Introduction

With the increase of anthropogenic emissions of carbon dioxide, many climate change mitigation plans have looked towards either reducing carbon emissions or expanding and protecting carbon storage habitats, also known as carbon sinks. In a national political climate that ranges from hesitant to obstinate with regards to controlling carbon emissions, interest has shifted towards developing carbon abatement and mitigation tools on a regional scale to curtail climate change without the gridlock of national politics.

Within the past decade, the scientific community has focused its attention on evaluating the carbon sink capacity of marine ecosystems including mangroves, tidal marshes, and seagrass beds. Coined “Blue Carbon”, recent research has determined that these three ecosystems store significant amounts of carbon within the plant biomass and sediments of these habitats despite their small global footprint (Duarte et al., 2005). Seagrasses, in particular, have received increased attention due to their small geographic extent and their potentially high carbon sink capacity (Kennedy et al., 2012; Fourqurean et al., 2012; Duarte et al., 2005). However, there is a great deal of variation in the carbon storage reported for seagrass habitats. This variation is likely a product of differences in biological variables such as plant species, distribution, and plant morphological characteristics, and physical variables such as turbidity, wave height and hydrogeomorphic variations in the habitats where seagrasses are found (Samper-Villarreal et al., 2016; Saintilan et al., 2013). Untangling the biotic and abiotic factors that influence the amount of carbon that is stored in seagrass meadows has been the focus of many studies (Samper-Villarreal et al., 2016; Lavery et al., 2013).

Seagrasses encourage sediment deposition at the site of meadow habitats (Duarte et al., 2013) primarily through the presence of a dense canopy. Seagrass canopies can reduce water flow by dampening currents and waves which causes particles and sediment in the water column to settle out, resulting in localized accretion of sediments (Bos et al., 2007; Fonseca and Fisher, 1986). Apart from the physical changes in the depositional environment, seagrass biomass is also stored at these sites in the form of plant detritus and rhizomes deposited in the sediment. Seagrass plant matter is slow to break down due to the low phosphorus and nitrogen content within plant tissues (Enriquez et al., 1993; Duarte, 1990; Harrison, 1989). Most importantly, any carbon deposited within marine sediments is additionally inhibited by low oxygen levels that discourages decomposition (Duarte, 2005). The origin of the organic carbon stored within the sediment can be determined using carbon and nitrogen isotopic signatures. Other studies have shown that seagrasses isotopic signatures are more enriched in  $\delta^{13}\text{C}$  when compared to other coastal sources of carbon such as plankton, algae or terrestrial plants (Rohr et al., 2016; Kennedy et al., 2010). These distinctive signatures can be used to identify the sources of the organic carbon stored in the sediment.

Seagrass habitat has been declining at a rapid rate that matches mangrove habitat loss and has bypassed the rate of loss of tropical forests (Waycott et al., 2009; Orth et al., 2006). Most of this habitat loss is due to the fact that seagrasses inhabit relatively shallow water and have high light requirements when compared to phytoplankton and benthic algae (Denisson et al., 1993). The shallow coastal distribution increases the risk that seagrass habitat is impaired by coastal development and land use activities. When seagrass habitats are degraded, not only is active carbon storage potential diminished, but carbon stored in the sediment can be released. It is

estimated that disturbed seagrass beds can release up to 299 Tg of carbon per year (Fourqurean et al., 2012).

In the Salish Sea, the dominant native seagrass species is *Zostera marina*. *Z. marina* is an aquatic flowering vascular plant with blades that reach 2 meters in length. Roots branch out from a rhizomatous mat (Mumford et al., 2007) which keep it embedded within the sediment. Regional threats to *Z. marina* habitat include competition with algal blooms, shading by overwater structures, shoreline armoring, sediment loading, and vessel moorage (Washington State Department of Natural Resources, 2015). In 2011, the Puget Sound Partnership, a Washington State agency, adopted seagrass coverage as an indicator of the biophysical conditions within Puget Sound. The agency and partners developed the *Puget Sound Eelgrass Recovery Strategy* with the goal to increase seagrass extent by 20% by 2020 (Washington State Department of Natural Resources, 2015). The Washington State Department of Natural Resource (DNR) has taken on the primary role of monitoring seagrass habitat within the Puget Sound region.

Little research has been conducted on the potential seagrass carbon sink within the Salish Sea. A previous study (Poppe, 2015) found surprisingly low carbon accumulation rates in Padilla Bay when compared to global estimates. However most global estimates have been based on studies conducted in Australian or Mediterranean meadows of *Posidonia* species (Lavery et al., 2013). These estimates may not be broadly generalizable to other seagrass species and other regions due to plant morphological and site physical characteristics. Understanding the localized biotic and abiotic variables that contribute to the success of the seagrass carbon sink can help regional managers better understand the controls on carbon storage in these environments and the potential for carbon release if these habitats are degraded.

To better understand how location and vegetation can influence carbon storage capacity in nearshore sediments, I estimated the carbon storage capacity of sediments of two shallow estuaries in the Salish Sea, WA, and measured stable isotopes of carbon and nitrogen. I compared vegetated and unvegetated plots in an active delta, Skagit Delta, versus an inactive delta, Padilla Bay. These two sites have a distinctive land use history. Historically, the two sites were fed by the same Skagit and Samish river systems. In the early 1900s the construction of agricultural dikes disconnected one site from its primary sediment source, creating an inactive delta. The active delta maintains a connection to the watershed and its primary sediment source, the Skagit River.

I hypothesized that:

- a) carbon storage capacity would be higher in vegetated plots compared with unvegetated plots,
- b) carbon storage capacity would be higher in the active delta compared with the inactive delta, and
- c) profiles of stable isotopes of carbon and nitrogen would differ based on geomorphic and vegetation characteristics of each site.

I compared the results with global estimates and evaluated the findings in the context of regional management.

## Methods

### Study Area

I focused my study on two shallow estuaries, the Skagit Delta and Padilla Bay, in the Salish Sea, WA. The Salish Sea is an international inland waterbody that borders the United States, Washington and Canadian, British Columbian coasts. It is made up of a network of straits, bays and sounds with over 64 freshwater inflows (Mohamedali, et al., 2011). I chose the Skagit River Delta as an example of an active river delta and nearby Padilla Bay as an example of an inactive river delta. Both sites are classified as intertidal shallow seagrass habitat and are subject to semidiurnal tides (Bulthuis 2010).

The Skagit River is the third largest river that drains into the Salish Sea after the Fraser and Snohomish Rivers (Mohamedali, et al., 2011). The river's average annual discharge is approximately  $470 \text{ m}^2 \text{ s}^{-1}$  (Sutherland et al., 2011) and contributes up to 35-50% of freshwater to the Puget Sound (Hood 2007; Yang & Khangaonkar 2008). At 240 km in length, the Skagit River's drainage basin encompasses  $8500 \text{ km}^2$ , including productive agricultural lands along a floodplain (Hood 2007; Webster et al., 2013). River outflow is distributed between the North Fork at 55-70% and the South Fork at 30-45% respectively (Webster et al., 2013; Hood 2007; Yang and Khangaonkar 2009). The Skagit River is the primary source of sediment to the marine environment.

In the Skagit Delta estuary, *Z. marina* distribution ranges from 0.03 ft. mean lower low water (MLLW) to -17.40 ft MLLW (Washington State Department of Natural Resources, 2016). Surveys of the Skagit Delta seagrass habitat show declining *Z. marina* growth in the North Fork and stable *Z. marina* growth in the South Fork (Washington State Department of Natural Resources, 2016). Core samples were taken along the length of the Skagit Delta at the north fork, the middle, and the south fork.

The Padilla Bay estuary is a shallow inactive delta located to the north of the Skagit River. Historically it was one of the distributary channels of the Skagit and Samish rivers during periods of high river discharge (Bulthuis, 2010). In the early 1900s, Padilla Bay was diked to support lands suitable for agriculture, essentially cutting off all freshwater sediment sources to the bay. Now Padilla Bay only receives freshwater from small sloughs maintaining the agricultural fields and to drain the adjacent floodplain (Bulthuis, 2010). Padilla Bay has a large *Z. marina* meadow that grows alone or in mixed stands with the smaller *Zostera japonica* species. *Z. marina* and *Z. japonica* distribution ranges from 5.80 ft. MLLW to -16.80 ft. MLLW. Surveys of the Padilla Bay seagrass habitat show increasing *Z. marina* growth (Washington State Department of Natural Resources, 2016). Core samples were taken along an established monitoring transect in the northeast of the reserve.

## Sediment Sampling

I collected sediment samples in August and September 2016 during low tide periods. At each site, I collected three sediment cores from vegetated habitat and three from unvegetated (bare) mudflat. Sediment cores were taken with a 4 inch polyvinyl chloride (PVC) corer to a maximum of 1 meter depth. The coring device was hammered into the sediment and capped with a mechanical test plug at the top of the core and a flexible PVC cap at the bottom. Cores were extracted from the sediment while maintaining a vacuum using a winch puller and tripod. Excess water was drained off the top of the core and the entire core was transported back to the laboratory in an upright position where it was refrigerated at 37°F until extruded. Cores were extruded and subsampled every 2 cm from the top of the core to 20 cm in depth, every 5 cm from 20-50 cm in depth and every 10 cm from 50 cm in depth to the end of the core. Total core length averaged 70.8 cm; because core lengths varied, I normalized the length to 50 cm for all analyses.

I retained a portion of each subsample for grain size analysis and to calculate bulk density. For analysis of bulk density, one 30 cc aliquot of each subsample (rhizomes included) was weighed (wet). For grain size analysis, subsamples were wet-sieved for 10-20 minutes using a 63  $\mu\text{m}$  sieve to separate fine particles from sand-sized particles. All samples were then dried in an oven at 55°C for ~72 hours. When dry, the samples were weighed again to calculate bulk density or sand and fine fractions. Because calcium carbonate precipitates were not visible within the cores, I did not acidify the samples before subjecting them to elemental analysis. Consequently, the values I measured represent total carbon, not total organic carbon. I followed the procedure outlined by The Blue Carbon Initiative (Howard et al., 2014) to calculate carbon density ( $\text{mg}/\text{cm}^3$ ) of each of my subsamples in order to determine total carbon stock in  $\text{Mg C ha}^{-1}$  within each core. Compaction within the core ranged from 1-14 cm in the Skagit Delta and from 1-7 cm in Padilla Bay. I did not correct total carbon values for core shortening and the potential increase in bulk density through compaction.

To measure total carbon, total nitrogen, and associated isotopic ratios, aliquots of the homogenized subsamples from each core segment were placed in vials and freeze-dried with a VirTis Benchtop SLC at -40°C for ~72 hours. Samples were then weighed with a microbalance and encapsulated in 5x9 mm tin capsules and sealed in 96-well trays to be shipped for analysis. Total carbon, nitrogen and isotopic analysis was carried out by the University of California Stable Isotope Facility (Appendix A).

Carbon and nitrogen isotopic ratios were expressed using the following equation, where  $R$  is the ratio between  $^{14}\text{C}:^{13}\text{C}$  or  $^{15}\text{N}:^{14}\text{N}$ :

$$\text{Heavy Isotope} = \frac{(R_{\text{sample}} - R_{\text{standard}})}{R_{\text{standard}}} \times 1000$$

## Statistical Analysis

I used a two-way ANOVA to test for differences between the factors of active/inactive delta and vegetated/unvegetated *Z. marina* sites on my response variable of total carbon stock. I also used a two-way ANOVA to test the same factors against the response variables of carbon and nitrogen isotopic ratios.



I applied multiple linear mixed effects models using data from all core subsamples normalized to 50 cm. For these models I used carbon density,  $\delta^{13}\text{C}$ , and  $\delta^{15}\text{N}$  isotopic ratios as the response variables. I used fixed factors of active and inactive delta, presence or absence of vegetation, whether the sample was located in the top 20 cm of the core, and percent fine grain size. I visually inspected each model's residuals to ensure they met classical linear model assumptions. Within the carbon density model, heteroskedasticity was present. To correct for this, the response variable of carbon density was  $\log_{(10)}$  transformed. Significance values were obtained from likelihood ratio tests of the full model against an alternative model with that factor removed.

## Results

### Carbon Content

Carbon stocks did not differ between sites or vegetation status (Figure 1). Mean total carbon stock in the top 50 cm of sediment from an active delta (Skagit Delta;  $14.9 \pm 2.1 \text{ Mg C ha}^{-1}$ ) was not significantly different from carbon stock measured from an inactive delta site (Padilla Bay;  $19.3 \pm 2.8 \text{ Mg C ha}^{-1}$ ;  $F_1 = 1.68$ ,  $P < 0.05$ ). Nor did carbon stocks differ between vegetated and unvegetated plots ( $F_1 = 0.01$ ,  $P < 0.05$ ). Contrary my hypothesis (a/b), the core with the highest sedimentary carbon content was taken from unvegetated habitat in the inactive delta site. The lack of statistically significant differences between cores from vegetated versus unvegetated plots could be explained by the high variance and low sample size.

According to the results of a linear mixed effects model, the percent of fine sediment in each subsample was the best predictor of the carbon density in subsamples ( $X^2(1) = 84.6$ ,  $P < .05$ ). The second best predictor of carbon density was location in the core, specifically whether or not the sample came from the top 20 cm ( $X^2(1) = 9.96$ ,  $P = 0.002$ ). Neither vegetation status (vegetated/unvegetated) ( $X^2(1) = 2.14$ ,  $P = 0.1436$ ) or geomorphic status (active/inactive delta) ( $X^2(1) = 0.39$ ,  $P = 0.53$ ) had any effect on carbon density with depth.

### Sediment Grain Size

Carbon stock was highest in a core taken from the inactive delta (Padilla Bay), located in unvegetated, bare mudflat habitat. This finding could be due to an isolated deposit of fine sediment that was absent from neighboring cores. This interpretation is consistent with a finding of a significant moderate positive relationship ( $r(14) = .85$ ,  $P < 0.05$ ) between percent fine sediment and carbon density within this core that did not appear in other samples ( $r(190) = .62$ ,  $P < 0.05$ ).

**Table 1. Sediment carbon results of this study compared to other published results. Values of carbon density were within the lower range of other *Z. marina* findings. Carbon stock results from this study were also low compared to other published seagrass results.**

<i>Source</i>	<i>Seagrass Species</i>	<i>Study Location</i>	<i>Carbon Density mg/cm<sup>3</sup></i>	<i>C<sub>org</sub> Stock (25cm) g C/cm<sup>2</sup></i>	<i>C<sub>org</sub> Stock Mg C/ha</i>	<i>% C<sub>org</sub></i>
This Study	<i>Z. marina</i>	Skagit Delta, USA	3.36	0.08**	8.30**	0.36**
		Padilla Bay, USA	4.77	0.12**	11.62**	0.49**
		Finland	2.60	0.06*	6.27*	
Rohr et al. 2016	<i>Z. marina</i>	Limfjorden, Denmark	17.45	0.26*	26.44*	
		Funen, Denmark	24.32	0.60*	60.05*	
		Gullmar Fjord, Sweden		0.35	35*	2.79
		Ria Formosa, Portugal		0.10	10*	0.61
Dahl et al. 2016	<i>Z. marina</i>	Asko, Sweden		0.05	5*	0.18
		Sozopol, Bulgaria		0.05	5*	0.17

\* Calculated value from published results

\*\* Results in total carbon

**Table 2. Results of statistical testing (two-way ANOVA) for significant effects of Active/Inactive Delta and Vegetated/Unvegetated on the total carbon stock in the top 50 cm of sediment. Total carbon was not significantly different between sites located in an active/inactive delta or between sites located in vegetated/unvegetated areas.**

ANOVA

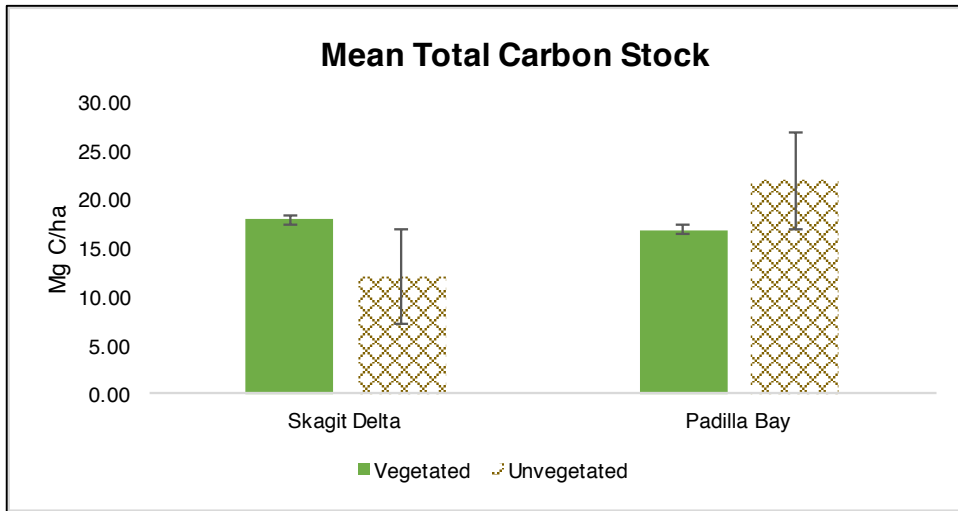
<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Active/Inactive Delta	57.94	1	57.94	1.68	0.23	5.32
Vegetated/Unvegetated	0.48	1	0.49	0.01	0.91	5.32
Interaction	88.85	1	88.85	2.58	0.15	5.32
Within	275.46	8	34.43			
Total	422.73	11				

**Table 3. Results of statistical testing (two-way ANOVA) for significant effects of Active/Inactive Delta and Vegetated/Unvegetated on mean  $\delta^{13}\text{C}$ . Carbon isotopes were significantly different between sites located in an active/inactive delta and between sites located in vegetated/unvegetated areas.**

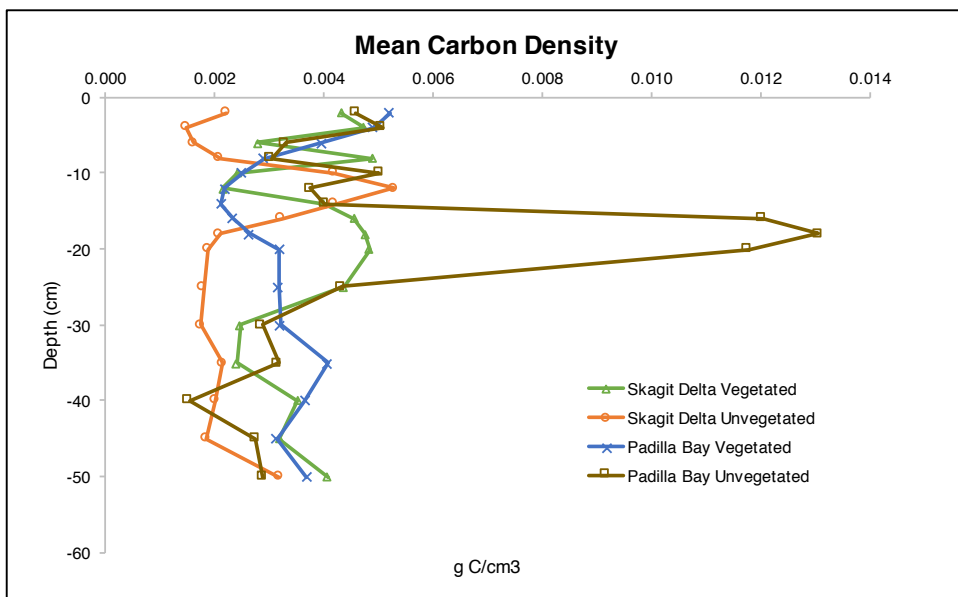
ANOVA						
<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Active/Inactive Delta	45.94	1	45.94	88.41	<b>1.34E-05</b>	5.32
Vegetated/Unvegetated	20.41	1	20.41	39.28	<b>2.41E-04</b>	5.32
Interaction	29.03	1	29.03	55.87	<b>7.10E-05</b>	5.32
Within	4.16	8	0.52			
Total	99.55	11				

**Table 4. Results of statistical testing (two-way ANOVA) for significant effects of Active/Inactive Delta and Vegetated/Unvegetated on mean  $\delta^{15}\text{N}$ . Nitrogen isotopes were significantly different between sites located in an active/inactive delta and between sites located in vegetated/unvegetated areas.**

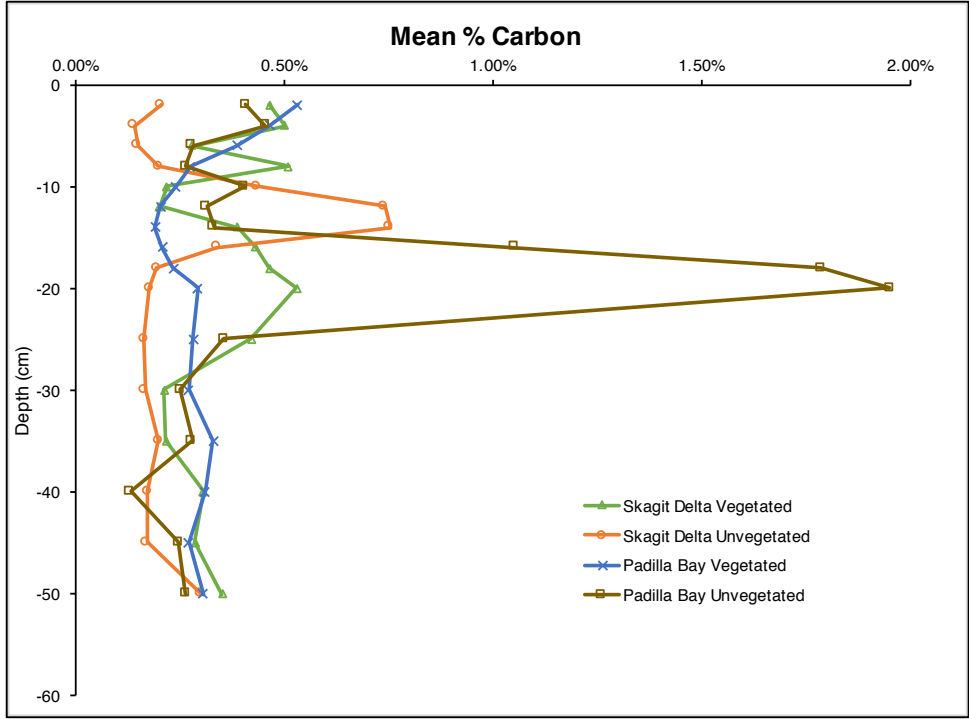
ANOVA						
<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Active/Inactive Delta	8.31	1	8.31	125.06	<b>3.66E-06</b>	5.32
Vegetated/Unvegetated	8.85	1	8.85	133.15	<b>2.89E-06</b>	5.32
Interaction	4.89	1	4.89	73.58	<b>2.634E-5</b>	5.32
Within	0.53	8	0.07			
Total	22.58	11				



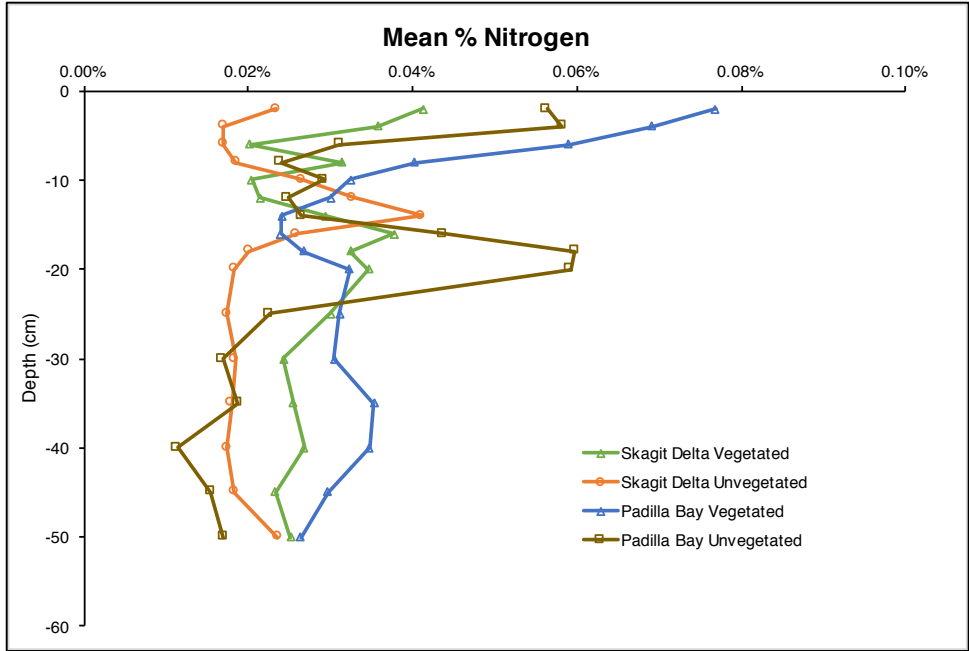
**Figure 1: Mean total carbon stock of the top 50 cm of sediment (Mg C/ha) for Skagit Delta and Padilla Bay by habitat type**



**Figure 2: Mean carbon density (g/cm<sup>3</sup>) with depth by site and habitat type**



**Figure 3: Mean percent carbon content with depth by site and habitat type**



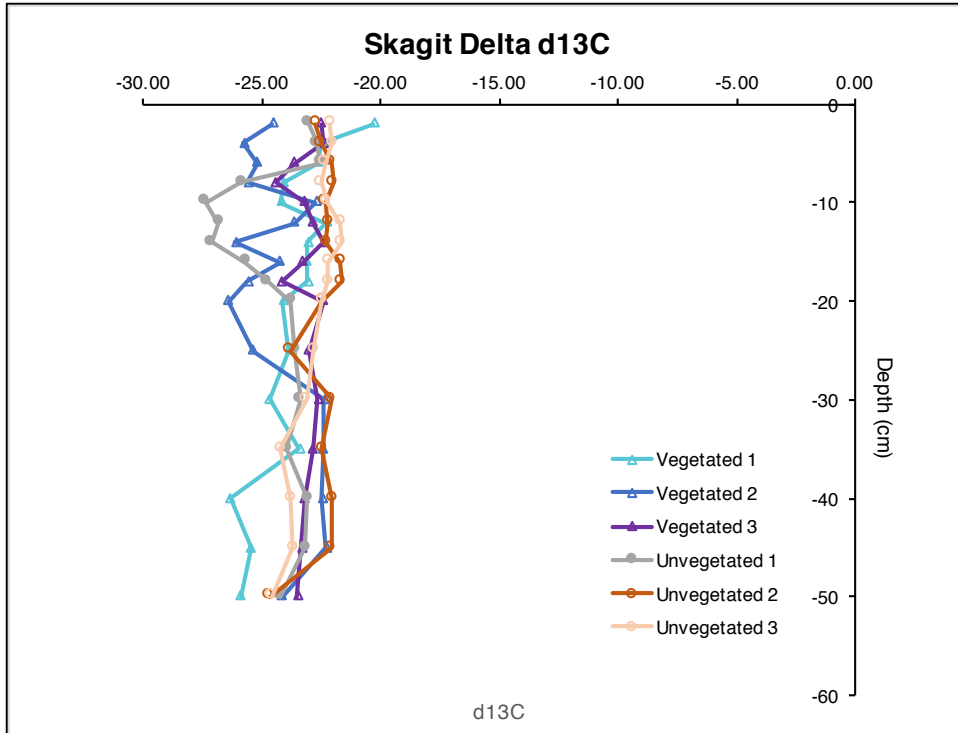
**Figure 4: Mean percent nitrogen content with depth by site and habitat type**

### Carbon and Nitrogen Isotopic Ratios

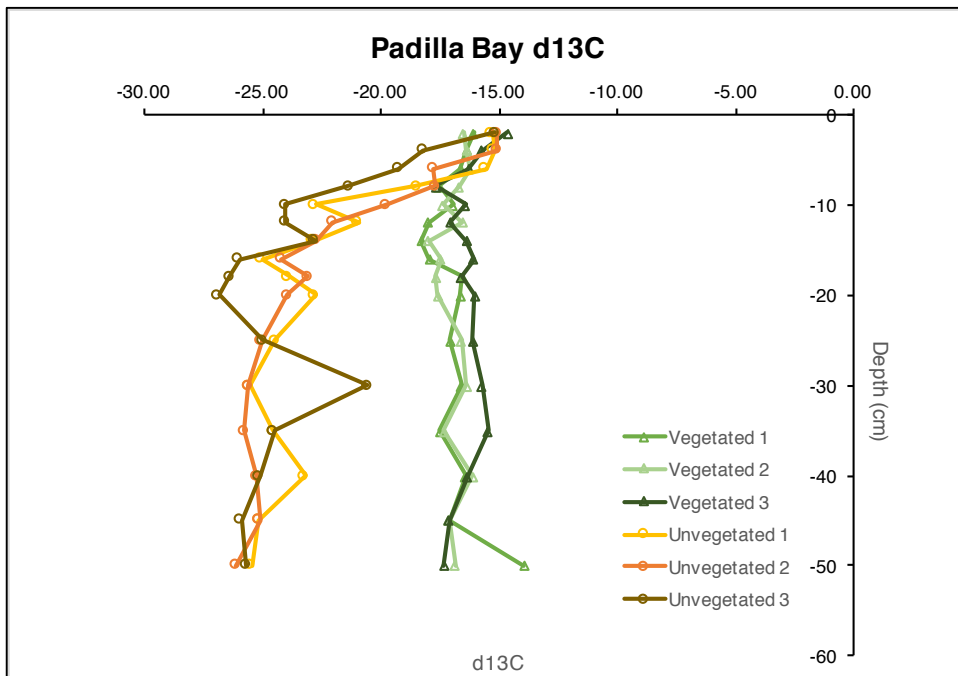
Two-way ANOVA confirmed hypothesis c and revealed isotopic ratios of (Table 3 & 4) carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) to be significantly different between the two sites (carbon;  $F_1 = 88.4$ ,  $P < .05$ ; nitrogen;  $F_1 = 125$ ,  $P < .05$ ) and between habitat type (carbon;  $F_1 = 39.3$ ,  $P < .05$ ; nitrogen;  $F_1 = 133.2$ ,  $P < .05$ ). However, whether vegetated cores were more depleted or enriched in  $^{13}\text{C}$  than their unvegetated counterparts depended on site. In the Skagit Delta, cores taken from vegetated habitat were depleted in  $\delta^{13}\text{C}$  ( $-23.73 \pm 0.16$ ) compared with cores taken from unvegetated habitat ( $-17.42 \pm 0.11$ ). Conversely, in Padilla Bay, cores taken from vegetated habitat were enriched in  $\delta^{13}\text{C}$  ( $-16.70 \pm 0.13$ ) compared with cores taken from unvegetated habitat ( $-22.42 \pm 0.87$ ). These site-specific differences in isotopic carbon and nitrogen could reflect differences in the source(s) of carbon and nitrogen supplied to each site.

In the Skagit Delta, there was little variation in  $\delta^{13}\text{C}$  with depth throughout all cores regardless of habitat type (Figure 5). However, in Padilla Bay,  $\delta^{13}\text{C}$  from vegetated and unvegetated areas diverge with distance from the surface (Figure 6).

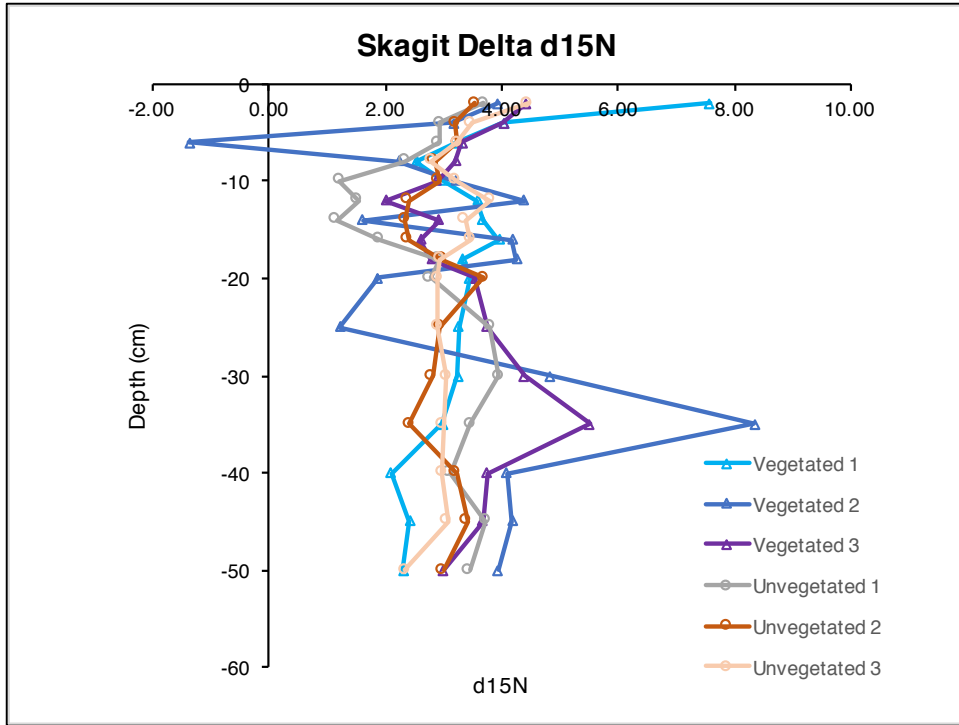
Linear mixed effects models revealed that percent fine grained sediment was the best predictor of  $\delta^{13}\text{C}$  content ( $X^2(1) = 20.37$ ,  $P < 6.38\text{e-}06$ ), followed by location in the core (that is, whether the sample was from the top 20 cm of the core), and if the core was taken from a vegetated plot or if it was taken from an active delta. The best predictor for  $\delta^{15}\text{N}$  content was whether the sample was from the top 20 cm of the core ( $X^2(1) = 18.02$ ,  $P < 2.184\text{e-}05$ ), followed by whether the sample was from an active delta, or if it was from a vegetated plot. Percent fine grained sediment was not a good predictor of  $\delta^{15}\text{N}$  content.



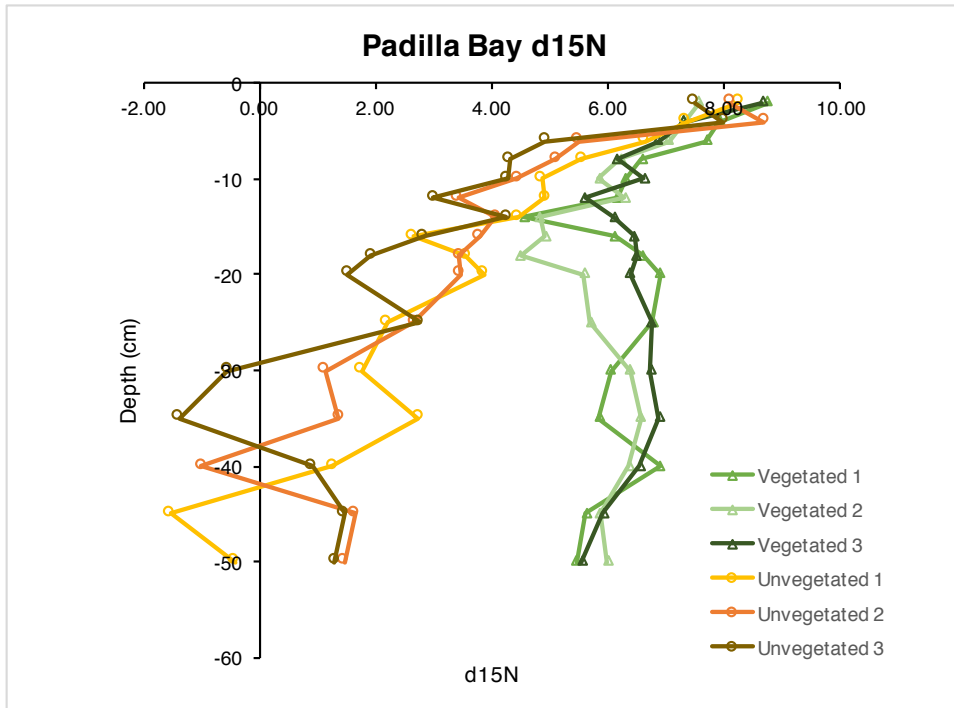
**Figure 5: Skagit Delta  $\delta^{13}\text{C}$  with depth by habitat type**



**Figure 6: Padilla Bay  $\delta^{13}\text{C}$  with depth by habitat type**



**Figure 7: Skagit Delta  $\delta^{15}\text{N}$  with depth by habitat type**



**Figure 8: Padilla Bay  $\delta^{15}\text{N}$  with depth by habitat type**



## Discussion

I found that carbon density was not statistically different between unvegetated plots and vegetated plots at both of the sites I studied. This finding is inconsistent with my hypotheses and the view, widely reported in the literature, that habitats vegetated by seagrass or other marine angiosperms store carbon at higher densities than those that are not vegetated. Moreover, I found that this was robust to depositional status: that is, carbon density did not differ between a highly depositional active delta and inactive delta. Notably, carbon densities at both sites were low compared with global averages. These findings provide evidence that the linkages between seagrass habitat and carbon storage reported from the Mediterranean Sea and from tropical seagrass habitats are not generalizable to the Salish Sea.

I did find significant differences in stable carbon and nitrogen isotopic signatures between each site and habitat type. This could indicate that the carbon and nitrogen stored within the top 50 cm of sediment comes from different sources and is influenced by deltaic processes and presence or absence of vegetation. Finer analysis of the origin of isotopic signatures of carbon and nitrogen is required to determine the source of the carbon and nitrogen stored in these sites.

Among my samples, the mean carbon densities integrated over the top 25 cm of sediment for both sites were low compared with those reported in the literature (Table 1). Moreover, total carbon stock of the top 50 cm for all sites, when doubled to estimate carbon stock over 1 meter in depth, is considerably lower than global estimates of from mixed species seagrass meadows (165.6 Mg C ha<sup>-1</sup>; Fourqurean et al., 2012).

### Potential Drivers of Carbon Storage in the Salish Sea

These findings suggest that physical forces, not seagrasses, could be the primary determinant of carbon deposition and storage at my study sites. Previous studies in Padilla Bay (Poppe, 2015; Kairis and Rybcyzk, 2010) have highlighted threats to the bay's seagrass meadow due to the loss of its primary sediment source and sea level rise. Padilla Bay experiences net erosion each year, which may transport stored carbon away from the bay. This may be the primary reason I found low carbon content in cores taken within this inactive delta.

Based on previous sediment deposition studies in the Skagit Delta (Webster et al., 2013), a potential explanation for a lack of high carbon storage at this active deltaic site could be that physical processes scour fine sediment off the tidal flats into deeper water where *Z. marina* does not grow. This could mean given the abiotic and biotic variables contributing to carbon storage in the Salish Sea, the presence of *Z. marina* may not be effective at trapping carbon at the site of these meadows. The importance of seagrass structural complexity and its role in trapping carbon in these habitats has been described in previous studies (Samper-Villarreal et al., 2016; Hendirks et al., 2008). Samper-Villarreal et al., (2016) found that carbon content was higher with higher plant structural complexity and low turbidity but not when turbidity increased. Therefore, *Z. marina*'s limited structural complexity, when compared with other seagrass species, may be less effective at decreasing water flow rates and promoting deposition or protecting sediments from erosion (Samper-Villarreal et al., 2016; Bos et al., 2007; Fonseca and Fisher, 1986). The relatively small rhizomatous mat characteristic of *Z. marina* may not withstand the physical processes typical of the Skagit River delta. Therefore, throughout Padilla Bay and the Skagit

River delta, physical forces could work against carbon burial in seagrass meadows. This is consistent with studies from mangrove and saltmarsh habitat that have found linkages between carbon stocks and hydro-geomorphic setting (Samper-Villarreal et al., 2016; Breithaupt et al., 2012; Donato et al., 2011). The particulate carbon and seagrass detritus produced within the sites I studied may be exported to deeper areas and stored there.

### Implications for Climate Mitigation and Resource Management

Despite the uncertainty surrounding the abiotic factors that affect variability of carbon stored in the Salish Sea, it is still possible to suggest management action based on carbon stock results. My findings that unvegetated bare mudflat does not differ in carbon content from vegetated *Z. marina* habitat may provide opportunities for resource management of the seafloor.

In May of 2017, California's Green House Gas cap and trade auction settlement price for allowances of emissions per metric ton reached \$13.80 USD (California Environmental Protection Agency, 2017). Using this value and the mean carbon stock of all sites within the top 50 cm (17.5 Mg C ha<sup>-1</sup> based on my data) equates to 64 metric tons of carbon dioxide per hectare. The amount of carbon dioxide potentially stored in my study sites is equivalent to 13.5 passenger vehicles driven for one year (Environmental Protection Agency, 2017). At the current market price, the value of carbon stored in my study sites would equal \$885 per hectare or \$229,214 per square mile. This value could influence policy or project planning cost-benefit analyses regarding projects that propose to disturb the seafloor at these sites.

However, incorporating this value for seafloor resource management is not the same as incorporating these findings into climate mitigation plans and carbon offset frameworks. As described by Sutton et al., (2016), projects that plan to enroll for carbon credits must show quantifiable added value to carbon emission reductions or additionality. If carbon storage is not tied to a biological indicator, such as the presence of *Z. marina*, it becomes difficult to demonstrate that conservation or mitigation efforts will add value by reducing carbon emissions instead of supporting an already functioning carbon storage system. However, understanding the value of these ecosystems, be it unvegetated mudflat, in terms of carbon stored currently may affect climate mitigation plans to prevent the disturbance and potential release of carbon stored in the seafloor.

Future research should continue to quantify carbon stock in the Salish Sea within multiple habitat types and depositional areas. These carbon stock assessments should also incorporate radiometric dating to determine accumulation rates of carbon. In addition, future studies should aim to identify the sources of carbon stored in Salish Sea habitats in order to narrow resource management and climate mitigation actions to continue to support carbon burial at these sites.

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## Appendix

### **Appendix A:** University of California Davis Stable Isotope Facility Methods (Stable Isotope Facility, 2017):

PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20-20 isotope ratio mass spectrometer (Sercon Ltd., Cheshire, UK). Samples are combusted at 1000°C in a reactor packed with chromium oxide and silvered copper oxide. Following combustion, oxides are removed in a reduction reactor (reduced copper at 650°C). The helium carrier then flows through a water trap (magnesium perchlorate) and an optional CO<sub>2</sub> trap (for N-only analyses). N<sub>2</sub> and CO<sub>2</sub> are separated on a Carbosieve GC column (65°C, 65 mL/min) before entering the IRMS.... During analysis, samples are interspersed with several replicates of at least two different laboratory standards. These laboratory standards, which are selected to be compositionally similar to the samples being analyzed, have been previously calibrated against NIST Standard Reference Materials (IAEA-N1, IAEA-N2, IAEA-N3, USGS-40, and USGS-41). A sample's preliminary isotope ratio is measured relative to reference gases analyzed with each sample. These preliminary values are finalized by correcting the values for the entire batch based on the known values of the included laboratory standards. The long term standard deviation is 0.2 permil for <sup>13</sup>C and 0.3 permil for <sup>15</sup>N.