

Comparison of Three Methods for the Quantification of Sediment Organic Carbon in Salt Marshes of the Rubicon Estuary, Tasmania, Australia

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Abstract

With the increasing need to accurately quantify carbon sinks of coastal wetlands, this study compares three methods in use. Sediment cores ($n = 4$) were collected from three salt marsh sites of the Rubicon Estuary, Tasmania Australia and analysed for organic carbon using three methods for measuring soil C concentration. The first method of elemental analysis quantified carbon using a Thermo Finnigan EA 1112 Series Flash Elemental Analyser. The second method used the loss on ignition technique at 550°C for 4 hours and applied a standard carbon conversion. The third method used loss on ignition technique at 450°C for 8 hours and applied a standard carbon conversion. Results from each method found significantly different quantities of carbon in replicate samples, however each method differentiated similar carbon trends within datasets. Each methodology investigated was found to have a useful application in carbon science, including the broad scale use of the standard carbon conversion and the accuracy of the elemental analysis method. The application of the standard carbon conversion in this study showed an underestimate of carbon stores in salt marsh sediment. Wetlands are well known to be among the most carbon rich ecosystems of the world, however this study highlights that in some cases those carbon stores may have been underestimated, and verification may further increase the value of wetlands as carbon sinks.

Keywords: salt marsh, soil organic carbon, dry combustion, loss on ignition

1. Introduction

The largest component of organic carbon found in wetland systems is in the sediment (Chmura, Anisfeld, Cahoon & Lynch, 2003; Bouillon et al., 2008; Donato et al., 2011), where organic carbon accumulates from the dead matter of leaves, stems and root matter of wetland vegetation during sediment accretion (Adam, 1990; Craft, 2007). The ability of wetlands to store carbon has provided an important conservation value in a period when the vulnerability of these unique ecosystems is increasing due to degradation, reclamation, erosion and sea level rise (Chmura et al., 2003; Howe, Rodriguez, & Saco, 2009; Elsey-Quirk, Seliskar, Sommerfield, & Gallagher, 2011; Donato et al., 2011).

A common method used to quantify carbon stores in sediment is a conversion variable, used to convert organic matter content data into organic carbon values (Allen, Grimshaw, Parkinson & Quarmby, 1974; Craft et al., 1991; Chmura et al., 2003; Pribyl, 2010; Yu & Chmura, 2010; Drexler, 2011). The Loss on Ignition (LOI) method is a simple and relatively inexpensive method used to estimate organic matter, though the temperature and length of time for best estimation of organic matter loss varies (Mook & Hoskin, 1982; Pribyl, 2010). Heiri, Lotter, and Lemcke (2001) found that exposure time, sample size and ignition temperature are all factors that affect LOI results, while other researchers express concern regarding the loss of structural water in the sample, giving an over estimation of organic matter, particularly in clay soils (Mook & Hoskin, 1982; Connor et al., 2001; Schumacher, 2002; Pribyl, 2010).

It has been assumed that soil organic matter contains between 58-60% carbon and that a conversion from organic matter to organic carbon can be achieved using either divisors or factors (Allen et al., 1974; Connor et al., 2001; Chmura et al., 2003). This is a simple procedure requiring the use of a conversion variable, conventionally a

divisor of 1.72, to interpret soil organic carbon from organic matter content, though this has been recently challenged (Pribyl, 2010).

Pribyl (2010) reviewed conversion variables in use back to the 1820's used to convert soil organic matter to organic carbon, finding that the divisor of 1.72 is generally too low, and that a divisor of 2 would be more accurate. Organic carbon content is however influenced by vegetation cover, organic matter composition, depth in profile as well as the amount of organic matter and clay in situ, suggesting that any carbon conversion variable cannot be a physical universal constant (Pribyl, 2010; Schumacher, 2002).

Conversion variables have been developed for different wetland ecosystems to allow relative comparisons of systems from data readily available, such as LOI. For seagrass meadows, Fourqurean et al. (2012) determined a positive relationship between organic matter and organic carbon from a broad variety of seagrass habitats, using studies that reported LOI results. For mangrove wetlands, carbon stores are estimated using conversion factors and used to give global mangrove stored carbon estimates and rates of carbon burial (Cintr'ón, Lugo, Pool, & Morris, 1978; Chmura et al., 2003; Twilley, Chen & Hargis, 1992; Breithaupt, Smoak, Smith III, Sanders, & Hoare, 2012).

Conversion factors developed for estuarine marsh soils have been calibrated by dry combustion (Morris & Whiting, (1986) and Craft et al. (1991), and range between 0.2 and 0.6. The range of divisors and factors in use suggests that a single conversion to organic carbon from LOI results may yield erroneous estimates of organic carbon (Pribyl, 2010).

Craft et al. (1991) developed an equation specifically for estuarine marshes to transform organic matter data into organic carbon data based on the proportional increase of organic carbon with organic matter. This was similar to previous methods for the development of conversion variables, however improving the accuracy of the equation through replicated sampling, calibration with elemental analysis, and testing on 250 samples sourced from 10 estuarine marshes in North Carolina. The equation produced is in quadratic form:

$$\text{Organic C} = (0.4 \pm 0.01) \text{ LOI} + (0.0025 \pm 0.0003) \text{ LOI}^2 \quad (1)$$

However the application of the equation globally could be questionable given the local variability found, along with the reliance on LOI as a measure of organic matter (Connor et al., 2001; Chmura et al., 2003; Howe et al., 2009; Donato et al., 2011).

The elemental analysis procedure first became popular during the 1940's and 50's in the oil industry (Down & Himus, 1941; Forsman & Hunt, 1958). Since then, technological advances have seen the application and accessibility of the analysis markedly improve and diversify. Using elemental analysis as a procedure for the determination of organic carbon is highly accurate (Pribyl, 2010; Schumacher, 2002), however while the technology remains expensive and requires specialised equipment which is often only available in limited laboratories, the costs have been decreasing. This accurate method allows assessment of the merits of other methods more widely used for previous salt marsh soil carbon evaluations.

This research has the objective of comparing and contrasting three methodologies of evaluating salt marsh soil organic carbon. The three methodologies evaluated are:

- 1) Elemental analysis of percent organic carbon,
- 2) The LOI method outlined by Heiri et al. (2001) and applying the quadratic equation from Craft et al. (1991),
- 3) The LOI method outlined by Craft et al. (1991) and applying the quadratic equation from Craft et al. (1991).

These were tested for native salt marsh sites and salt marsh sites dominated with *Spartina anglica* in the Rubicon estuary, Tasmania.

2. Method

2.1 Study Sites

Tasmania extends across a latitudinal range of 39° 40'-43° 20' S and is the southernmost state in Australia. As the only island state, Tasmania has longer coastlines per unit land area of c. 2,200 km, and a salt marsh area of 47 km² (Zann, 1995). Tidal ranges are highest on the northern coast, with an estimated range at the Rubicon of 2.1 m (Edgar, Barrett, & Graddon, 1999). The Rubicon Estuary on the central north coast of Tasmania (41°09'S 146°33'E; Figure 1) has a total catchment area of 262 km² and an overall low topographic relief (Edgar, Barrett, & Graddon, 1999). The mean annual temperature is 16°C with a mean monthly range of 13-22°C, and the mean precipitation is 648 mm a⁻¹ with all mean monthly totals over 30 mm and a winter maximum (Bureau of Meteorology 2011). The climatic conditions within the greater Rubicon estuary area have been historically favourable to land use changes such as agriculture and forestry that has resulted in a high sediment supply

(Cotching, 1995; Sims & Cotching, 2000).

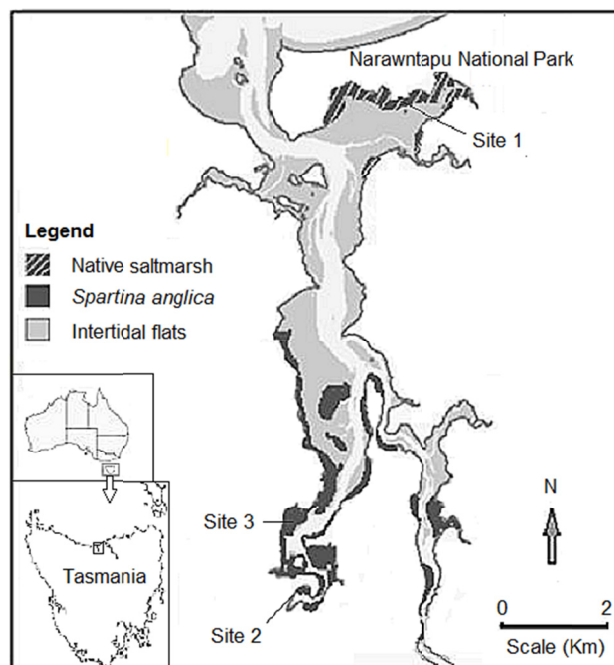


Figure 1. Location of the Rubicon Estuary, Tasmania showing intertidal zones and sample sites

Site 1 was located in native salt marsh within the Narawntapu National Park (Figure 1), which has been managed and maintained as a rice grass free zone (DPIWE, 2002). Site 2 was located in the upper estuary, in dense pioneer cover of introduced rice grass *S. anglica* vegetation with no native salt marsh species present. Site 3 was located on the western margin of the Rubicon estuary in dense well-established *S. anglica* vegetation (Figure 1).

2.2 Sample Collection and Analysis

Two cores were collected at site 1, one from *Sarcocornia quinqueflora* vegetation and the second from *Juncus kraussii* vegetation and one core was collected from sites 2 and 3 during the summer of 2011. A Russian peat corer was used to extract sediment 8.5 cm in diameter to a depth of approximately 0.6 m. Compaction was negligible owing to the sidewall sample design of the corer. The cores were placed in a PVC pipe and wrapped securely for transportation to the analytical laboratory, and were carried horizontally at all times to minimize vertical compaction.

In the laboratory cores were opened in a horizontal position and sampled at 5 cm increments from the surface at 0 cm down each core. Sediment was sampled from ± 0.5 cm at each increment. Samples from each increment were mixed and then divided by volume into 10 ml replicates to be tested by each methodology as described in the below section.

2.3 Percent Organic Carbon Determination

2.3.1 Method 1 - Elemental analysis of percent organic carbon

To identify carbon content in organic matter, samples from each core were prepared and analysed in a Thermo Finnigan EA 1112 Series Flash Elemental Analyser after being weighed in a Sartorius Microbalance SE2 with a precision of 0.1 μ g. A pre-treatment of HCl was required for the removal of inorganic carbons from the samples prior to the elemental analysis. Following the method described by Craft et al. (1991), samples were immersed with 6N HCl to test for carbonates. Treatment was applied to subsamples and not to the entire sample, as HCl may dissolve part of the organic matter present within the sample, leading to an underestimation of the organic matter content (Schumacher, 2002). Therefore unnecessary exposure to HCl was avoided. Where effervescence was present, samples were immersed in 6N HCl for 10 minutes then rinsed 5 times with distilled water to remove remnant traces of HCl.

The elemental analyser measured the amount of carbon, hydrogen and nitrogen in the material by the combustion of small amounts (1-2 mg) of the sediment sample in pure oxygen and in the presence of catalysts at high

temperature. The combustion products were separated by passing them through a packed column and quantified using a thermal conductivity detector.

2.3.2 Method 2 - LOI method from Heiri et al. (2001) applying the quadratic equation from Craft et al. (1991)

Following the methodology described by Heiri et al. (2001), samples were ground into a fine powder using a mortar and pestle, and remnant vegetation fragments were removed by passing through a 425 μm sieve after oven drying at 60°C for 72 hours until a constant dry weight was obtained. Samples were then placed in a muffle furnace at 550°C for 4 hours. The LOI results were used in the equation developed by Craft et al. (1991) resulting in percent carbon data.

2.3.3 Method 3 - LOI method from Craft et al. (1991) applying the quadratic equation from Craft et al. (1991)

A HCl treatment was applied to the samples following the method described by Craft et al. (1991), and outlined above in method 1.

Samples were then weighed and oven dried at 60°C for 72 hours until a constant dry weight was obtained. Once dry, samples were ground into a fine powder using a mortar and pestle, and remnant vegetation fragments were removed by passing through a 425 μm sieve. Samples were placed in a muffle furnace at 450°C for 8 hours. The LOI results were used in the equation developed by Craft et al. (1991) resulting in percent carbon data.

2.4 Statistical Analysis

Kruskal-Wallis one-way ANOVA was used in the statistics program XLSTAT, where using statistical analysis that compares ranks rather than means will also result in the output being less affected by outliers in the dataset (Kruskal & Wallis, 1952). The hypothesis was tested “That the 3 different laboratory procedures used to calculate below ground carbon will yield the same carbon results (the distribution of carbon is the same across categories of method)”.

The Mann Whitney U test and the Conover-Iman procedure / Two-tailed test were conducted to evaluate pairwise differences among the three groups to determine where the differences were within the populations. Due to the possible inflation of a Type I error occurring during multiple hypotheses testing a Bonferroni correction was used; whereby a greater level of significance is required in order for the results to be deemed significant. The Bonferroni correction divides the pre-specified α -level equally among the tests of significance (j) being contemplated.

3. Results

3.1 Method 1

Organic carbon content results using the dry combustion method showed a relationship of decreasing carbon with depth at each site (Figure 2). The rice grass sites showed overall higher levels of carbon content than the native salt marsh site, apart from a surface outlier at site 1.1 (*J. kraussii*). Overall carbon levels found were 0.3-2.4% at 50 cm depth, 0.2-2.2% at 30 cm depth, 1.8-4.0% at 10 cm depth and 1.4-7.1% at the surface, data is provided in Table 1.

Table 1. Method 1 percentage carbon results

Depth (cm)	% C			
	Site 2	Site 3	Site 1.1	Site 1.2
5	4.18	3.45	7.16	1.47
10	3.99	3.08	1.82	2.44
15	3.13	3.14	1.07	1.70
20	1.68	3.26	0.98	0.66
25	1.04	2.07	1.19	0.41
30	0.86	2.21	0.43	0.19
35	0.94	2.15	0.17	0.17
40	1.29	2.35	0.19	0.23
45	1.09	1.82	0.29	0.22
50	1.31	2.44	0.34	0.30

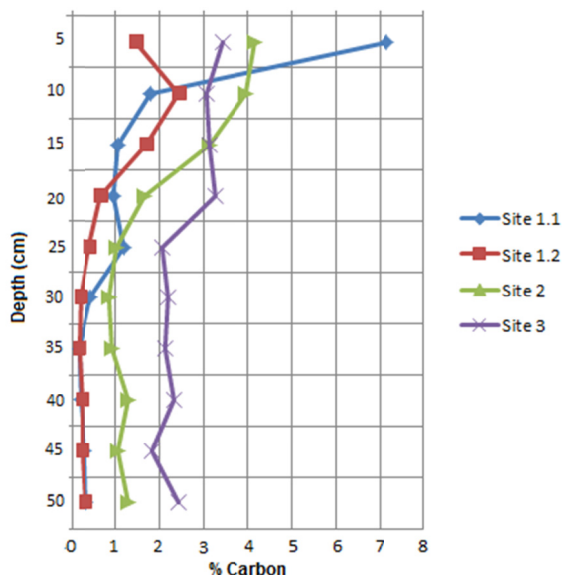


Figure 2. Relationship between percentage carbon with depth from each core using method 1- Elemental analyser

3.2 Method 2

Organic matter content results using the LOI technique of 4 hours at 550°C are shown in Figure 3, with a trend of decreasing organic matter with depth in cores found at all sites. The rice grass sites showed overall higher levels of carbon content than the native salt marsh site, apart from surface outliers at both native sites. Overall organic matter levels found were 0.7-1.3% at 50 cm depth, 0.5-1.8% at 30 cm depth, 1.5-2.3% at 10 cm depth and 1.3-3.4% at the surface. Site 3 and site 1.2 had the most even distribution of organic matter that decreased by less than 1% down the 48 cm core. Site 1.1 had a high surface organic content (3%) however low mean organic content. Site 1.2 had the lowest mean organic content, while the LOI results from sites 2 and 3 were within 0.1% of each other.

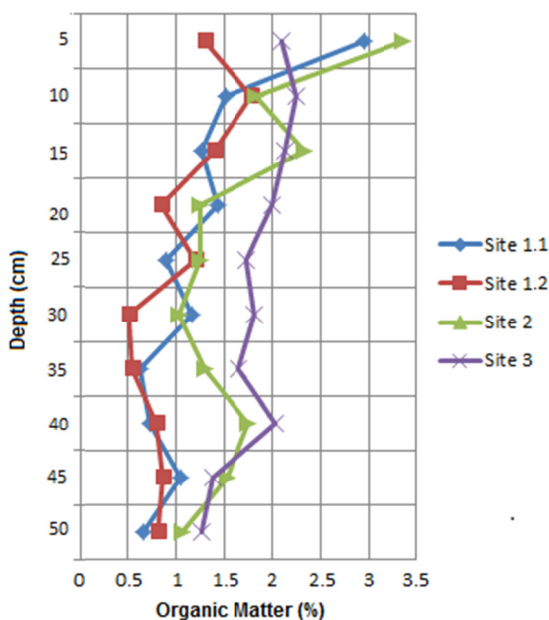


Figure 3. Organic matter content (%) with depth from each core using method 2: Heiri et al. (2001) LOI method of LOI at 550°C for 4 hours

Percentage carbon results found using the LOI 4 hour method and applying the Craft et al. (1991) carbon calculation equation are shown in Figure 4, and data provided in Table 2. Results showed a relationship of decreasing carbon with depth at each site, and overall carbon levels found were 0.27-0.51% at 50 cm depth, 0.47-0.73% at 30 cm depth, 0.61-0.92% at 10 cm depth and 1.37-0.53% at the surface. The rice grass sites 2 and 3 again showed the most carbon stored throughout the core; however the overall percentages were half of the amounts quantified using the dry combustion method.

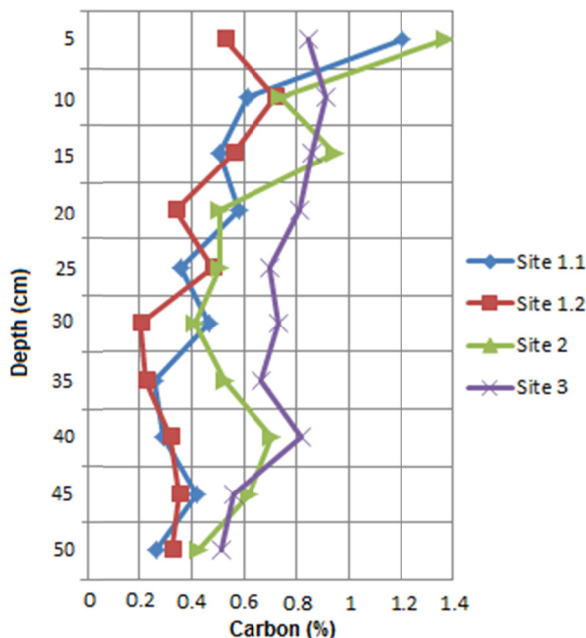


Figure 4. Percentage carbon with depth in each core using method 2: LOI 4 hour method and applying the quadratic equation developed by Craft et al. (1991) converting LOI to SOC

Table 2. Method 2 percentage organic matter (OM) and percentage carbon results

Depth (cm)	% C				% OM			
	Site 2	Site 3	Site 1.1	Site 1.2	Site 2	Site 3	Site 1.1	Site 1.2
5	1.37	0.85	1.21	0.53	2.96	1.30	3.36	2.10
10	0.74	0.92	0.61	0.72	1.52	1.78	1.84	2.26
15	0.95	0.86	0.51	0.56	1.26	1.40	2.33	2.13
20	0.51	0.81	0.58	0.34	1.43	0.85	1.26	2.01
25	0.51	0.70	0.36	0.48	0.90	1.20	1.25	1.74
30	0.42	0.74	0.47	0.21	1.16	0.52	1.04	1.82
35	0.53	0.67	0.26	0.23	0.64	0.56	1.30	1.65
40	0.71	0.82	0.30	0.32	0.74	0.80	1.76	2.03
45	0.62	0.56	0.42	0.35	1.05	0.88	1.54	1.39
50	0.43	0.51	0.27	0.33	0.67	0.82	1.07	1.27

3.3 Method 3

Results of organic matter content using the LOI technique of 8 hours at 450°C are shown in Figure 5, and data provided in Table 3. There is a clear trend of decreasing organic matter with depth for the native marsh sites 1.1 and 1.2, while introduced *S. anglica* sites 2 and site 3 both show a similar trend but with more variation. Overall organic matter levels found were 0.3-0.8% at 50 cm depth, 0.2-0.7% at 30 cm depth, 0.7-1.4% at 10 cm depth

and 0.7-1.6% at the surface. The lowest mean organic matter content was at site 1.2 while the highest mean organic matter content was at site 3 (Table 4).

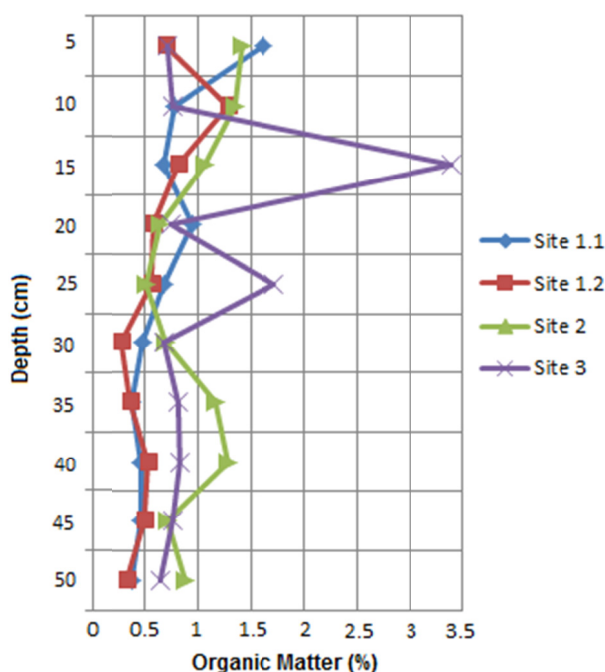


Figure 5. Organic matter content (%) with depth using method 3: LOI at 450°C for 8 hours

Table 3. Method 3 percentage organic matter and percentage carbon results

Depth (cm)	% C				% OM			
	Site 2	Site 3	Site 1.1	Site 1.2	Site 2	Site 3	Site 1.1	Site 1.2
5	0.55	0.34	0.49	0.21	1.62	0.71	1.42	0.71
10	0.30	0.37	0.25	0.29	0.79	1.28	1.34	0.78
15	0.38	0.35	0.20	0.23	0.69	0.82	1.07	3.41
20	0.20	0.33	0.23	0.14	0.95	0.59	0.65	0.75
25	0.20	0.28	0.14	0.19	0.68	0.57	0.53	1.73
30	0.17	0.30	0.19	0.08	0.48	0.28	0.70	0.69
35	0.21	0.27	0.10	0.09	0.38	0.37	1.16	0.82
40	0.29	0.33	0.12	0.13	0.47	0.54	1.29	0.84
45	0.25	0.23	0.17	0.14	0.46	0.50	0.71	0.77
50	0.17	0.21	0.11	0.13	0.38	0.34	0.88	0.65

Percentage carbon results found using the LOI 8 hour method and applying the equation of Craft et al. (1991) showed a relationship of decreasing carbon with depth (Figure 6). Overall carbon levels found were 0.13-0.36% at 50 cm depth, 0.19-0.28% at 30 cm depth, 0.31-0.54% at 10 cm depth and 0.28-0.65% at the surface. The decrease in carbon with depth at each site is not as clear in these results (Figure 6) as in the previous methods (Figures 2-5). Additionally the quantity of carbon is approximately a quarter less than the carbon quantified using the dry combustion method and less than half of the carbon quantified using the LOI 4 hour method (Table 4).

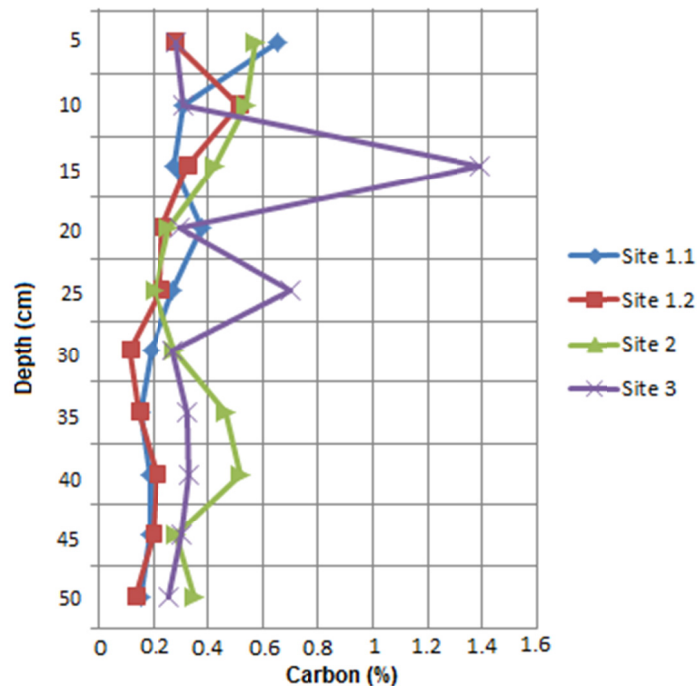


Figure 6. Relationship between percent carbon with depth in each core using the LOI 8 hour method and applying the quadratic equation developed by Craft et al. (1991) converting LOI to SOC

Table 4. Organic carbon content (%) mean \pm SD results from Rubicon estuary sites from the three methods

	Site 1.1	Site 1.2	Site 2	Site 3
Method 1	1.37 \pm 2.1	0.78 \pm 0.8	1.95 \pm 1.3	2.6 \pm 0.5
Method 2	0.5 \pm 0.3	0.41 \pm 0.2	0.68 \pm 0.3	0.74 \pm 0.1
Method 3	0.27 \pm 0.2	0.24 \pm 0.1	0.39 \pm 0.1	0.45 \pm 0.4

3.4 Statistical Analysis of the Organic Carbon Quantification Methodologies

A Kruskal-Wallis one-way ANOVA was performed on the percentage carbon results from the three methods. The analysis indicated a significant difference between the methods ($\chi^2(2) = 5.911$, $p < 0.0001$). Multiple pairwise comparisons using the Conover-Iman procedure / Two-tailed test were conducted to evaluate pairwise differences among the three groups, controlling for Type I error across tests by using a Bonferroni correction. The analysis indicated a significant difference between all groups ($p < 0.01$) shown in Figure 7.

Results from the Kruskal-Wallis one-way ANOVA performed on the percentage carbon results from the three groups of methods were categorized into communities (native and non-native). The analysis indicated a significant difference between communities within each of the three methods ($\chi^2(1) = 3.841$, $p < 0.0001$).

Results from the multiple pairwise comparisons using the Conover-Iman procedure / Two-tailed test conducted to evaluate pairwise differences among communities, controlling for Type I error across tests by using a Bonferroni correction are shown in Figure 8. The analyses indicated that the native community was significantly different to the non-native community in all of the methods (method 1 $p < 0.0001$, method 2 $p < 0.0001$, method 3 $p = 0.0001$).

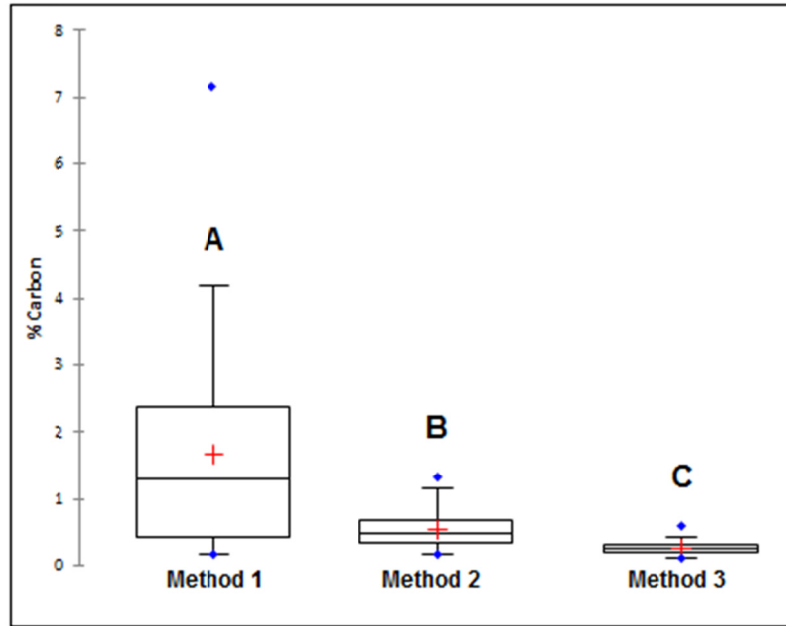


Figure 7. Results of percent carbon of the three methods of carbon quantification (method 1, method 2, and method 3) shown as box plots. Means with different letters are significantly different ($p < 0.001$)

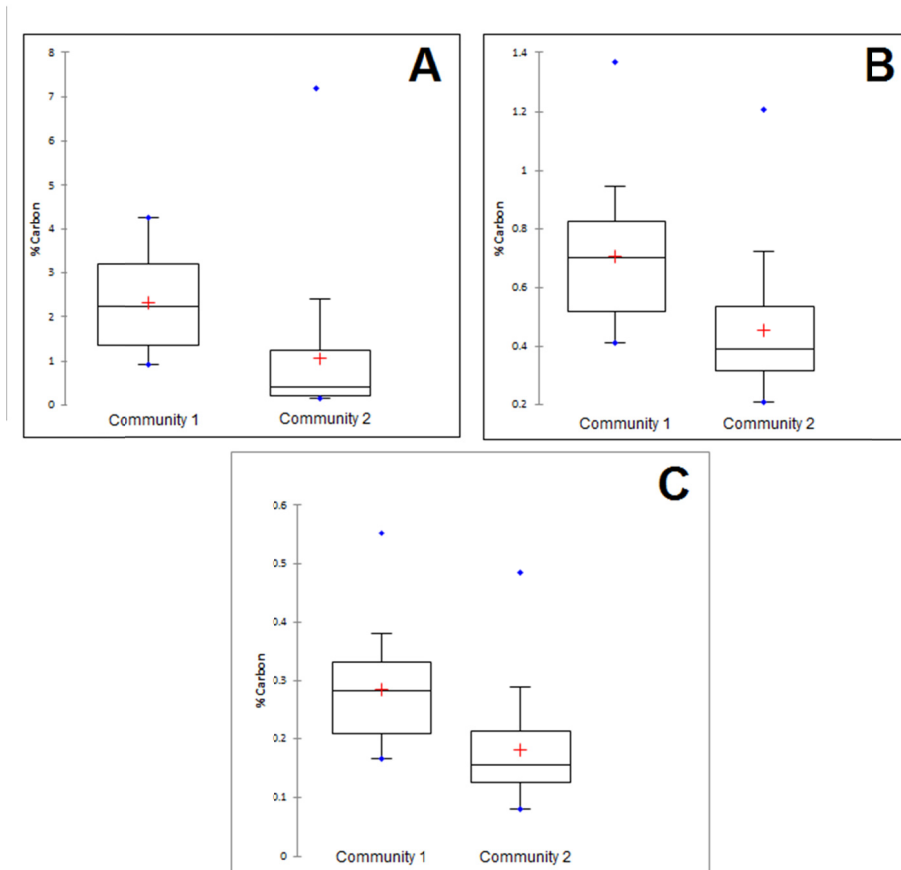


Figure 8. Results of percent carbon from each method (A – method 1, B – method 2, C – method 3) shown as box plots and categorized by community (community-1 – *S. anglica*, community-2 – native marsh). Means with different letters are significantly different ($p < 0.001$)

Mann-Whitney U Tests showed that method 2 and method 3 were significantly different in native sites ($p=0.0001$) and non-native sites ($p=0.0001$). Results from method 1 and method 2 were significantly different in both native sites ($p=0.0001$) and non-native sites ($p=0.0001$). Method 1 and method 3 were significantly different in both native sites ($p=0.0001$) and non-native sites ($p=0.0001$).

4. Discussion

Results from each method found a similar trend of decreasing carbon with depth within and between sites (Figures 2-6), however the results demonstrate the variability in carbon results from different methodologies (Table 4).

Method 1 using dry combustion by elemental analysis is accurate, precise and reliable (Pribyl, 2010) and results from soils of the Rubicon salt marshes (Figure 2) showed the organic carbon values to be consistently declining with depth in all cores, similar to organic carbon results in South Carolina estuarine marshes at 32° N (Sanger, Holland & Scott, 1999), and marsh sites located in West Greenland at 69°N (Jensen, Schmidt, Hollesen & Elberling, 2006).

By contrast, samples from method 2 and 3 were found to contain significantly less carbon, with less consistency in the proportional decline of carbon with depth.

Determining an organic carbon value using method 2 is a cheap and easy alternative without the reliance on specialised equipment (Pribyl, 2010). The LOI method is a standard technique for determining the organic matter of sediments, although the absolute precision of the method is often questioned (Dean, 1974; Mook & Hoskin, 1982; Bengtsson & Enell, 1986; Craft et al., 1991; Connor et al., 2001; Heiri et al., 2001; Pribyl, 2010). Average organic carbon results were found to be significantly lower than the dry combustion method 1 (Table 4), and showed less difference between sites and with depth (Figures 3 and 4).

Applying the quadratic equation of Craft et al. (1991) to LOI results using exposure times and temperatures outside of those specified by Craft et al. (1991), has been tested previously (Connor et al., 2001; Chmura et al., 2003; Yu & Chmura, 2010). The equation has allowed re-evaluation of previous studies reporting organic matter values, by allowing organic carbon percentages to be determined without further sample analysis (Chmura et al., 2003).

However the results from this study show organic carbon results from the LOI method to be lower than similar studies (Figure 4) (Sanger et al., 1999; Connor et al., 2001; Howe et al., 2009; Palomo & Neil, 2009) and far lower than results obtained from replicate samples analysed by the dry combustion method (Figure 2). This suggests that the reliance of LOI as the primary data source may affect the reliability of results determined using the modelled equation.

The method 3 organic carbon results (Figure 6) determined from using the method outlined by Craft et al. (1991) also gave lower results than the dry combustion method (Figure 2) as well as results found in similar studies (Sanger et al., 1999; Connor et al., 2001; Howe et al., 2009; Palomo & Neil, 2009). The results indicate that the low percentages found in organic carbon are due to the low organic matter results obtained from the LOI procedure (Figure 5). This method is also a cheap and easy method without the reliance on specialised equipment (Pribyl, 2010).

4.1 Comparison of the Methods

It was found that by varying the control factors within the LOI method, such as exposure time and ignition temperature, the results between methodologies were not comparable. Method 3 had a longer exposure time and lower ignition temperature which resulted in half the organic matter reported from method 2 (Figures 3 and 5). Findings from Allen et al. (1974), Heiri et al. (2001), Abella (2007) and Pribyl (2010) suggested that varying the ignition temperature in the LOI procedure will produce different results, and our results from salt marsh sediments concur with this finding.

Previous studies suggest that LOI estimates of organic matter will in general produce an overestimate of organic matter due to the loss of structural water, thus making results from clay soils particularly risky (Mook & Hoskin, 1982; Craft et al., 1991; Connor et al., 2001; Pribyl, 2010). The Craft et al. equation was developed for soils with low clay content (0-11%), and while that condition may not hold for our samples the results from this study however produced underestimates of organic matter using both methods. The underestimates were extenuated using method 3 likely to be as a consequence of the longer exposure time (Table 4). The underestimate was determined from the percentage carbon results given by the dry combustion method that showed an average of 1.6% carbon in the samples (Table 4), while the average organic matter from the cores using method 2 was 1.4% while method 3 was 0.8%. Both organic matter percentages were lower than the carbon percentage, when it

would be expected that the organic matter in the samples would be greater given that organic carbon is a component of the organic matter. This suggests that both LOI methods are reporting an underestimate of organic matter in the sediment.

The Kruskal-Wallis one-way ANOVA showed that the difference in methodologies were statistically significant (Figure 7). However further analysis using a Mann-Whitney U test showed that each method was able to independently recognize a significant difference between the communities of salt marsh (*S. anglica*) and native marsh (Figure 8). This shows that each methodology is able to identify trends within the data sets, but comparisons using different methods, should be made with caution.

5. Conclusions

Over the past 40 years, significant progress has occurred in advancing capability to quantify sediment carbon in a diverse range of ecosystems, enhancing understanding of this value (Allen et al., 1974; Heiri et al., 2001; Craft et al., 1991; Drexler, 2011). The objective of this study was to compare and contrast three common methodologies used to quantify carbon in salt marsh soil, and each was found to have a useful application in carbon science, including the broad scale use of the quadratic equation and the accuracy of the elemental analysis method. This study has shown that with recent advances in technological capability and reducing costs and logistical complexity, techniques previously underutilised or unavailable due to cost and logistical constraints can be more widely applied to the quantification of soil carbon.

The application of the quadratic equation in this study showed an underestimate of carbon stores in salt marsh sediment. This raises the question of underestimation in other studies that have relied on similar methods for carbon quantification. Wetlands are well known to be among the most carbon rich ecosystems of the world (Donato et al., 2011), however this study highlights that in some cases those carbon stores may have been underestimated, and verification may further increase the value of wetlands as carbon sinks.

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References

- Abella, S. R., & Zimmer, B. W. (2007). Estimating organic carbon from Loss-On-Ignition in Northern Arizona forest soils. *Soil Science Society of America Journal*, 71, 545-550. <http://dx.doi.org/10.2136/sssaj2006.0136>
- Adam, P. (1990). *Salt marsh ecology*. Cambridge: Cambridge University Press.
- Allen, S. E., Grimshaw, H. M., Parkinson, J. A., & Quarmby, C. (1974). *Chemical analysis of ecological materials*. New York: Wiley.
- Bengtsson, L., & Enell, M. (1986). Chemical analysis. In E. B. Berglund (Ed.), *Handbook of Holocene palaeoecology and palaeohydrology* (pp. 423-455). Caldwell, New Jersey: The Blackburn Press.
- Bouillon, S., Borges, A. V., Castaneda-Moya, E., Diele, K., Dittmar, T., Duke, N. C., ... Twilley, R. R. (2008). Mangrove production and carbon sinks: A revision of global budget estimates. *Global Biogeochemical Cycles*, 22, GB2013. <http://dx.doi.org/10.1029/2007GB003052>.
- Breithaupt, J. L., Smoak, J. M., Smith III, T. J., Sanders, C. J., & Hoare, A. (2012). Organic carbon burial rates in mangrove sediments: Strengthening the global budget. *Global Biogeochemical Cycles*, 26, GB3011. <http://dx.doi.org/10.1029/2012GB004375>.
- Bureau of Meteorology (2011). Climate statistics for Australian locations, Monthly climate statistics. Retrieved from http://www.bom.gov.au/climate/averages/tables/cw_091126.shtml.
- Cintr' on, G., Lugo, A. E., Pool, D. J., & Morris, G. (1978). Mangroves of arid environments in Puerto Rico and adjacent islands. *Biotropica*, 10, 110-121. <http://dx.doi.org/10.2307/2388013>
- Chmura, G. L., Anisfeld, S. C., Cahoon, D. R., & Lynch, J. C. (2003). Global carbon sequestration in tidal, saline wetland soils. *Global Biogeochemical Cycles*, 17(4), 1111. <http://dx.doi.org/10.1029/2002GB001917>, 2003.
- Connor, R. F., Chmura, G. L., & Beecher, C. B. (2001). Carbon accumulation in Bay of Fundy salt marshes: Implications for restoration of reclaimed marshes. *Global Biogeochemical Cycles*, 15, 943-954. <http://dx.doi.org/10.1029/2000GB001346>
- Cotching, W. E. (1995). A review of the challenges for long term management of krasnozems in Australia. *Australian Journal of Soil and Water Conservation*, 8, 18-27.

- Craft, C. B., Seneca, E. D. & Broome, S. W. (1991). Loss on ignition and Kjeldahl Digestion for estimating organic carbon and total Nitrogen in estuarine marsh Soils: Calibration with dry combustion. *Estuaries*, *14*, 175-179. <http://dx.doi.org/10.2307/1351691>
- Craft, C. (2007). Freshwater input structures soil properties, vertical accretion, and nutrient accumulation of Georgia and U.S. tidal marshes. *Limnology and Oceanography*, *52*, 1220-1230.
- Donato, D. C., Kauffman, J. B., Murdiyarso, D., Kurnianto, S., Stidham, M., & Kanninen, M. (2011). Mangroves among the most carbon-rich forests in the tropics. *Nature Geoscience*, *4*, 293-297. <http://dx.doi.org/10.1038/NCEO1123>
- Down, A. L., & Himus, G. W. (1941). A preliminary study of the chemical constitution of kerogen. *Journal of the Institute of Petroleum*, *27*, 436-445.
- DPIWE [Department of Primary Industries, Water and Environment] (2002). *Strategy for the Management of Rice Grass (Spartina anglica) in Tasmania, Australia* (2nd ed.). Hobart: Department of Primary Industries and Water.
- Dean, W. E. (1974). Determination of carbonate and organic matter in calcareous sediments and sedimentary rocks by loss on ignition: comparison with other methods. *Journal of Sedimentary Petrology*, *44*, 242-248.
- Drexler, J. Z. (2011). Peat formation processes through the millennia in tidal marshes of the Sacramento-San Joaquin Delta, California, USA. *Estuaries and Coasts*, *34*, 900-911. <http://dx.doi.org/10.1007/s12237-011-9393-7>
- Edgar, G. J., Barrett, N. S., & Graddon, D. J. (1999). *A classification of Tasmanian estuaries and assessment of their conservation significance using ecological and physical attributes, population and land use*. Hobart, Tasmania: Marine Research Laboratories, Tasmanian Aquaculture and Fisheries Institute.
- Elsy-Quirk, T., Seliskar, D. M., Sommerfield, C. K., & Gallagher, J. L. (2011). Salt marsh carbon pool distribution in a Mid-Atlantic lagoon, USA: Sea level rise implications. *Wetlands*, *31*, 87-99. <http://dx.doi.org/10.1007/s13157-010-0139-2>.
- Forsman, J. P., & Hunt, J. M. (1958). Insoluble organic matter (kerogen) in sedimentary rocks of marine origin In L.G. Weeks, (Ed.) *Habitat of oil* (pp. 747-778). Tulsa, Oklahoma: American Association Petroleum Geologists.
- Fourqurean, J. W., Duarte, C. M. Kennedy, H., Marba, N., Holmer, M., Mateo, M. A., ... Serrano, O. (2012). Seagrass ecosystems as a globally significant carbon stock. *Nature Geoscience*, *5*, 505-509. <http://dx.doi.org/10.1038/ngeo1477>
- Heiri, O., Lotter, A. F., & Lemcke, G. (2001). Loss on ignition as a method for estimating organic and carbonate content in sediments: reproducibility and comparability of results. *Journal of Paleolimnology*, *25*, 101-110. <http://dx.doi.org/10.1023/A:1008119611481>
- Howe, A. J., Rodriguez, J. F., & Saco, P. M. (2009). Surface evolution and carbon sequestration in disturbed and undisturbed wetland soils of the Hunter estuary, southeast Australia. *Estuarine, Coastal and Shelf Science*, *84*, 75-83. <http://dx.doi.org/10.1016/j.ecss.2009.06.006>
- Jensen, L. A., Schmidt, L. B., Hollesen, J., & Elberling, B. (2006). Accumulation of soil organic carbon linked to Holocene sea level changes in West Greenland. *Arctic, Antarctic and Alpine Research*, *38*, 378-383. [http://dx.doi.org/10.1657/1523-0430\(2006\)38\[378:AOSACL\]2.0.CO;2](http://dx.doi.org/10.1657/1523-0430(2006)38[378:AOSACL]2.0.CO;2)
- Kruskal, W., & Wallis, A. (1952). Use of ranks in one-criterion variance analysis. *Journal of the American Statistical Association*, *47*, 583-621.
- Mook, D., & Hoskin, C. (1982). Organic determinations by ignition: Caution advised. *Estuarine, Coastal and Shelf Science*, *15*, 697-699. [http://dx.doi.org/10.1016/0272-7714\(82\)90080-4](http://dx.doi.org/10.1016/0272-7714(82)90080-4)
- Morris J. T., & Whiting, G. J. (1986). Emission of gaseous dioxide from salt-marsh sediments and its relation to other carbon losses. *Estuaries*, *9*, 9-19. <http://dx.doi.org/10.2307/1352188>
- Palomo, L., & Neill, F. X. (2009). Primary production and nutrient budgets of *Sarcocornia perennis* ssp. *alpini* (Lag.) Castroviejo in the salt marsh of the Palmones River estuary (Southern Spain). *Aquatic Botany*, *91*, 130-136. <http://dx.doi.org/10.1016/j.aquabot.2009.04.002>
- Pribyl, D. W. (2010). A critical review of the conventional SOC to SOM conversion factor. *Geoderma*, *156*, 75-83. <http://dx.doi.org/10.1016/j.geoderma.2010.02.003>

- Sanger, D. M., Holland, A. F., & Scott, G. I. (1999). Tidal creek and salt marsh sediments in South Carolina coastal estuaries: I. Distribution of trace metals. *Archives of Environmental Contamination and Toxicology*, 37, 445-457.
- Schumacher, B. (2002). *Methods for the determination of total organic carbon (TOC) in soils and sediments*. Las Vegas. Ecological Risk Assessment Support Center Office of Research and Development US. Environmental Protection Agency.
- Sims, C., & Cotching, W. (2000). Turbidity and sediment loads from selected catchments in north-west Tasmania. *Natural Resource Management Journal*, 3, 8-14.
- Twilley, R. R., Chen, R. H., & Hargis, T. (1992). Carbon sinks in mangroves and their implications to carbon budget or tropical coastal ecosystems. *Water, Air and Soil Pollution*, 64, 265-288. <http://dx.doi.org/10.1007/BF00477106>
- Yu, O. T., & Chmura, G. L. (2010). Soil carbon may be maintained under grazing in a St Lawrence Estuary tidal marsh. *Environmental Conservation*, 36, 312-320. <http://dx.doi.org/10.1017/S03710.1017/S0376892910000184>
- Zann, L. P. (1995). *Our Sea, Our Future: Major findings of the state of the marine environment report for Australia*. Townsville: Great Barrier Reef Marine Park Authority.

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