# Estimation of the sediment carbon storage in shallow bays of the Stockholm archipelago

Betty Gubri

BIG, Stockholm University Degree project in Marine Biology - 45 Hp Date: 15.04.22

Examiner: Jonas Gunnarsson, DEEP department, Stockholm University

Supervisors: Sofia Wikström, Östersjöcentrum, Stockholm University, Michael Tedengren, DEEP department, Stockholm University



# Estimation of the sediment carbon storage in shallow bays of the Stockholm archipelago

#### **Betty Gubri**

# Abstract

Seagrass meadows, mangroves, and salt marshes are commonly referred to as "blue carbon habitats", due to their natural ability to act as carbon sinks. Their protection and restoration are proposed as a measure to mitigate climate change. Today, it is important to identify new areas that could also act as important carbon reservoirs and contribute to these mitigation efforts. This study proposes a first approach toward the identification of a new potential blue carbon habitat in the Baltic Sea: the shallow sheltered lagoon-like bays of the Stockholm archipelago. These bays are complex littoral ecosystems with abundant rooted-macrophyte vegetation communities that are not dominated by seagrass species. The current isostatic land-uplift isolates them gradually from the sea, changing their sedimentation and biological processes over time. This phenomenon could increase their capacity to accumulate sediment and thus, their capacity to store large quantities of carbon. This study aims to (1) quantify the amount of carbon stored in the sediment of these shallow sheltered bays, and (2) explore the abiotic and biotic factors that influence these carbon stocks. The results revealed that carbon stocks estimates for the shallow bays are comparable to estimates obtained for seagrass meadows in the Baltic Sea. The sediment carbon content was mostly influenced by topographic openness and sediment density, but the differences in carbon stocks between the bays were not as noticeable as expected. This highlights the complexity of the processes occurring in these sheltered ecosystems. The results from this study are encouraging, but further research is recommended to support the present findings and to investigate their potential to act as a natural carbon sink. The results from this study suggest that the shallow sheltered bays of the archipelago are worthy of further investigation as local carbon storage that might contribute to climate change mitigation efforts.

**Keywords** semi-enclosed shallow bays, coastal vegetated ecosystems, blue carbon, sediment carbon stocks, soft-bottoms, Stockholm archipelago, Baltic Sea

# Popular Science Summary

Vegetated coastal ecosystems are recognized to be important natural carbon sinks and referred to as "blue carbon habitats". Mangroves, salt marshes, and seagrass meadows have the natural ability to trap and store atmospheric carbon in their sediment. The stored carbon can remain for centuries to millennia if the ecosystem is undisturbed, which offers a great advantage in mitigating the effects of climate change and the current rise of CO<sub>2</sub> emissions. Today, there is a necessity to research alternative habitats that could help with these mitigation efforts. The present study proposes to investigate a potential new blue carbon habitat in the Baltic Sea: the semi-enclosed bays of the Stockholm archipelago. These sheltered bays are complex ecosystems with abundant vegetation and they are progressively isolated from the sea due to the current post-glacial land rise in Scandinavia. This phenomenon gives special characteristics to the numerous bays of the archipelago and could increase the bays' capacity to store carbon in their sediment. This study aims to quantify the sediment carbon storage of these shallow bays and also to investigate which environmental factors influence their capacity to store carbon. The results revealed that carbon stocks estimates for the bays are comparable to estimates obtained for seagrass meadows in the Baltic Sea. It also highlights the complexity of the processes occurring in these sheltered ecosystems and further research is recommended to investigate their potential to act as a natural carbon sink. The results of this study suggest that the shallow sheltered bays of the archipelago are worthy of further investigation as local carbon storage and might possibly be regarded as a new blue carbon habitat if future studies support the present findings.

# Ethical considerations

This project did not involve the voluntary collection of animals. However, some benthic organisms were found in the sediment samples or attached to the vegetation samples. No extended harm to these organisms was done, as they died the moment that the samples were frozen and were discarded during the samples' preparation process.

# Contents

Introduction	1
Natural carbon sinks	1
Submerged blue carbon habitats	1
Blue carbon research for the Baltic Sea	2
Coastal bays of the Stockholm archipelago	3
Aim	4
Material and methods	5
Sampling design	5
Samples processing	8
Data analysis	10
Results	13
Carbon stocks, sediment and vegetation characteristics	13
Influence of sediment and environmental variables	16
Influence of above-ground vegetation	20
Carbon sources	21
Discussion	22
Environmental factors influencing the sediment carbon content	22
Carbon storage estimates in shallow sheltered bays	25
Origin of the bays' sediment carbon content	27
Methodological improvements and future research	29
Conclusion	31
Acknowledgements	33
References	34
Appendixes	39

# Introduction

# Natural carbon sinks

The importance of vegetated coastal ecosystems acting as natural carbon sinks was fully recognized with the emergence of the term "Blue Carbon Habitat" in 2005. Since then, there has been strong evidence that blue carbon coastal habitats like mangroves, tidal marshes, and seagrass meadows can sequester a significant proportion of carbon in their sediment (Hori et al., 2019; Macreadie et al., 2019; Trevathan-Tackett et al., 2015). It has been estimated that globally these ecosystems cover less than 2% of the ocean floor, but sequester more than half of the carbon buried in marine sediment (Chen & Xu, 2020; Hori et al., 2019; Ortega et al., 2020). These sediment carbon stocks can reside for long time scales (up to millennia), which qualify them as long-term sinks. This is of importance because they can help to mitigate climate change; yet, their rapid degradation can alter their capacity to function as a carbon sink (Hori et al., 2019; Mcleod et al., 2011; Ortega et al., 2020).

Blue Carbon Science quantifies global carbon stocks that can contribute to this effort toward climate mitigation (Chen & Xu, 2020; Mcleod et al., 2011; Ortega et al., 2020). It is still a new field and the consideration of marine macrophytes as carbon sinks was first introduced fifty years ago (Smith, 1981). Therefore significant gaps and uncertainties remain in the understanding of the carbon sequestration mechanisms in these ecosystems. The sink capacity of blue carbon (BC) habitats can be disrupted by anthropogenic pressures: this can result in a shift from their status of a net sink of carbon to a net source of carbon to the atmosphere (Hori et al., 2019; Kuwae et al., 2016; Mcleod et al., 2011). It has been argued that they could act simultaneously as net emitters of  $CO_2$  and net carbon reservoirs in human-dominated shallow coastal areas.

Thus, these ecosystems are still worth further investigation as contributors to local climate change mitigation (Hori et al., 2019; Kuwae et al., 2016). Today, it is important to identify other potential overlooked coastal biotopes that could also act as local carbon sinks (Chen & Xu, 2020; Hori et al., 2019; Krause-Jensen et al., 2018).

## Submerged blue carbon habitats

Seagrass meadows are the most well-researched submerged BC ecosystem to this date. The water column separates them from the atmosphere, therefore their carbon stocks are not directly associated with the removal of atmospheric CO<sub>2</sub>. The biogeochemical processes of inorganic & organic carbon in the water column are complex and knowledge gaps remain concerning the functioning of the CO<sub>2</sub> sequestration process (Krause-Jensen et al., 2018; Macreadie et al., 2019; Scheffold & Hense, 2020).

The carbon sink capacity of seagrass meadows is partly due to its vegetation primary production, but a key mechanism to their efficiency at sequestering carbon is their effectiveness in trapping sediments and carbon compounds from outer sources beyond their natural boundaries (Mcleod et al., 2011). The efficiency of these habitats to store carbon is influenced by several biotic and abiotic environmental variables and their interactions.

For example, sediment carbon stocks are influenced by wave exposure and the intensity of hydrodynamic processes. Sheltered habitats have been shown to accumulate finer sediment, which facilitate the deposition of organic matter; as opposed to more exposed habitats, where the hydrodynamics conditions result in higher exportation of organic compounds farther away and can favor erosion (Dahl et al., 2016; Jankowska et al., 2016; Moksnes et al., 2021; Röhr et al., 2016).

Usually, coastal sediments containing high proportions of mud content are also associated with high organic matter content (Mazarrasa et al., 2018). The accumulation of fine-grained particles and organic matter are both influenced by the hydrodynamics conditions, and the sediment type may also affect the carbon remineralization process, enforcing the correlation (Dahl et al., 2016; Mazarrasa et al., 2018; Serrano et al., 2014; Röhr et al., 2016).

The species diversity, cover, and density of the benthic plant communities can have a significant impact on the burial and storage of carbon below it (Jankowska et al., 2016; Lavery et al., 2013; Mazarrasa et al., 2018; Samper-Villarreal et al., 2016). Vegetated bottoms have generally higher sediment carbon stocks than unvegetated bottoms. The type of vegetation and community also influence the sediment stability: it enhances sediment trapping and thus its accumulation while reducing resuspension of finer particles. The meadow density and its patchiness impact the deposition of carbon. Hence, dense meadows with larger persistent species are associated with higher sediment carbon stocks than less dense and uneven ones (Dahl et al., 2016; Ewers Lewis et al., 2020; Lavery et al., 2013; Serrano et al., 2016).

Lastly, water depth has been shown to affect sediment carbon stocks (Mazarrasa et al., 2018; Serrano et al., 2014). Water depth is related to turbidity, irradiance, and light attenuation, which directly influence the meadows' growth and productivity (Macreadie et al., 2017; Mazarrasa et al., 2018). Generally, it is expected that shallower meadows have higher carbon storage that gradually decreases with depth, but this trend can be disrupted due to differences in local hydrodynamics conditions. Irradiance affects the seagrass meadow's carbon storage, especially between -2 m and -4 m (Lavery et al., 2013; Samper-Villarreal et al., 2016; Serrano et al., 2014).

## Blue carbon research for the Baltic Sea

In the Baltic Sea, the BC spotlight has been mainly focused on seagrass meadows, which tend to have lower sediment carbon contents compared to meadows in other marine regions. Seagrass commonly grows on exposed to semi-exposed, sandy bottom with good water exchange and they are present up to the Åland archipelago (Boström et al., 2003).

This makes them less likely to capture carbon and favor the exportation of the produced organic matter outside the habitat's boundaries(Dahl et al., 2016; Röhr et al., 2018). In addition, the low salinity of the Baltic Sea might influence the seagrass plant growth, which may affect the local production that could end up in the carbon pool.

A lot of organic material stored in the seagrass meadow sediment derives from surrounding areas, independently of local production (Dahl et al., 2016; Röhr et al., 2018).

They are composed of *Zostera sp.* and often mixed with limnic angiosperms, such as *Myriphyilum spp.* and *Potamogeton spp.*. These characteristics could alter their capacity to capture and retain organic matter to a certain extent for the most exposed meadows (Boström et al., 2003; Dahl et al., 2016).

Other vegetated areas composed of mixed rooted-macrophytes of freshwater origin are common in the coastal areas of the Baltic Sea and occupy more sheltered sediment habitats. These vegetation communities can be found in shallow bays of the Baltic Sea that are present up to the Gulf of Bothnia (Appelgren & Mattila, 2005; Schiewer, 2008).

There is very limited knowledge concerning the carbon sequestration of rooted-macrophyte habitats that are not dominated by seagrass species, but it could be argued that they share similar characteristics with seagrass meadows and they could also contribute to some extent to carbon sequestration. Thus, investigating these ecosystems represents an opportunity to identify a new habitat that could be considered as a carbon sink (Mazarrasa et al., 2018), and the shallow sheltered lagoon-like bays of the Stockholm archipelago could be a candidate towards this goal.

Generally, the accumulation bottoms prevail in the shallow, sheltered areas below 15m in the archipelago (Hill & Wallström, 2008). Considering the importance of hydrodynamic conditions for carbon storage in the sediment, then the semi-enclosed bays could be ideal spots for sediment carbon accumulation. Until the last three decades, these bays have been greatly overlooked by the research and monitoring programs in the Baltic Sea for the benefit of other more open coastal ecosystems (Hansen, 2013). The European Union Habitats Directive (Council Directive 92/43/EEC) implementation led to renewed interest in these particular biotopes and until now, they have never been studied as a potential BC habitat.

# Coastal bays of the Stockholm archipelago

The Stockholm archipelago is strongly influenced by its brackish water coastal area, which displays a gradient of salinity towards the inner part, due to an extensive outflow of freshwater from lake Mälaren. The postglacial land uplift is responsible for gradually isolating its numerous shallow bays from the sea. This phenomenon causes the bays' openness to slowly decrease, which limits progressively the water exchange between the bays and the sea. With time, the water properties and bottom-substrate characteristics of the bays are being altered, which directly impacts their ecological structure (Hansen, 2013; Hill & Wallström, 2008).

They are recognized as important ecological biotopes in the Baltic Sea (Hansen, 2013; Hill & Wallström, 2008), but their ecological functions can be affected in several ways by human activities such as increasing concentrations of phosphorus, turbidity, and level of boating activities inside the bays (Hansen and Snickars, 2014; Hill & Wallström, 2008; Eriksson et al., 2004).

The bays are characterized by soft-bottom sediments and their progressive separation from the sea has a direct impact on their submerged vegetation (Appelgren & Mattila, 2005; Hansen 2013). The vegetation consists of diverse plant communities of aquatic phanerogams and charophytes: mostly from freshwater to brackish water origin and marine origin (e.g. *Potamogeton perfoliatus, Myriophyllum spicatum, Stuckenia pectinata, Ceratophyllum demersum, Najas marina, Chara sp. ...*). Loose-lying algae (e.g. *Fucus vesiculosus*) can also be present. These communities are a mix of annual and perennial species, where their composition and density varies with the bottom characteristics and depth of the bays. The changes in species composition over time have been strongly linked to the openness of the bay (Hansen, 2013; Hansen et al., 2008; Hill & Wallström, 2008).

The shore-level displacement gradually decreases the bays' wave exposure which enhances the deposition and sedimentation of fine particles and organic matter in the sediment (Hansen, 2013; Hill & Wallström, 2008). It could be suspected that this can also result in decreased carbon outwelling for the most enclosed bays, based on explanations of the outwelling hypothesis from Santos et al. (2021). This could mean that the mobile carbon fraction would be less likely to be exported toward the sea, which could potentially increase its chance of sequestration in the bay's sediment with time and act as long-term carbon storage.

Finally, it would be expected that the ratio between terrestrial and marine organic carbon would increase with the decrease in the topographic openness of the bays (Hansen, 2013). The more enclosed bays would capture more allochthonous terrestrial carbon, due to higher input of water run-off from land compared to a relatively decreasing seawater inflow. The origin of organic carbon can be deduced from  $\delta^{13}$ C values of organic matter, so changes in carbon origins would be reflected in the  $\delta^{13}$ C-signal values of the organic matter in the sediment, but also in the  $\delta^{13}$ C-signal of the particulate organic matter in the water column (Bohlin et al., 2006; Hansen, 2013; Jönsson et al., 2005). However, Hansen (2013) reported that the particulate organic matter in the water column did not show noticeable changes along the gradient of isolation for the studied bays of the archipelago. The resulting pattern observed could be the consequence of several processes happening simultaneously, underlying the complexity of these biotopes.

#### Aim

The purpose of the study was to investigate the sediment carbon pool of shallow bays: as a first step in investigating if they could be considered as local blue carbon habitats that are worth further study. We hope that this study will contribute to gaining knowledge on the sediment carbon stocks of the vegetated shallow bays in the archipelago and, more generally, on rooted-macrophyte habitats not dominated by seagrass species.

The aim is to quantify the amount of carbon stored in the sediment of nine shallow bays and to determine if the variation in carbon stocks can be explained by specific environmental variables. The variables looked into by this study are water depth, the openness of the bay, sediment type (DBD, porosity, finer-grain particles content), and vegetation characteristics (composition, coverage, biomass).

I expected that the most enclosed bays have higher amounts of stored carbon compared to the more open ones and that these stocks are comparable to values obtained by seagrass meadows in the Baltic. I expected that the carbon stocks vary with sediment characteristics and are influenced also by vegetation characteristics. As the vegetation is composed of seasonal marine macrophyte communities, the variation explained by the vegetation characteristics was only investigated for the top sediment layer.

# Material and methods

# Sampling design

A total of nine bays were selected to represent a gradient of openness (Table 1). The bay selection was restricted to two areas to find bays that were reasonably close to each other, to facilitate the fieldwork. Four bays are located in the Vaxholm/Värmdö area, at the east of Stockholm, and five bays are located in the Furusund area, further north in the archipelago (Figure 1).





The topographic openness (Ea) is calculated based on the equation described by Hansen (2013):

$$Ea = 100 x \left( A_t / a \right) \tag{1}$$

where  $A_t$  is the narrowest section of the bay's opening connecting the bay to the sea (calculated from the mean water depth and the length of the bay opening on the field) and a is the water surface area (derived from topographic maps using GIS tools).

The selected bays represent a gradient of Ea from enclosed bays (Ea<0.05) to open bays (Ea<0.2). It is needed to keep in mind that a qualified "open bay" in this report should be interpreted as *the most open* of the semi-enclosed bays studied, as all the bays in this study are enclosed bays with narrow and shallow openings (Appendix 1).

Boy	Latituda	Longitudo	Locality	Area	Openness
Бау	Laulude	Longitude	Locality	(ha)	(Ea)
Östra Myttingeviken	59.380349	18.494024	Vaxholm/Värmdö	6	0.003
Eke fjärd	59.404603	18.259106	Vaxholm/Värmdö	19	0.009
Långbroviken	59.369418	18.472738	Vaxholm/Värmdö	10	0.013
Söderfladen	59.66371	18.86769	Furusund	7	0.017
Handfatet	59.674935	18.791428	Furusund	3	0.056
Fårholmsfladen	59.678792	18.795633	Furusund	7	0.101
Västra Lermaren	59.674458	18.865242	Furusund	13	0.259
Siviken	59.430328	18.309008	Vaxholm/Värmdö	8	0.403
Edvassviken	59.642959	18.768082	Furusund	4	0.591

#### Table 1. Locations, size and openness index of the studied bays.

The samples were collected between mid and late September 2021. The sites were investigated based on a stratified random sampling design and the sample collection was adapted from the methodology for seagrass meadows described by Howard et al. (2014).

The sites were investigated based on a stratified random sampling design and the sample collection was adapted from the methodology for seagrass meadows described by Howard et al. (2014). Two random samples were taken from the shallow and two from the deep part of each bay. The shallow samples were taken between 0.5 and 1.3 m depth and the deep samples between 1.7 and 2.9 m depth (the maximum depth in the study bays was 3 m). In one of the bays (Eke fjärd) the shallow area was all covered in reeds and one single "shallow" sample was taken at 1.5 m depth.

For each station, the water depth was taken with a plummet. The water depth measured was adjusted to the seawater level at the time of sampling, using data from the water level measurement station in Stockholm (provided by SMHI).

Then, a long sediment core was collected with a gravity corer (h = 50 cm,  $\emptyset$  = 6 cm). Once out of the water, the length of the core was measured and a compression factor was calculated on the spot following the equation in Skilbeck et al. (2017). The average core length was 34.94 cm (± 7.61), and the average compression was 8.3 % (± 7.7).

The sediment cores were divided into three parts representing the topsoil section (0 to 5 cm depth), an intermediate section (5 to 12.5 cm depth), and the bottom section (rest of the core). When the bottom section was long, only half of it was collected by cutting the section vertically to the sediment depth.

The sections were cut accounting for the compression factor, before being placed in labeled pre-weighed plastic bags and stored in a cooler with ice. This represents a total of 106 samples that were used for carbon content and grain-size analysis.

After the sediment sampling, a vegetation community survey was carried out by snorkeling, where a 1 m<sup>2</sup> visual quadrat was used to assess the total percentage of vegetation cover and the percentage cover of present species. Finally, a vegetation sample was collected with a 0.25 m<sup>2</sup> quadrat, placed in a labeled bag, and stored in the cooler, in order to measure the biomass and the richness of benthic vegetation. It should be noted that depending on the bay's characteristics, there were not always 4 vegetation samples collected, as some bays only had vegetation in their shallow parts (e.g Siviken, Långbroviken, Eke fjärd) or in their deeper parts (e.g Östra Myttingeviken). Some bays had vegetation present in both shallow and deeper areas (e.g Söderfladen, Handfatet, Fårholmsfladen, Edvassviken), while Västra Lermaren had a vegetation cover of less than 2 % in each of the sampling stations and no samples were retrieved. A total of 23 quantitative samples were collected.

In order to investigate if submerged or emergent vegetation affects the carbon stocks on a small scale (within bays), we did an additional sampling in three of the bays' shallower parts: Fårholmsfladen, Söderfladen, and Handfatet (Appendix 1). For this, we sampled four small cores of sediment with a syringe corer (h = 8.1 cm, ø = 2.7 cm) in each of three vegetation types: reeds, submerged vegetation, and bare sediment. No compression factor was applied, as it was judged to be insignificant on the field.

For each of the syringe cores in submerged vegetation, a corresponding vegetation sample was retrieved using a 25 cm<sup>2</sup> quadrat, to measure vegetation biomass. In the reed vegetation, the number of long, medium, and short reed shoots were counted with the quadrat. In addition, all reed vegetation in the quadrat was sampled in one of the bays (Handfatet) and the reed biomass for the other bays was calculated from the biomass/shoot ratio from Handfatet. All the biomass samples were placed in labeled plastic bags and stored in the cooler.

Back from the field, all samples were kept at -18 °C in a freezer until further analysis. A list of the vegetation species recorded for each bay is shown in Appendix 3.

## Samples processing

The sediment samples were divided into two sub-samples, one for grain-size analysis and one for analysis of carbon content. The vegetation samples were dried to estimate the dry weight of the above-ground plant biomass at each station.

#### Carbon content analysis

After defrosting, the sediment samples were homogenized and 30 mL of each sample (syringe and each section of the long cores) was placed in an aluminum form and weighed, before being let to dry in the oven at 60 °C for about 36 h. After the weight became constant, the samples were weighed again and big shells and marine fauna were retrieved.

The samples were then ground using a mortar and pestle and if needed, a mixer mill was used (adding 5 to 6 balls for about 5 to 7 min) to desegregate the dry sediment.

Using a milligram scale, tin capsules were weighed empty and 13 mg to 15 mg of sample was added before closing the capsules with pliers. The same process was repeated for silver capsules, except that after the sample deposit, 1M of HCl solution was added to each silver capsule to eliminate carbonates. After the reaction, the silver capsules were let to dry in the oven at 60 °C for 24 h before being closed using pliers. The organic carbon content and carbonate content of each sample were quantified by a mass spectrometer at the Center for Physical Sciences and Technology in Vilnius, Lithuania. The <sup>13</sup>C:<sup>12</sup>C isotope ratio ( $\delta^{13}$ C in ‰) was also measured in the process. No samples of potential carbon sources were taken on the field (e.g surrounding terrestrial plants, submerged vegetation, plankton...), so the reference values were taken from the literature (Bohlin et al., 2006; Hansen et al., 2012; Jönsson et al., 2005).

The sediment characteristics, e.g porosity, dry bulk density (DBD), and organic matter (OM), were calculated following equations from Smeaton et al. (2020), based on measured dry and wet weights of the samples and sample volume.

#### Grain-size analysis

Chemical oxidation followed by wet sieving was used to determine the particle size distribution. The choice of method was based on the timing and budget of this project, and also taking into consideration that our samples were likely to have a high content of fine-grained material. In fact, wet oxidation using hydrogen peroxide  $(H_2O_2)$  is commonly used to avoid problems of grain aggregate formation that arise with LOI methods for samples with high content of fine material and organic matter (López, 2017; Vaasma, 2008). Standard geological sieves were used.

After defrosting, the sediment samples were homogenized, placed in an aluminum form, weighed, and left in the oven at 60 °C for about 48 h to 72 h until the constant dry weight was reached. Big roots, benthic shells, or organisms were retrieved and the dry sample was weighed again. The samples were then placed in 500mL labeled containers for wet oxidation and the organic matter was removed by adding 3-10 % diluted  $H_2O_2$  solution to the sediment. This also helps to disperse the aggregation of particles by organic matter.

The samples were left to digest for at least 48 h and were stirred every day. After 48 h, the supernatant (if present) was removed with a pipette. If the reaction had ceased, a few drops of  $H_2O_2$  were added to see if all the organic matter was removed. If no reaction occurred, the sample was ready for wet sieving. If effervescence appeared, more diluted  $H_2O_2$  was added and the sediment sample was left to digest further until no more bubbling occurred.

A stack of sieves of decreasing mesh size (2 mm, 1 mm, 0.5 mm, 0.2 mm, 0.125 mm, 0.063 mm) was placed over a bucket and the sample was passed from the uppermost sieve through the sieves stack by using water. The content of each sieve was carefully collected with the help of a wash bottle in pre-labeled containers and left to dry in the oven at 60°C for 24 h until the weight was constant.

The next day, the dry samples were weighed to determine the different size fractions of the sediment section. The water in the bucket containing the mud fraction (silt and clay) was left to decant for 48 to 36 h. When settled, the surface water was carefully poured out, the mud was collected in a container and placed in the oven at 60 °C for 24 h. Afterward, it was weighed to the closest 0,001 g.

The results were presented in percentage composition of gravel (> 2 mm), very coarse sand (2 - 1 mm), coarse sand (1 - 0.5 mm), medium sand (0.5 - 0.2 mm), fine sand (0.2 - 0.125mm), very fine sand (0.125mm - 63  $\mu$ m) and mud (< 63  $\mu$ m corresponding to silt + clay).

Due to time restrictions, further analysis of finer material (<  $63 \mu m$ ) has not been made and the finer material measured, including clay and silt, was together classified as "mud content". From this process, the organic matter content of the sample, the proportions of the different size-grain classes, and the relative percentage of gravel/sand/mud for each sample were derived. A summary table of sediment classification per core depending on the bay, water depth, and sediment depth is shown in Appendix 2. The classification was determined by the proportion of gravel, sand, and mud content of each sample, following Folk's classification system.

#### Vegetation biomass quantification and identification

After defrosting, the vegetation samples were placed in a tray and cleaned with water. The different species of each sample were separated and identified to the lowest taxonomic level, before being placed in pre-weighed aluminum forms and left to dry in the oven at 60 °C for about 48 h. Afterward, the dry samples were weighed to the closest 0,001 g. The vegetation corresponding to the syringe cores (Najas, filamentous algae, and Chara) were grounded and placed into tin capsules to be analyzed for carbon content.

From this process, the detailed vegetation composition and the vegetation biomass quantification corresponding to a 25 cm<sup>2</sup> quadrat were estimated, as well as, the carbon content for the flora corresponding to the syringes samples.

## Data analysis

#### Carbon stocks

The total sedimentary carbon stocks in the upper 25 cm sediment layer were calculated following equations from Howard et al. (2014). The lengths of the cores were different, so for the analysis, they were all standardized to a total length of 25 cm.

First, the inorganic carbon was deduced from the total organic carbon for each sample, and then the organic carbon per unit area  $(gC_{org}.cm^{-2})$  was calculated for each section.

The bottom section value of  $gC_{org}.cm^{-2}$  of each core was calculated so it would represent a depth of 12.5 to 25 cm.

The carbon content of each sample is represented by sediment organic carbon per unit area  $(gC_{org}.cm^{-2})$ , and sediment organic carbon percentage (% $C_{org}$ ).

The carbon content values for each sample were calculated by applying a weight factor corresponding to each section's length out of the total core length over 25 cm.

The sum of each section, with applied weight factor, was summed up to obtain the total carbon content for each core.

The aim is to have a representative value over the 25 cm depth. Knowing that the bottom section accounts for half of the core, then it needs to be accounted for in the total carbon content of each core (see equation 2).

$$C_{core} = S_A x \ 0.2 + S_B x \ 0.3 + S_C x \ 0.5 \tag{2}$$

where  $C_{core}$  is the total organic carbon content of a core (% or  $gC_{org}$ .cm<sup>-2</sup>),  $S_A$  is the carbon content of the depth section 0 - 5cm,  $S_B$  is the carbon content for the section 5 - 12.5cm and  $S_C$  is for the section 12.5 - 25 cm.

The mean sediment carbon stock ( $C_{stocks}$ ) of a bay was calculated by averaging the total carbon content ( $gC_{org}.cm^{-2}$ ) of all cores from the same bay.

As explained in Röhr *et al.* (2018),  $C_{\text{stocks}}$  can be extrapolated from 25 cm depth to 100 cm depth and expressed as a projected total  $C_{\text{stocks}}$  in MgC, after being multiplied by the habitat area. Here, due to the bay's characteristics and the lack of knowledge concerning the depth of the post-glacial clay layer: there is no extrapolation in this study, in order to be cautious with the results' interpretations. The projected total  $C_{\text{stocks}}$  were calculated for the top 25 cm sediment layer following the equation:

projected total 
$$C_{stocks} = C_{stocks} x (Mg / 1 \ 000 \ 000 \ g) x (100 \ 000 \ 000 \ cm^2 / ha) x A_{Bay}$$
 (3)

where projected total  $C_{stocks}$  is in MgC over the upper 25 cm sediment,  $C_{stocks}$  is the mean carbon stocks of a bay (g $C_{org}$ .cm<sup>-2</sup>), Mg represents 1 megagram, ha represents 1 hectare, and  $A_{Bay}$  is the size of the bay (ha). The projected total  $C_{stocks}$  can be found in Appendix 6.

When investigating the mean of  $C_{\text{stocks}}$  per bay in the results section, the  $C_{\text{stocks}}$  are given in  $gC_{\text{org.}}\text{m}^{-2}$  for a better visualization. Values in  $gC_{\text{org.}}\text{m}^{-2}$  are obtained by multiplying the values in  $gC_{\text{org.}}\text{cm}^{-2}$  by a factor of 10000. The results were compared to carbon pools estimations for seagrass meadows in the eastern Baltic Sea and the Kattegat-Skagerrak area, presented in Appendix 5.

The organic carbon per unit area values (or carbon stocks per unit area) are expressed in  $gC_{org}.cm^{-2}$  for the rest of the report.

#### Sediment characteristics

The sediment properties, i.e. dry bulk density, and porosity, were calculated by following equations used by Smeaton et al. (2020):

$$DBD = M_s / V_s \tag{4}$$

where DBD is Dry Bulk Density (g.cm<sup>-3</sup>),  $M_s$  is the dry mass of the sample (g) and  $V_s$  is the wet sample volume (cm<sup>3</sup>).

$$p = M_w / [M_w + (M_s / DBD)] \times 100$$
(5)

where p is the porosity (%),  $M_w$  is the mass of water in the sediment (g),  $M_s$  is the dry mass of the sample (g) and DBD is the dry bulk density (g.cm<sup>-3</sup>).

To obtain the values of the different sediment characteristics for each core, each variable had weight factors applied depending on the section depth range, following equation 2 with the values corresponding to each variable. The mean values for each bay were obtained by averaging the values of all the cores inside a bay.

#### Statistical analysis

The response variables for carbon content were checked for normality and homogeneity of variances before the statistical analysis.

The differences in mean sediment percent of organic carbon (% $C_{org}$ ) and carbon per unit area (g $C_{org}$ .cm<sup>-2</sup>) between the bays were tested using a one-way ANOVA. The model residuals were approximately normally distributed and had homogeneous variances, but the model of % $C_{org}$  had two influential outliers in the residuals. Re-doing the analysis excluding them did not change the model's results, so I report the results from the model including the outliers. When ANOVAs were performed and the results were significant, a Tukey's test was run to check which bays differed.

To investigate which predictor variables were influencing the most carbon content in the upper 25 cm of sediment, several analyses were attempted. Principal Component Analysis, Partial Least Square Regression, and Multiple Regressions were tested, but due to the high correlation between the predictor variables, the restricted number of samples, and some outliers influencing the residuals, it was abandoned in favor of simpler models.

A correlation matrix was created from a model including all predictor variables for  $C_{org}$  and  $g_{C_{org}}.cm^{-2}$ , to visualize the individual relationship of each response variable to the different predictor variables. Linear Mixed-Effects Models (LMEM) were used to investigate the relationship between carbon content and a single predictor at a time. For some variables, the relationships had non-linear patterns, so Generalized Additive Mixed Models (GAMM) were run to smooth the regression line and compared to the LMEM results to see which regression model gave a better fit. GAMM were kept when results fitted better with the observations with a stronger correlation.

This was the case for the relationships between  $C_{org}$  - topographic openness, and carbon content - DBD (both  $C_{org}$  and  $gC_{org}$ .cm<sup>-2</sup>). All mixed models had bay as a random factor.

The linear models had approximately normally distributed residuals and homogenous variances, but three influential outliers occurred in all models with sediment characteristics. The regression models were re-tested on a dataset excluding them and compared to see their influence on the models.

The relationship between the vegetation characteristics and the carbon content in the upper 5 cm of sediment was examined with pairwise plots. Only the 5 cm upper section was taken into account in this case, due to the unknown sediment accumulation rate in the different bays and the unknown history of the vegetation community composition evolution with the isolation of the bay. Since there were no clear patterns, no regression models were run on this data due to lack of time.

Instead, the influence of the vegetation on surface carbon content was investigated more closely using the syringe samples from the three bays.

The effects of above vegetation on surface  $gC_{org}.cm^{-2}$  over the first 5cm of sediment, were tested using a permutational ANOVA, as the normality assumption was not met.

The differences in mean carbon source  $\delta^{13}$ C between the bays were analyzed using a one-way ANOVA. The model residuals were approximately normally distributed and had homogeneous variances. Two influential outliers were present in the model residuals, so the analysis was replicated excluding them. The results obtained were similar to the ones where they were included, so I present the results from the first model.

Four samples were lost following the carbon analysis by the mass spectrometer, and the three corresponding cores were discarded from the statistical analysis involving sediment carbon content and characteristics (two for Östra Myttingeviken, one for Långbroviken).

The bottom section of one core from Västra Lermaren was entirely constituted of post-glacial clay, so the  $\delta^{13}$ C value for this section was replaced by the mean of the bottom sections of the three other cores from the same bay.

All the statistics were performed using R Statistical Software (v4.1.2; R Core Team 2021) and the packages "car", "dplyr", "gamm4", "ggpubr", "ggplot2", "ggpmisc", "gt", "gridExtra", "lemon", "MuMIn", "mgcv", "multcompView", "nlme", "permuco", and "rstatix".

For conciseness in the rest of the report, organic carbon percentage will be referred to as  $C_{org}$ , and carbon stock per unit area as  $gC_{org}$ .cm<sup>-2</sup> (or  $gC_{org}$ .m<sup>-2</sup>).

# Results

## Carbon stocks, sediment and vegetation characteristics

There were significant differences in mean  $C_{org}$  among the bays (p = 6.58e-07, df = 8, F = 13.07, Figure 2A). The most enclosed bay (Östra Myttingeviken) had higher  $C_{org}$  values than all the other bays. The other enclosed bays (Söderfladen, Långbroviken and Eke fjärd) had higher  $C_{org}$  values than some or all of the most open bays (Västra Lermaren, Siviken and Edvassviken). Västra Lermaren had a lower %Corg than all the enclosed and one of the semi-enclosed bays (Fårholmsfladen).



**Figure 2. Sediment organic carbon content for the different bays.** Bays are displayed from the most enclosed to the most open. Values are presented as means ( $\pm$  SD) of %C<sub>org</sub> (A) and gC<sub>org</sub>.m<sup>-2</sup> (B). The percent organic carbon %C<sub>org</sub> is displayed as the mean carbon content for the upper 25 cm. The carbon per unit area gC<sub>org</sub>.m<sup>-2</sup> is presented as the total amount of carbon accumulated for the sediment profile (0 - 25cm). Letters above the bars represent the distinct groups according to the Tukey's test

Also  $gC_{org}$ .m<sup>-2</sup> differed between the bays (p = 0.0321, df = 8, F = 2.65, Figure 2B). The only significant difference was between Siviken and Långbroviken.

Siviken had the highest mean value of all sites, while Långbroviken had the lowest mean  $gC_{org}.m^{-2}$ . They did not differ from any of the other bays. Västra Lermaren had the highest variation in  $C_{org}$ stocks. The total projected carbon stocks in the upper 25 cm layer of the studied bay are presented in Appendix 5. Considering the small differences in mean  $gC_{org}.m^{-2}$  among the bays, the results suggest that the size of the bay influences to a greater extent the total carbon stocks for the shallow bays of the archipelago.

The sediment profiles obtained for %C<sub>org</sub>, are displayed in Figure 3 below.



Figure 3. Organic carbon content along the sediment profile. Bays are displayed from the most enclosed to the most open. Values are presented as means ( $\pm$  SD) of percent organic carbon %C<sub>org</sub> by depth sections. Depth sections represent the upper layer (0 - 5cm), intermediate layer (5 - 12.5 cm) and bottom layer (12.5 - 25 cm).

The %C<sub>org</sub> varied across the depth profile for Östra Myttingeviken, Långbroviken, and Söderfladen, where lower values were found in the bottom section (12.5 - 25 cm). There were more variations in %C<sub>org</sub> values at all depth sections for Söderfladen, especially for the upper 12.5 cm, compared to the other bays. Eke Fjärd, Handfatet, Fårholmsfladen, Västra Lermaren, Siviken and Edvassviken did not display any strong noticeable variations in %C<sub>org</sub> along the depth profile. The sediment depth profiles for carbon density are presented in Appendix 6. An increase in gC<sub>org</sub>.cm<sup>-3</sup> mean values with the depth profile was present for Östra Myttingeviken, Eke Fjärd, Söderfladen, Handfatet, Fårholmsfladen and Edvassviken. The bottom section had higher gC<sub>org</sub>.cm<sup>-3</sup> mean values compared to the upper 5 cm layer, and these bottom sections had twice more gC<sub>org</sub>.cm<sup>-3</sup> than the upper section in the case of Östra Myttingeviken, Handfatet and Fårholmsfladen.

There were more variations in  $gC_{org}$ .cm<sup>-3</sup> values at all depth sections for open Västra Lermaren, especially below 5 cm depth, compared to the other bays. The enclosed Långbroviken was the only bay with slightly higher  $gC_{org}$ .cm<sup>-3</sup> values in the upper 5 cm sediment, compared to the values in the 5 - 25 cm depth profile. Västra Lermaren, Siviken and Långbroviken did not display any strong noticeable variations in  $gC_{org}$ .cm<sup>-3</sup> along the depth profile.

Sediment and vegetation characteristics for the studied bays are shown in Table 2 and Appendix 7. The species richness per bay is shown in Appendix 3.

**Table 2. Summary of sediment and vegetation characteristics per bay.** Bays are displayed from the most enclosed to the most open one. Values are presented as mean values ( $\pm$  SD). Values for the sediment characteristics (DBD, porosity, mud content, and OM) are standardized over the upper 25 cm sediment layer. Vegetation cover and dry biomass represent the above-ground vegetation characteristics.

B	DBD	Porosity	Mud content	OM	Vegetation cover	Vegetation dry biomass
Вау	(g.cm-3)	(%)	(%)	(%)	(%)	<b>(</b> g)
Östra Myttingeviken	0.060 ± 0.002	46.4 ± 0.8	95.1 ± 0.0	48.8 ± 9.6	100 ± 0	8.21 ± 1.48
Eke fjärd	0.207 ± 0.080	45.0 ± 1.7	79.8 ± 7.0	26.9 ± 3.0	34 ± 57.2	1.69 ± 2.93
Långbroviken	0.103 ± 0.019	45.3 ± 0.6	88.2 ± 3.1	32.1 ± 5.8	23.7 ± 40.1	2.37 ± 4.11
Söderfladen	0.201 ± 0.091	43.3 ± 1.8	78.7 ± 20.3	32.9 ± 9.0	90 ± 11.5	9.33 ± 6.25
Handfatet	0.209 ± 0.036	44.5 ± 0.8	91.7 ± 5.2	21.2 ± 4.1	82.5 ± 23.6	16.1 ± 12.3
Fårholmsfladen	0.185 ± 0.084	44.5 ± 1.3	91.4 ± 4.9	24.5 ± 11.0	82.5 ± 23.6	2.69 ± 1.80
Västra Lermaren	0.585 ± 0.300	40.8 ± 3.1	67.8 ± 30.2	10.3 ± 5.8	1 ± 1.2	0 ± 0
Siviken	0.324 ± 0.029	43.3 ± 0.9	74.6 ± 7.0	16.1 ± 1.5	42.5 ± 50.6	1.81 ± 2.20
Edvassviken	0.481 ± 0.132	40.6 ± 1.1	69.4 ± 13.7	10.2 ± 1.7	45 ± 10	6.62 ± 3.72

Higher values of carbon density were found in more open bays, the same as for DBD. Eke Fjärd is qualified as an enclosed bay, but its sediment carbon characteristics values differ from the means for other enclosed bays. Porosity, mud content, and organic matter had lower mean values for the most open bays.

Graphs D and E of Appendix 7 also show the mean values of gravel and sand content per bay. The gravel content was higher for Söderfladen and Västra lermaren, and both bays had more variation compared to the rest of the bays that had minimum gravel content. The sand content was slightly higher in more open bays (Eke-Fjärd and Söderfladen) compared to the rest of the enclosed and semi-enclosed bays. For both sand and mud content, the highest variation among the cores was recorded for Söderfladen, Västra Lermaren, and Edvassviken.

From the sediment type classification given in Appendix 2, there is a clear difference in sediment class between the bays, and also among the cores in each bay. The most enclosed bays had more mud content compared to more open bays, except Eke-Fjärd for the 0-5cm depth section. More open bays were more sandy and had more gravel generally. The water depth of the core had some influence on the sediment type, especially for some depth intervals (e.g Östra Myttingeviken section 0 - 5 cm, Eke-Fjärd section 0 - 5 cm, Handfatet section 12.5 - 25 cm, Siviken section 12.5 - 25 cm).

The mean vegetation cover per bay, as well as the variation among the sampling stations within the bay, depended largely on the site. Eke Fjärd and Siviken had the most variation from 0 to 100% vegetation cover. Söderfladen, Handfatet, and Fårholmsfladen had an average of 90% vegetation cover, while Västra Lermaren had less than 2% vegetation cover. Note that the vegetation cover for Östra Myttingeviken is only representative of the deeper sampling stations, as the shallower ones were discarded due to the loss of sediment samples following the carbon analysis. From field observations, the shallow sampling stations from Östra Myttingeviken had an average of 25% vegetation cover.

The mean dry weight of the vegetation biomass was significantly higher for Handfatet, followed by Söderfladen, Östra Myttingeviken, and Edvassviken. Handfatet had the highest variation in vegetation dry weight.

In terms of vegetation communities, the most enclosed bays Östra Myttingeviken, Eke Fjärd, and Långbroviken had a community composed mainly of angiosperms with several species of filamentous algae. The most open bays Västra Lermaren, Siviken and Edvassviken were mainly composed of angiosperms. Söderfladen and the two semi-enclosed bays Handfatet and Fårholmsfladen had the richest vegetation communities with angiosperms, macro-algae (*Chara* sp. meadows), and several filamentous algae species. The common reed *Phragmites australis* was present along the shore of all bays.

## Influence of sediment and environmental variables

The correlation matrix present in Appendix 8.1 shows that there is a high correlation between  $%C_{org}$  and DBD, porosity, openness, sand, and mud. DBD had a high correlation coefficient with  $%C_{org}$  (r = -0.81), while porosity and mud content had a lower correlation coefficient (respectively: r = 0.68 and r = 0.56). The predictor variables were also highly inter-correlated with each other. Mud and sand content showed an almost perfect correlation (r = -0.97), so only mud content was investigated further here.

To avoid multicollinearity, DBD, porosity, and mud content were not tested in the same model, and simple regressions were used to investigate the relationships of the most correlated sediment characteristics with carbon content. The correlation matrix for  $gC_{org}$ .cm<sup>-2</sup> (Appendix 8.2) had different results concerning its correlation with the predictor characteristics and openness was the most correlated to  $gC_{org}$ .cm<sup>-2</sup> (r = 0.47).

The relationship between topographic openness and % $C_{org}$  was tested using a GAMM and the results showed a non-linear negative correlation of % $C_{org}$  with the bay's isolation to the sea (p = 0.01, F = 0.464, adjusted R<sup>2</sup> = 0.47, Figure 4A).



Topographic openness (Ea)

Figure 4. Relationship between topographic openness and carbon content for the different bays. Indication of the category limits, with dashed lines delimiting the cores from "enclosed", "semi-enclosed", and "open" bays. Values for  $%C_{org}$  (A) are presented as the mean carbon content for the upper 25cm. Values for  $gC_{org}$ .cm<sup>-2</sup> (B) are presented as the total amount of carbon accumulated for the sediment profile (0 - 25 cm).

LMEM were used to test the relationship between topographic openness and  $gC_{org}.cm^{-2}$ , and the results showed a weak but significant positive linear correlation (p = 0.041, R<sup>2</sup> = 0.22, Figure 4B).

LMEM was also used to test the relationship between water depth and carbon content. There was no distinct relationship between water depth and % $C_{org}$  (p = 0.871, R<sup>2</sup> = 0.004), nor between water depth and g $C_{org}$ .cm<sup>-2</sup> (p = 0.946, R<sup>2</sup> = 0.0001).

Linear regressions with one predictor variable were used for all variables, but the relationship between carbon content and DBD did not appear to be linear. Thus, GAMM with a single predictor variable was tested and compared to the results obtained by LMEM, to see which regression model was more fitting to the observations (Appendix 9 and 10). The carbon content had a linear relationship with porosity and mud content, while it had a non-linear pattern for DBD. Generally, there was a stronger relationship between %C<sub>org</sub> and the predictor variables than for gC<sub>org</sub>.cm<sup>-2</sup>.

The carbon content was highly correlated to DBD, in term of both  $\[\%C_{org}\](p < 2e-16, F = 11.21, adjusted R^2 = 0.83, Appendix 10A) and gC_{org}.cm^{-2}\](p = 7.41e-06, F = 8604, adjusted R^2 = 0.57, Appendix 10D). Carbon content was less correlated with porosity, but remained a significant predictor variable for <math>\[\%C_{org}\](p = 0.010, R^2 = 0.46, Appendix 10B)\]$ , but not for gC<sub>org</sub>.cm<sup>-2</sup> as it was not significant (p = 0.669, R<sup>2</sup> = 0.04, Appendix 10E). Carbon content was even less correlated with mud content. It was a significant predictor variable for  $\[\%C_{org}\](p = 0.004, R^2 = 0.32, Appendix 10C)\]$ , but again, it was not a significant predictor variable for gC<sub>org</sub>.cm<sup>-2</sup> (p = 0.903, R<sup>2</sup> = 0.016, Appendix 10F).

From this first process, three outliers appeared to drive the observation pattern for all sediment characteristics, especially for DBD (Appendix 10D). They are mean values for one shallow core in Edvassviken and the two shallow cores in Västra Lermaren. Two of the three cores had reached the post-glacial clay layer when they were being sampled.

The core from Edvassviken had the clay layer starting at -22 cm depth, but it was excluded from the mean values in the statistics as it was a really small fraction of the bottom section. The core from Västra Lermaren had the clay layer starting at -11 cm depth, so the bottom section was completely constituted of post-glacial clay and it was therefore included in the analyses. The LMEM and GAMM regressions were repeated on a dataset that excluded these outliers and the results are presented in Appendix 9 and Figure 5. Overall, it did change the results obtained for carbon content, but it affected more the results obtained for  $gC_{org}$ .cm<sup>-2</sup> (Figure 5 below).



Figure 5. Relationships between carbon content (% $C_{org}$  and  $gC_{org}$ .cm<sup>-2</sup>) and the sediment characteristics (outliers excluded). Observations represent the core mean values of carbon content and predictor variables over the 25 cm upper sediment layer. The first line shows the relationships between % $C_{org}$  and (A) DBD (g.cm<sup>-3</sup>), (B) porosity (%), (C) mud content (%). The second line shows the relationships between  $gC_{org}$ .cm<sup>-2</sup> and the predictor variables: (D) DBD (g.cm<sup>-3</sup>), (E) porosity (%), (F) mud content (%). The same graphs with the outliers included can be found in Appendix 10).

By excluding the outliers, the correlation between  $C_{org}$  and the predictor variables decreased and the relationship was less significant. DBD remained highly correlated to  $C_{org}$  (p < 2e-16, F = 11.36, adjusted R<sup>2</sup> = 0.82, Figure 5A). Porosity became insignificant as a predictor variable for  $C_{org}$  (p = 0.064, R<sup>2</sup> = 0.32, Figure 5B). Mud content remained a significant predictor variable for  $C_{org}$  (p = 0.013, R<sup>2</sup> = 0.17, Figure 5C).

On the contrary, the relationships between  $gC_{org}$ .cm<sup>-2</sup> and the predictor variables became all significant and the correlations increased, when the outliers were excluded. Thus, the  $gC_{org}$ .cm<sup>-2</sup> became highly correlated to DBD, which became a significant predictor variable (p p < 2e-16, F = 31796, adjusted R<sup>2</sup> = 0.73, Figure 5D).

Note that the LMEM results for DBD relationships with  $gC_{org}$ .cm<sup>-2</sup> became significant, but with a R<sup>2</sup> value lower than for the GAMM regression (Appendix 10). The porosity became more correlated to  $gC_{org}$ .cm<sup>-2</sup> and became a significant predictor variable (p = 0.020, R<sup>2</sup> = 0.35, Figure 5E). The mud content became poorly correlated to  $gC_{org}$ .cm<sup>-2</sup> values, and it was weakly significant (p = 0.185, R<sup>2</sup> = 0.18, Figure 5F).

### Influence of above-ground vegetation

The permutational ANOVA showed no significant differences in  $gC_{org}$ .cm<sup>-2</sup> means between the vegetation types (p = 0.348, df = 2, F = 1.098), no significant differences in  $gC_{org}$ .cm<sup>-2</sup> means between bays (p = 0.156, df = 2, F = 1.989), and the interaction between these variables was not significant either (p = 0.606, df = 4, F = 0.689).



Figure 6. Sediment  $gC_{org}$ .cm<sup>-2</sup> depending on the above vegetation type for the different bays. Values are presented as means (± SD) of  $gC_{org}$ .cm<sup>-2</sup> of the upper 5 cm of

**different bays.** Values are presented as means ( $\pm$  SD) of gC<sub>org</sub>.cm<sup>2</sup> of the upper 5 cm of sediment. The submerged vegetation species were different among bays, with dominant *Chara sp.* for Söderfladen and *Najas marina* for Fårholmsfladen and Handfatet.

Figure 6 shows that there was a larger variation in  $gC_{org}$ .cm<sup>-2</sup> in Fårholmsfladen compared to the other bays and that the variation was larger in the sediment in the reeds area.

The submerged vegetation composition was different between the three bays with Söderfladen having predominantly *Chara* meadows, while Fårholmsfladen and Handfatet had predominantly *Najas* meadows. Handfatet had significant filamentous algae cover compared to the other bays. The differences in species communities did not influence the results significantly in terms of surface sediment carbon content.

#### **Carbon sources**

There were significant differences in mean  $\delta^{13}$ C per bay, for the upper 25 cm sediment layer (p = 8.88e-10, df = 8, F = 22.27, Figure 7). There was a significant difference between Söderfladen and the rest of the other enclosed bays, and Långbroviken differed from both Söderfladen and Östra Myttingeviken.



Figure 7. Mean  $\delta^{13}$ C per bay over the upper 25cm sediment layer. Bays are displayed from the most enclosed to the most open one. Values are presented as core means of  $\delta^{13}$ C in ‰ (± SD), over the depth profile (0 - 25cm). Letters represent the distinct groups according to the Tukey's test.

Söderfladen had the highest  $\delta^{13}$ C value of all sites. Handfatet and Fårholmsfladen had higher  $\delta^{13}$ C values than most of the bays. Eke Fjärd and Långbroviken had lower  $\delta^{13}$ C values than the rest of the sampled bays, with Långbroviken having the lowest  $\delta^{13}$ C value of all sites.

The mean  $\delta^{13}$ C values showed no consistent trend with the topographic openness of the bay. All the bays with low  $\delta^{13}$ C values were located in the southern study area Vaxholm/Värmdö and the bays with the highest  $\delta^{13}$ C values were situated in the northern area Furusund.

There were a few patterns in  $\delta^{13}$ C values between the depth sections of the sediment profile (Appendix 11). The  $\delta^{13}$ C values increase gradually with sediment depth for some bays (Eke Fjärd, Söderfladen, and Handfatet). Other bays only had higher  $\delta^{13}$ C values in the bottom section compared to the values of the above sediments (Fårholmsfladen and Siviken). Two bays did not show a strong increase or decrease in  $\delta^{13}$ C values across the depth profile (Edvassviken and Östra Myttingeviken).

# Discussion

# Environmental factors influencing the sediment carbon content

A clear pattern was observed for the sediment carbon content, where an increase in topographic openness was associated with a decrease of  $C_{org}$  in the sediment. There was also a decrease in sediment  $C_{org}$  with an increase in sediment density. On the other hand, a decrease in  $C_{org}$  was associated with an increase in sediment porosity and mud content. The sediment variables and the topographic openness were all significant predictors for  $C_{org}$  and DBD was the predictor variable that explained the most the  $C_{org}$  values observed. The topographic openness was the second predictor explaining partially the  $C_{org}$  values obtained, and it was also highly intercorrelated with DBD. Porosity and mud did not have a strong relationship with the observed  $C_{org}$  values.

The  $%C_{org}$  along the sediment profiles differed between bays, but it was generally higher in the uppermost layer of sediment. The sediment layer containing very high carbon content had different thicknesses in the different bays, while the carbon density in the sediment increased along with the depth profile. It suggests that the uppermost layer of this sediment had low density but high OM content. This pattern was observed on the field while retrieving the sediment samples. The sediment cores from more enclosed bays had a layer of decomposing organic matter that was sitting on top of a denser sediment layer, and the limit between these two layers was not always distinguishable.

This type of sediment was previously described in the Pomeranian Bay by Emeis et al. (2002) and in lagoons of the southern Baltic Sea by Ulyanova et al. (2014). It is referred to as "fluffy layer suspended matter" (FLSM) and described as deposited organic-rich material that is resting on the sea bottom. This layer has high water content and looks like it is 'floating' over the bottom, corresponding to what was seen on the field. In sandy areas, the limit between FLSM and

the bottom sediment can easily be seen. But in soft-bottom areas, this layer may not be easily delimited from the underlying muddy sediment and can show a gradual change in physical and chemical properties (Emeis et al., 2002). It is reported that the suspended matter is incorporated by both physical and biological processes into the underneath sediment (Pempkowiak et al., 2002).

On the contrary, the sediment from more open bays had a negligible layer of organic matter at the top of the core. The sediment was generally more compact and the post-glacial clay layer was visible in two cores retrieved in two different open bays. This indicates that the fluffy layer properties change depending on the hydrodynamic conditions of the shallow bays. In fact, it was reported that the formation of a high-density fluffy layer on the sediment is optimal in calm weather conditions, but it can be quickly resuspended and mixed in the presence of waves (Pempkowiak et al., 2002; Ulyanova et al. 2014). Moreover, spatial changes in FLSM were observed due to variable river input, while temporal changes were also noticed due to the hydrological factors and biological activity variations induced by the climate (Emeis et al., 2002; Pempkowiak et al., 2005; Ulyanova et al. 2014).

The hydrodynamic conditions of the more open sheltered bays are responsible for the erosion of the surface sediment, which might prevent rapid sediment accumulation and increase the sediment compaction. Thus, the sediment layer with organic content for Edvassviken and Västra Lermaren was so thin that the core section retrieved extended down to the post-glacial clay layer. On the contrary, the weak hydrodynamics of more enclosed bays would be more favorable to the sediment accumulation and would lead to the accumulation of organic matter in the form of a fluffy layer on the sea bottom. This layer can easily be resuspended in the water column, but the sheltered characteristics associated with the small opening of the bays might naturally prevent it.

The carbon content in terms of  $gC_{org}.cm^{-2}$  showed an opposite relationship with all predictor variables, compared to the pattern seen with %C<sub>org</sub>. Then an increase of  $gC_{org}.cm^{-2}$  was associated with an increase in topography openness and sediment density. On the contrary, a decrease of  $gC_{org}.cm^{-2}$  was related to an increase in sediment porosity and mud content. DBD was the factor explaining the most the variations in  $gC_{org}.cm^{-2}$  values, followed by topographic openness. Porosity and mud content were not good predictors for explaining  $gC_{org}.cm^{-2}$ .

In the results, three cores appeared to be different from the other samples for almost all the sediment variables and they all came from two of the more open bays with more intense hydrodynamic conditions (Edvassviken and Västra Lermaren). Once they were removed, the relationship between  $gC_{org}$ .cm<sup>-2</sup> and porosity became significant, while the relationship of  $gC_{org}$ .cm<sup>-2</sup> with mud content remained insignificant. As for %C<sub>org</sub>, DBD had a strong relationship with  $gC_{org}$ .cm<sup>-2</sup> values and was the best predictor variable. Topographic openness did not have a strong relationship with observed  $gC_{org}$ .cm<sup>-2</sup> values. Porosity was only a significant variable when the three outliers were excluded, but the relationship with  $gC_{org}$ .cm<sup>-2</sup> was not strong either. Mud content did not influence the  $gC_{org}$ .cm<sup>-2</sup> values in this study.

The water depth and the vegetation characteristics did not explain the sediment carbon content variations observed. Some of the shallow bays had vegetation all the way down to their deeper part and there was no consistent pattern in terms of vegetation cover and biomass with water depth in the studied sites.

It has been shown that macrophyte communities can enhance locally the sediment carbon content, compared to non-vegetated bottoms (Jankowska et al., 2016; Röhr et al., 2016; Scheffold & Hense, 2020). This is not the case in the investigated bays. The vegetation characteristics did not have any effect on the small-scale level and were not linked to noticeable variation in  $gC_{org}$ .cm<sup>-2</sup>.

The vegetation biomass and cover are highly dynamic in both space and time, making the results of an up-close study difficult to interpret accurately over such a short time. In fact, the vegetation may differ between years, and the patches investigated may have had different vegetation, while the carbon content in the uppermost 5 cm integrates over a longer time period than the current growth season. The sampling was done in late summer and the aquatic species recorded are mostly seasonal, so this study does not account for annual variations. The common reed (*Phragmites australis*) is a perennial species that was present to a large extent along the shoreline of all bays. Reeds start to decay in autumn but keep rigid stems during winter. The submerged vegetation is mainly composed of fast-growing annual species that lose all or at least a significant part of their above-ground biomass during winter, due to bottom freezing conditions (Appelgren & Mattila, 2005). If the roots remain, the perennial species can come back every year and could still contribute to some extent with sediment-binding properties during the colder season or when the above-ground biomass is low. Additional sampling during winter would be required to confirm that the plant community presence and composition have a negligible influence on the surface gC<sub>org</sub>.cm<sup>-2</sup>.

However, it could be assumed that the vegetation community has a minor influence on the particle resuspension if the bay has extensive dense macrophyte meadows, especially during spring and summertime. The summer season is associated with higher intensity of recreational boating which can increase the hydrodynamics of the bay (Appelgren & Mattila, 2005; Eriksson et al., 2004). The plant community would then contribute to the stabilization of the seabed with their roots or rhizomes, which could reduce the resuspension process. A study by Austin et al. (2017) highlighted that a high cover of aquatic vegetation can reduce water turbidity during spring and summer in shallow bays along the central Swedish Baltic Sea coast. This could be the case for bays similar to Söderfladen, Handfatet, and Fårholmsfladen. The number of sampled bays was restricted and further research needs to be carried out to confirm this hypothesis. It is certain that if the vegetation played a great role in the sequestration process of carbon in the sediment, then it would be reflected in the results even from a small dataset.

These results obtained for the shallow bays of the archipelago contrast clearly with those from studies investigating the factors driving sediment  $C_{org}$ stocks in seagrass meadows of the Baltic Sea, over the same sediment depth (Dahl et al., 2016; Röhr et al., 2016; Röhr et al., 2018). Commonly, the most important predictors of carbon sink capacity of seagrass meadows are sediment characteristics (mud content, porosity, DBD, grain size), water depth, and plant characteristics (species, biomass, canopy complexity, cover, density). In seagrass studies, the sediment carbon content values in  $%C_{org}$  and  $gC_{org}$ .cm<sup>-2</sup> are usually following a similar pattern across sites with more contrasting results than the ones obtained for the shallow bays. As stated before, seagrass meadows are present in more exposed areas and higher hydrodynamics exposure was associated with higher  $C_{org}$ stocks (Dahl et al., 2020; Röhr et al., 2018).

The exposed seagrass meadows have less delimited boundaries compared to the shallow sheltered bays of the archipelago. Consequently, it has been discussed that there is a large quantity of organic carbon produced by seagrasses that is exported outside the ecosystems' boundaries (Röhr et al., 2018).

The vegetation has a more important role in seagrass meadows to trap and increase sediment accumulation locally, while the more enclosed bays did not display a major role of the vegetation, probably due to the calmer conditions of the environmental settings.

Additionally, the seagrass sediment generally has more sand content than the sediment of the soft-bottom bays, which could suggest that in more sandy areas the proportion of fine-grained sediment is more important to predict  $C_{org}$  stocks.

Overall, the proportion of mud in the sediment was not a good predictor of carbon content for the shallow sheltered bays of the archipelago. Mud content was recognized to not be a universal variable to predict sediment carbon content for seagrass meadows (Serrano et al., 2016). The silt or clay fraction, as well as the organic matter content, were sometimes better predictors to explain variations in sediment  $C_{org}$ stocks (Dahl et al., 2020; Serrano et al., 2016). Here, the limitations in time and budget did not allow us to separate the mud content into the silt and clay fractions and it was not possible to investigate the possible impact of the clay fraction on the sediment carbon content.

More details concerning the bottom topography, the sediment rate accumulation, and the fauna and flora community temporal changes, would help to better understand the complex processes that are occurring in those ecosystems. Contrary to seagrass meadows, there is no considerable spatial variability in the amount of carbon stored in the sediment (Lavery et al., 2013). The small differences of  $gC_{org}$ .cm<sup>-2</sup> observed occurred over a wide range of topography openness (thus of hydrodynamic conditions). So the investigation of which specific variable would be the best at predicting  $gC_{org}$ .cm<sup>-2</sup> is not justified as a priority for further research.

## Carbon storage estimates in shallow sheltered bays

A summary of carbon pool estimations for seagrass meadows in the Baltic area and the Skagerrak-Kattegat strait can be found in Appendix 4. The  $C_{org}$ stocks estimates for the shallow bays of the archipelago are comparable to values obtained for seagrass meadows in the eastern Baltic Sea area. On average, the bays had twice as much carbon per unit area than values reported for meadows of the eastern Baltic Sea area. However, the bays'  $C_{org}$ stocks estimations represent at best half the previous estimates of sediment  $C_{org}$ stocks for the seagrass meadows in the Skagerrak-Kattegat strait.

Note that previous estimates of sediment  $C_{org}$  for these seagrass habitats are generally way lower than the ones obtained in this study. The values obtained for most exposed sites here (Västra Lermaren and Edvassviken) are close to the sediment  $C_{org}$  values obtained for the seagrass meadows of the Kattegatt-Skagerrak strait.

While there is a pattern in seagrass meadows where higher  $C_{org}$  is associated with overall higher sediment  $C_{org}$  stocks, this is not true for the shallow bays of the archipelago.

There is a significant correlation that the most enclosed bays have higher  $%C_{org}$  values compared to the most open bays, but there was no similar trend with the sediment  $C_{org}$ stocks. The results did not show a strong difference in  $gC_{org}$ .cm<sup>-2</sup> values between the bays, which suggests that there is no distinct pattern between the sediment  $%C_{org}$  values and the sediment  $C_{org}$ stocks for the shallow bays (at least for the 25 cm sediment depth profile). It was initially expected that the sediment  $C_{org}$ stocks estimates would be more noticeable along the topographic openness gradient than the results obtained by this study. The results indicate a possible counter-effect of the hydrodynamic conditions of the bays. Presumably, the extremely high  $%C_{org}$  values in the very loose organic sediment of the enclosed bay clearly indicate that the environment is favorable to the accumulation of organic matter on sheltered bottoms. This leads to low DBD and low carbon density in the upper part of the sediment and it can result in a low estimation of the carbon stocks when only looking at the uppermost 25 cm.

One question of interest is whether the sediment  $C_{org}$ stocks of these bays are particularly distinct from the amount of carbon in the sediment surrounding shallow coastal areas in the open archipelago. It could be assessed by comparing the values of this study with values obtained for the amount of carbon stored in the sediment of the surrounding shallow coastal areas in the archipelago. If the estimates for the bays are more important than the estimates of shallow exposed areas in the archipelago, then it would support the theory that the shallow semi-enclosed bays represent notable local carbon storage areas. Such carbon content value estimates were researched in sediment reports and scientific literature about the Stockholm archipelago sediment. Unfortunately, the estimates found were related to sediment sampled at deeper depths and were given in either total carbon percentage (%C<sub>total</sub>) or %C<sub>org</sub>. It is not possible to compare these estimates with the results from this study, both due to depth differences and to the %C<sub>org</sub> that was not representative of the carbon per unit area inside the bays.

The lack of knowledge concerning the bays limited our ability to conclude about the total projected  $C_{org}$ stocks for the entire bays. This study did not have prior knowledge about the specific sediment accumulation rate or the total thickness of the sediment layer where the carbon is stored (down until the post-glacial clay layer) for each bay. The results only reflect that there were no strong differences in average  $C_{org}$ stocks between the bays in the uppermost 25 cm sediment layer. It can be expected that the size of the bay would be an important factor influencing the most the total projected  $C_{org}$ stocks, at least for the 25 cm sediment depth profile. The results shown in Appendix 5 encourage this assumption.

The selected sites represent an acceptable sampling of both small and larger bays for each category defined by topographic openness. It suggests that there is a tendency for the larger bays to have a more important total projected  $C_{org}$  stocks compared to the smallest bays, regardless of their openness.

# Origin of the bays' sediment carbon content

Typically, the origin of organic matter in marine sediments is deduced using the stable carbon isotope ratio of the surrounding plant material (Bohlin et al., 2006; Jönsson et al., 2005). Indeed, the photosynthetic mechanisms and the carbon dioxide sources will differ between terrestrial plants and aquatic plants. These differences will influence the <sup>13</sup>C:<sup>12</sup>C isotope ratio, from which it is possible to infer the origin of the sediment organic matter (Jönsson et al., 2005).

Terrestrial plants derive their carbon from atmospheric carbon dioxide and have a  $\delta^{13}$ C range of -23 ‰ to -30 ‰, while aquatic plants from freshwater origin derive most of their carbon from dissolved inorganic carbon with  $\delta^{13}$ C values ranging between -18 ‰ and -28 ‰. Freshwater phytoplankton has an average  $\delta^{13}$ C of -30 ‰, meaning that it is not possible to distinguish lake-derived organic matter from land-derived organic matter with  $\delta^{13}$ C (Bohlin et al., 2006). Organic matter derived from marine phytoplankton generally have  $\delta^{13}$ C values ranging from -20 ‰ and -22 ‰ (Bohlin et al., 2006; Jönsson et al., 2005)

Commonly, low values of  $\delta^{13}C$  are interpreted as an indication of a large input of terrestrial organic carbon and high  $\delta^{13}C$  values generally indicate the dominance of carbon from marine origin.

In similar bays of the archipelago, the  $\delta^{13}$ C values of the shore vegetation were recorded at -27 ‰ for reeds and -29 ‰ for deciduous trees (Hansen et al., 2012). Higher  $\delta^{13}$ C values were recorded for submerged primary producers. The values recorded for angiosperms ranged from -9 ‰ to -15 ‰ (*Potamogeton sp., Myriophyllum sp.*), values for filamentous ephemeral algae ranged from -14 ‰ to -21 ‰, values for the macro-algae ranged from -12 ‰ to -16 ‰ for *Fucus vesiculosus* and from -10 ‰ to -13 ‰ for *Chara* spp. (Hansen, 2013).

The sediment  $\delta^{13}$ C values of the organic carbon in the sediment did not display a gradual decrease in relation to the bay isolation as could be expected. These results support and complement the findings of Hansen et al., (2012) and Hansen (2013), where an increase in the ratio of terrestrial to marine organic carbon with increased isolation of the bays was not reflected in the  $\delta^{13}$ C-signal of particulate organic matter in the water, nor further in the food-web structure.

All the bays with low  $\delta^{13}$ C were situated in the southern Vaxholm/Värmdö area and the bays with the highest values were located in the northern Furusund area. This could indicate a large-scale difference in carbon sources between the areas.

To assess whether the carbon source is dependent on the geography, additional research would be needed with a focus on the sediment carbon source of this type of bay and a higher number of sites should be investigated in both regions. However, it is interesting to note that the southern study area is situated next to lake Mälaren and as a consequence, it potentially receives a larger input of freshwater than the northern study area.

According to a map from Hill & Wallström (2008), the Vaxholm/Värmdö region is located in the outflow of surface water from Lake Mälaren. It is reported that this outflow is the source of 39% of the carbon in the sediments found in the innermost part of the archipelago near Stockholm.

The organic matter from the Vaxholm/Värmdö region had relatively high  $\delta^{13}$ C values ranging between -22 ‰ and -26 ‰ on average.

A previous study by Jönsson et al. (2005) indicated that the phytoplankton from the inner part of the archipelago could have values as low as -22 ‰ to -24 ‰ due to the alkalinity of the seawater and the large input of dissolved terrestrial carbon in some areas. For this part of the archipelago, previous estimates of sediment  $\delta^{13}$ C-signal had an average of -24 ‰, which was reported to be typical of marine plankton from the Baltic. The C/N ratio from the same study suggested a dominance of the sediment organic matter of planktonic origin, rather than from terrestrial sources.

The present investigation of carbon sources is too limited to carry a more detailed analysis of the origin of the sediment carbon content for the bays. However, considering the closeness of the sites investigated here with the ones by Jönsson et al. (2005), it is possible that similar results could be obtained for the Vaxholm/Värmdö region. Additional analysis on the carbon sources of the bays' sediment is needed to support this assumption.

According to the same map from Hill & Wallström (2008), the Furusund area is located in the main southward coastal stream of the archipelago, with water flowing mainly from the Baltic Proper. It could be argued that the inflow of water entering the bay by its opening would be mainly composed of seawater from the Baltic Proper. The surrounding coastal region is also less populated compared to the dense Stockholm coastline. Then, the proportion of freshwater and organic carbon from terrestrial sources would be quantitatively less important than for the southern part. This assumption would lead to higher  $\delta^{13}$ C values in the sediment, suggesting a marine origin to the sediment's organic carbon. In the Furusund area, higher  $\delta^{13}$ C values in the sediment were obtained, which could support this assumption. However, more samples from this region would be required to confirm this hypothesis.

Variation in  $\delta^{13}$ C between the bays of similar areas could result from other processes specific to each bay and local additional inputs of terrestrial allochthonous carbon (Bohlin et al., 2006; Hansen et al., 2012). These could be related to the presence of anthropogenic pressures and their extent (sewage treatment plant, high nutrient water run-offs), hypoxic and anoxic conditions of the sediment, the periodic fluctuation of the inflow/outflow of water in the bay related to differences in water level, or the submerged vegetation community characteristics (biomass, coverage, composition).

The three bays with the highest  $\delta^{13}$ C values recorded (Söderfladen, Handfatet, Fårholmsfladen) are also the bays with the richest plant community composition, relatively high coverage of the vegetation community, and the most diverse plant composition (Appendix 3 and Appendix 7 G). These bays were the only sites with a presence of Chara meadows and *Chara* spp. was recorded with high  $\delta^{13}$ C values in similar bays (Hansen, 2013). When these macro-algae and the other angiosperms decompose in autumn, they could influence to some extent the sediment  $\delta^{13}$ C values. This remains a simple observation and no conclusion can be made regarding the vegetation characteristics influence on the average sediment  $\delta^{13}$ C with these results.

## Methodological improvements and future research

The methodology could be improved to gain time during the analysis of the samples, particularly concerning the grain-size analysis. The wet sieving of all samples is tedious so laser techniques would be more efficient. It would also require less sediment to store and analyze.

The methodology would benefit from improvements for the vegetation sample collection. Here, the below-ground biomass of the submerged vegetation was discarded to focus on the above-ground biomass. Exploring the below-ground biomass contribution to the sediment carbon content would be needed to quantify the complete influence of the vegetation on the sediment carbon pool. While it was not a problem to differentiate the above-ground and below-ground biomass of submerged species, it was more difficult to sample the common reeds. This was due to the thick and extensive root network of *Phragmites australis* and a more appropriate sampling method would facilitate the sampling process.

The biomass and cover of seagrass meadows represent a shorter time scale compared to the sediment carbon sequestration processes (Dahl et al., 2016). Thus, the actual vegetation properties of these perennial species do not fully represent the trends happening over decades or centuries (a more appropriate time scale for the carbon storage in the sediment). This assumption can also be made about the rooted-macrophyte communities of the shallow bays in the archipelago. Indeed, these vegetation communities were reported to change over time, both between years and with the gradual isolation of the bays (Appelgren & Mattila, 2005; Hansen 2013; Hill & Wallström, 2008).

Then, the impact of the above-ground vegetation on the surface carbon content might not even be representative of the first 5 cm sediment in all bays. The insufficient data concerning the history of the vegetation change of each bay make the investigation over the upper 5 cm sediment layer probably irrelevant. The comparison of these results among bays is not possible if the sediment accumulation rate is not known for each bay.

It is necessary to repeat this methodology in other bays of the archipelago to confirm that the amount of carbon stored in the upper 25 cm of sediment is independent of the openness of the bays. If a higher number of observations from various regions of the archipelago support this observation, then the total  $C_{org}$  stocks of the bays could be quantified over the entire archipelago with a spatial analysis.

It would also be required to take additional samples from shallow areas outside the bays to have a better idea of the capacity of these biotopes to capture carbon in comparison to the rest of the archipelago.

Also, it is necessary to date the sediment profile of the core so an average accumulation rate can be calculated for the different types of bay. The sediment accumulation rate would allow a better estimation of the carbon stocks over a similar time period and a more accurate comparison of these stocks between bays of different isolation stages. It would also give more details concerning the hydrodynamics processes in these ecosystems and confirm (or deny) the suggested counter effect of different hydrodynamic intensities on the amount of carbon stored in the sediment. The diversity of the numerous bays in the archipelago supports the suggestion that they might represent important sediment carbon storage on a regional scale. The shallow bays selected were of relatively small size, but larger bays can be found within the Stockholm archipelago. However, it is important to note that these bays are highly dynamic over time due to the shore-level displacement and it has to be taken into account when investigating further these carbon pools. Bays with a similar range of topographic openness were reported to represent about 700 years of shore-level displacement from the most open to the most enclosed bay (Hansen, 2013). The current isostatic land uplift of about 5 mm per year slowly isolates the semi-enclosed shallow bays from the sea, which can be described in a succession of different stages from Juvenile flad to Glo (Appelgren & Mattila, 2005; Hansen, 2013).

In the case that future research confirms the findings of this study and excluding anthropogenic impacts that could disrupt the natural processes occurring in the bays, the bays would follow a natural process of gradually being cut off from the Baltic Sea (Appelgren & Mattila, 2005; Hansen, 2013). During the succession, the water body has decreased exchanges with the surrounding seawater, coupled with a decrease in wave exposure and water circulation. There is an increase in freshwater influence until the most isolated stage of the succession. It consequently drives the shift from the dominance of marine vascular plants and algae in the less isolated stages towards the dominance of freshwater species when completely isolated.

Juvenile flads (e.g Edvassviken) are described as the less isolated transitional stage of the water body and still have significant water-exchanges with the sea. Then, flads (e.g Västra Lermaren) correspond to a delimited shallow water body that has a few narrow openings connected to the surrounding waters. Glo-flads (e.g Söderfladen, Östra Myttingeviken) still have continuous contact with the sea but their openings are overgrown by reeds.

Finally, the most isolated successional stage of a coastal lagoon is considered a Glo, which can be defined as a water basin whose opening has risen over the sea level and only has occasional contact with the sea, due to wave actions of high-water levels.

At this stage, the water body can be compared to lake environments due to the predominance of freshwater input (Appelgren & Mattila, 2005; Hansen, 2013). It would stop sequestering allochthonous carbon from marine origin, but will continue to receive organic matter from terrestrial origin.

This could indicate that without major disturbances, the bays would have an evolution of their vegetation community that would contribute to stabilizing the surface sediment until their most isolated stage as a lake. The shoreline will continue to rise and the reeds will most likely colonize the bottom that is progressively emerging from the water. The plant primary succession of seashore meadows of the Baltic coast has been studied and terrestrial vegetation could develop on the shoreline if appropriate environmental conditions for their growth are met (Ecke & Rydin, 2000). In the best scenario possible, the human impacts are minimal and the progressive evolution of the vegetation could help to stabilize and maintain the sediment as long-term carbon storage during the entire process.

However, the natural processes occurring in the bays could be disrupted by climate change effects and local anthropogenic pressures (Appelgren & Mattila, 2005; Hulisz et al., 2016).

It was reported that the vegetation zonation pattern along the Baltic coastline was associated with water fluctuations (both from sea-level and amount of water run-offs inputs fluctuations), surrounding land-use intensity, and depends mainly on soil properties (e.g moisture, salinity) and flooding events (Hulisz et al., 2016; Jutila, 2001). In this case, the predicted sea-level rise due to climate change could counteract this shore-displacement phenomenon and influence the succession stages of the bay's isolation (Hulisz et al., 2016). Additionally, grazing was shown to affect the present vegetation and can significantly decrease the abundance of reeds along the shoreline (Ecke & Rydin, 2000; Jutila, 2001). If anthropogenic pressures increase over time, then it can be assumed that the surface carbon stored in the sediment could be released back into the atmosphere.

Shallow coastal ecosystems are recognized as both net carbon storage and net emitters of  $CO_2$  to the atmosphere, especially in human-dominated areas and it is therefore needed to identify the type and extent of anthropogenic impacts that could interfere with the natural processes occurring in the bays. Their ambiguous status is a matter of controversy concerning their contribution to climate change mitigation efforts (Hori et al., 2019; Kuwae et al., 2016). The properties that characterized them as emitters of  $CO_2$  are mainly related to anthropogenic pressures such as high nutrient inputs from nearby terrestrial areas, loads of treated wastewater effluents, and the presence of underwater hypoxic areas (Hori et al., 2019). It was previously suggested that Human activities indirectly influence the carbon storage of seagrass meadows due to urbanization and erosion of terrestrial soils can increase local sediment loads or pollution (Ewers et al., 2020). Anthropogenic disturbances can also have an impact on the sink capacity of seagrass meadows through the mechanical process of particles resuspension, such as recreational boating, which occurs especially in shallow areas (Moknes et al., 2021; Serrano et al., 2014).

Marinas, boating activities, dredging, and local nutrient drainage areas constitute a few sources identified as local anthropogenic impacts for these bays (Appelgren & Mattila, 2005; Eriksson et al., 2004; Hansen, 2013). It has been estimated that in shallow and sheltered coastal areas of Sweden nearly 20% of the soft-bottom vegetation was impacted by recreational boat activities (Moksnes et al., 2021). Previous studies have started to quantify and create indicators of anthropogenic pressures for this type of bays (Appelgren & Mattila, 2005; Hansen & Snickars, 2014). Their main focus was to investigate their impacts on the flora or fauna in the bays, but it could be argued that the same indicators could be used to explore anthropogenic influence on the sediment carbon content dynamics in the bays.

## Conclusion

This study results revealed that the amount of carbon stored in the upper 25 cm of the shallow shelter bays exceeds the previous estimates for seagrass meadows of the eastern Baltic Sea area over the same sediment depth profile. The estimated carbon stocks in this study might be underestimated due to the presence of FLSM that differed between the bays, as well as, the absence of data concerning the sediment rate accumulation.

Dating the sediment profile would be needed to allow a proper comparison of the carbon stocks over time between bays of different isolation succession stages.

The results also revealed that the amount of carbon stored in the sediment was not directly reflected in the percentage of organic carbon in the sediment. An increase in sediment carbon percentage was strongly associated with a decrease in topographic opening and in sediment density, but the average carbon stocks did not display important differences between the bays. It was suggested that this pattern is due to a counter effect of the different processes driven by contrasting hydrodynamics conditions, which highlights the complexity of these coastal ecosystems. Finally, the results supported that the sediment carbon content origin was not depending on the isolation of the bay to the sea, but rather depending on their location on a larger spatial scale and their proximity to large input of terrestrial organic material.

These results were based on a restricted dataset, but these findings are promising for a first approach to explore the sediment carbon storage capacity of these shallow bays. Future research is recommended to determine whether the shallow sheltered lagoon-like bays of the Stockholm archipelago can be considered local BC ecosystems.

# Acknowledgements

I'm extremely grateful to all the people involved in my master thesis.

A thousand thanks to Sofia Wikström for her continuous support and guidance throughout this project, always with a lot of patience, valuable advice and feedback.

Thanks to Michael Tedengren for providing the laboratory equipment necessary for the samples processing and his advice on various aspects of the project.

Thanks to Mats Björk for his advice and for funding the carbon content analysis of the project's samples.

Thanks to Joakim Hansen for helping in the bays' selection process, sharing knowledge on the shallow bays of the archipelago and lending its field material.

Thanks to Emil Rydin for lending his corer device and providing a quick training on the field. Thanks to Jonas Gunnarsson for lending a stack of sieves for the grain-size analysis.

Finally, thank you to Erik Rudolfsson for helping on the field with the vegetation sampling, to Ronja and Nikki for the good atmosphere in the laboratory, and to everyone who has read the project and given me feedback.

# References

- Appelgren, K., & Mattila, J. (2005). Variation in vegetation communities in shallow bays of the northern Baltic Sea. *Aquatic Botany*, *83*, pp.1–13. Available at <<u>https://doi.org/10.1016/j.aquabot.2005.05.001</u>>
- Austin, Å. N., Hansen, J.P., Donadi, S., & Eklöf, J.S. (2017). Relationships between aquatic vegetation and water turbidity: A field survey across seasons and spatial scales. *PLoS ONE*, 12(8) : e0181419. Available at <<u>https://doi.org/10.1371/journal.pone.0181419</u>>
- Bohlin, H.S., Mörth, C.M., & Holm, N.G. (2006). Point source influences on the carbon and nitrogen geochemistry of sediments in the Stockholm inner archipelago, Sweden. *Science of The Total Environment*, 36(11), pp. 337-349. Available at <<u>https://doi.org/10.1016/j.scitotenv.2005.07.016</u> >
- Boström, C., Baden, S.P. & Krause-Jensen, D. (2003). Chapter 1: The seagrasses of Scandinavia and the Baltic Sea. *In: World atlas of seagrasses*. Edition XII, University of California Press, Berkeley. pp.27-37. Available at <<u>https://www.researchgate.net/publication/249010147</u> The seagrasses of Scandinavia and the Baltic Sea
- Chen, Y. & Xu, C. (2020). Exploring new blue carbon plants for sustainable ecosystems. *Trend in Plant Science, vol.25 (11)*, pp. 1067-1070. Available at <<u>https://doi.org/10.1016/j.tplants.2020.07.016</u>>
- Dahl, M., Deyanova, D., Gütschow, S., Asplund, M. E., Lyimo, L. D., Karamfilov, V., ... Gullström, M. (2016). Sediment Properties as Important Predictors of Carbon Storage in *Zostera marina* Meadows: A Comparison of Four European Areas. *PLoS ONE 11(12): e0167493*. Available at <<u>https://doi.org/10.1371/journal.pone.0167493</u>>
- Ecke, F., & Rydin, H. (2000). Succession on a land uplift coast in relation to plant strategy theory. Ann. Bot. Fennici, 37(3), pp.163-171. Available at <<u>https://www.jstor.org/stable/pdf/23726897.pdf?refreqid=excelsior%3Ae8f31c4ba192c35409aa177ae7</u> 19cba7&ab\_segments=&origin=>
- Emeis, K., Christiansen, C., Edelvang, K., Jähmlich, S., Kozuch, J., Laima, M., Leipe, T., Löffler, A., Lund-Hansen, L.C., Miltner, A., Pazdro, K., Pempkowiak, J., Pollehne, F., Shimmield, T., Voss, M., & Witt, G. (2002). Material transport from the near shore to the basinal environment in the southern Baltic Sea: II: Synthesis of data on origin and properties of material, *Journal of Marine Systems, 35 (3-4)*, pp.151-168. Available at <<u>https://doi.org/10.1016/S0924-7963(02)00127-6</u>>
- Eriksson, B.K., Sandström, A., Isæus, M., Schreiber, H., & Karås, P. (2004). Effects of boating activities on aquatic vegetation in the Stockholm archipelago, Baltic Sea. *Estuarine Coastal and Shelf Science*, *61*, pp.339–349. Available at <<u>https://doi.org/10.1016/j.ecss.2004.05.009</u>>
- Ewers Lewis, C., Young, M., Baldock, J., Hawke, B., Sanderman, J., Carnell, P. & Macreadie, P. (2020). Drivers and modeling of blue carbon stock variability in sediments of Southern Australia. *Biogeosciences Discussions*, 17(7), pp.1-25. Available at <<u>https://doi.org/10.5194/bg-17-2041-2020</u>>

- Hansen, J.P. (2013). Effects of shore-level displacement on the ecology of Baltic Sea bays. Swedish Nuclear Fuel and Waste Management Company (SKB), Technical report TR 13-0. Available at <<u>https://skb.se/upload/publications/pdf/TR-13-01.pdf</u>>
- Hansen, J.P. & Snickars, M. (2014). Applying macrophyte community indicators to assess anthropogenic pressures on shallow soft bottoms. *Hydrobiologia*, *738*, pp.171–189. Available at <<u>https://doi.org/10.1007/s10750-014-1928-z</u>>
- Hansen, J.P., Wikström, S.A. & Kautsky, L. (2008). Effects of water exchange and vegetation on the macroinvertebrate fauna composition of shallow land-uplift bays in the Baltic Sea. *Estuarine, Coastal* and Shelf Science, 77(3), pp.535-547. Available at <<u>https://doi.org/10.1016/j.ecss.2007.10.013</u>>

Hansen, J. P., Wikström, S. A. & Kautsky, L. (2012). Taxon composition and food-web structure in a morphometric gradient of Baltic Sea land-uplift bays. *Boreal Environmental Research*, 17, pp.1–20. Available at <<u>https://www.academia.edu/56327012/Taxon\_composition\_and\_food\_web\_structure\_in\_a\_morphomet\_ric\_gradient\_of\_Baltic\_Sea\_land\_uplift\_bays</u>>

- Hill, C.& Wallström K. (2008). Chapter 14 The Stockholm Archipelago. In: Schiewer U. (eds) Ecology of Baltic Coastal Waters. *Ecological Studies (Analysis and Synthesis)*, 197. Springer, Berlin, Heidelberg. Available at <<u>https://doi.org/10.1007/978-3-540-73524-3\_14</u>>
- Hori, M., Bayne, C. J., & Kuwae, T. (2019). Blue Carbon: Characteristics of the Ocean's Sequestration and Storage Ability of Carbon Dioxide. *In: Kuwae T., Hori M. (eds) Blue Carbon in Shallow Coastal Ecosystems*. Springer, Singapore. Available at <<u>https://doi.org/10.1007/978-981-13-1295-3\_1</u>>
- Howard, J., Hoyt, S., Isensee, K., Pidgeon, E., Telszewski, M. (eds.) (2014). Coastal Blue Carbon: Methods for assessing carbon stocks and emissions factors in mangroves, tidal salt marshes, and seagrass meadows. *Conservation International, Intergovernmental Oceanographic Commission of UNESCO, International Union for Conservation of Nature*. Arlington, Virginia, USA. Available at <<u>https://www.iucn.org/sites/dev/files/english\_blue\_carbon\_lr.pdf</u>>
- Hulisz, P., Piernik, A., Mantilla-Contreras, J., & Elvisto, T. (2016). Main Driving Factors for Seacoast Vegetation in the Southern and Eastern Baltic. *Wetlands*, *36*, pp.909–919. Available at <<u>https://doi.org/10.1007/s13157-016-0803-2</u>>
- Jankowska, E., Michel, L. N., Zaborska, A., & Wlodarska-Kowalczuk, M. (2016). Sediment carbon sink in low density temperate eelgrass meadows (Baltic Sea). *Journal of Geophysical Research: Biogeosciences*, *122 (12)*, pp. 2918-2934. Available at <<u>https://doi.org/10.1002/2016JG003424</u>>
- Jönsson, A., Lindström, M., Carman, R., Mörth, C.M., Meili, M., & Gustafsson, Ö. (2005). Evaluation of the Stockholm Archipelago sediments, northwestern Baltic Sea Proper, as a trap for freshwater runoff organic carbon. *Journal of Marine Systems*, 56(1,2), pp. 167-178. Available at <<u>https://doi.org/10.1016/j.jmarsys.2004.11.001</u>>

- Jutila, H. (2001). How does grazing by cattle modify the vegetation of coastal grasslands along the Baltic Sea? Ann. Bot. Fennici, 38(3), pp.181-200. Available at <<u>https://www.jstor.org/stable/pdf/23726759.pdf?refreqid=excelsior%3A05670fe4c8d423416de2bbe866</u> <u>887e91&ab\_segments=&origin=></u>
- Krause-Jensen, D., Lavery, P., Serrano, O., Marbà, N., Masque, P., Duarte, C.M. (2018). Sequestration of macroalgal carbon: the elephant in the Blue Carbon room. *Biology Letters*, 14: 20180236. Available at <<u>http://dx.doi.org/10.1098/rsbl.2018.0236</u>>
- Kuwae, T., Kanda, J., Kubo, A., Nakajima, F., Ogawa, H., Sohma, A., & Suzumura, M., 2016. Blue carbon in human-dominated estuarine and shallow coastal systems. *Ambio*, 45(3), pp.290–301. Available at <<u>https://doi.org/10.1007/s13280-015-0725-x</u>>
- Lavery, P. S., Mateo, M. Á., Serrano, O., & Rozaimi, M. (2013). Variability in the carbon storage of seagrass habitats and its implications for global estimates of blue carbon ecosystem service. *PloS one*, 8(9), e73748. Available at <<u>https://doi.org/10.1371/journal.pone.0073748</u>>
- López, G.I. (2017). Grain Size Analysis. In: Gilbert A.S. (eds) Encyclopedia of Geoarchaeology. *Encyclopedia of Earth Sciences Series*. Springer, Dordrecht, pp.341-348. Available at <<u>https://doi.org/10.1007/978-1-4020-4409-0\_18</u>>
- Macreadie, P. I., Anton, A., Raven, J. A., Beaumont, N., Connolly, R. M., Friess, D. A., Kelleway, J. J., Kennedy, H., Kuwae, T., Lavery, P. S., Lovelock, C. E., Smale, D. A., Apostolaki, E. T., Atwood, T. B., Baldock, J., Bianchi, T. S., Chmura, G. L., Eyre, B. D., Fourqurean, J. W., ... Duarte, C. M. (2019). The future of Blue Carbon science. *Nature Communications*, 10(1), 3998. Available at <<u>https://doi.org/10.1038/s41467-019-11693-w</u>>
- Macreadie, P.I., Jarvis, J., Trevathan-Tackett, S.M. & Bellgrove, A. (2017). Seagrasses and Macroalgae: Importance, Vulnerability and Impacts. In: *Climate Change Impacts on Fisheries and Aquaculture*. Editions B.F. Phillips and M. Pérez-Ramírez. Available at <<u>https://doi.org/10.1002/9781119154051.ch22</u>>
- Mazarrasa, I., Samper-Villarreal, J., Serrano, O., Lavery, P.S., Lovelock, C.E., Marbà, N., Duarte, C.M. & Cortés, J. (2018). Habitat characteristics provide insights of carbon storage in seagrass meadows. *Marine Pollution Bulletin 134*, pp.106–117. Available at <<u>https://doi.org/10.1016/j.marpolbul.2018.01.059</u>>
- Mcleod, E., Chmura, G.L., Bouillon, S., Salm, R., Björk, M., Duarte, C.M., Lovelock, C.E., Schlesinger, W.H. & Silliman, B.R. (2011). A blueprint for blue carbon: toward an improved understanding of the role of vegetated coastal habitats in sequestering CO<sub>2</sub>. *Frontiers in Ecology and the Environment, 9*, pp. 552-560. Available at <<u>https://doi.org/10.1890/110004</u>>
- Moksnes, P.-O., Röhr, M. E., Holmer, M., Eklöf, J. S., Eriander, L., Infantes, E., and Boström, C. (2021). Major impacts and societal costs of seagrass loss on sediment carbon and nitrogen stocks. *Ecosphere*, *12(7)* :e03658. Available at <<u>https://doi.org/10.1002/ecs2.3658</u>>
- Natural Earth (2021). Natural Earth Free vector and raster map data. Admin 0 Countries, version 5.0.0. Available at <<u>https://www.naturalearthdata.com/downloads/10m-cultural-vectors/</u>>

- OpenMapTiles (2022). Open-source maps for self-hosting, Free OpenStreetMap Vector Tiles. Available at <<u>http://openmaptiles.org/</u>>
- Ortega, A., Geraldi, N.R. & Duarte, C.M. (2020). Environmental DNA identifies marine macrophyte contributions to Blue Carbon sediments. *Limnology and Oceanography*, *65*, pp.3139-3149. Available at <<u>https://doi.org/10.1002/lno.11579</u>>
- Pempkowiak, J., Bełdowski, J., Pazdro, K., Staniszewski, A., Leipe, T., & Emeis, K.C. (2002). The contribution of the fine sediment fraction to the Fluffy Layer Suspended Matter (FLSM). *Oceanologia*, 44, pp.513–527. Available at <<u>https://agro.icm.edu.pl/agro/element/bwmeta1.element.agro-article-f3c4f721-483e-4511-a1f0-04469f</u>690837/c/444pempk.pdf>
- Pempkowiak, J., Bełdowski, J., Pazdro, K., Staniszewski, A., Zaborska, A., Leipe, T., & Emeis, K.C. (2005). Factors influencing fluffy layer suspended matter (FLSM) properties in the Odra River Pomeranian Bay Arkona Deep System (Baltic Sea) as derived by principal components analysis (PCA), and cluster analysis (CA). *Hydrology and Earth System Sciences Discussions, European Geosciences Union, 9 (1/2)*, pp.67-80. Available at <a href="https://hal.archives-ouvertes.fr/hal-00304806/document">https://hal.archives-ouvertes.fr/hal-00304806/document</a>>
- QGIS Development Team (2021). QGIS Geographic Information System. QGIS Association. Available at <<u>http://www.qgis.org</u>>
- R Core Team (2021). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0. Available at <<u>http://www.R-project.org/</u>>
- Röhr, M. E., Boström, C., Canal-Vergés, P., & Holmer, M. (2016). Blue carbon stocks in Baltic Sea eelgrass(Zostera marina) meadows. *Biogeosciences*, *13(22)*, pp.6139-6153. Available at <<u>https://doi.org/10.5194/bg-13-6139-2016</u>>
- Röhr, M. E., Holmer, M., Baum, J. K., Björk, M., Boyer, K., Chin, D., ... Boström, C. (2018). Blue carbon storage capacity of temperate eelgrass (Zostera marina) meadows. *Global Biogeochemical Cycles*, *32*, pp.1457–1475. Available at <<u>https://doi.org/10.1029/2018GB005941</u>>
- Samper-Villarreal, J., Lovelock, C.E., Saunders, M.I., Roelfsema, C. and Mumby, P.J. (2016). Organic carbon in seagrass sediments is influenced by seagrass canopy complexity, turbidity, wave height, and water depth. *Limnology and Oceanography*, *61*, pp.938-952. Available at <<u>https://doi.org/10.1002/lno.10262</u>>
- Santos, I. R., Burdige, D. J., Jennerjahn, T. C., Bouillon, S., Cabral, A., Serrano, O., Wernberg, T., Filbee-Dexter, K., Guimond, J. A., & Tamborski, J. J. (2021). The renaissance of Odum's outwelling hypothesis in 'Blue Carbon' science. *Estuarine, Coastal and Shelf Science*, 255, 107361. Available at <<u>https://doi.org/10.1016/j.ecss.2021.107361</u>>
- Scheffold, M.I.E. & Hense, I. (2020). Quantifying Contemporary Organic Carbon Stocks of the Baltic Sea Ecosystem. *Frontiers Marine Science*, 7, 571956. Available at <<u>https://doi.org/10.3389/fmars.2020.571956</u>>

Serrano, O., Lavery, P. S., Duarte, C. M., Kendrick, G. A., Calafat, A., York, P. H., Steven, A., & Macreadie, P. I. (2016). Can mud (silt and clay) concentration be used to predict soil organic carbon content within seagrass ecosystems?. *Biogeosciences*, *13*, pp.4915–4926. Available at <<u>https://doi.org/10.5194/bg-13-4915-2016</u>>

Serrano, O., Lavery, P. S., Rozaimi, M., & Mateo, M. Á. (2014). Influence of water depth on the carbon sequestration capacity of seagrasses. *Global Biogeochemical Cycles, 28*, pp.950–961. Available at <<u>https://doi.org/10.1002/2014GB004872</u>>

- Skilbeck, C.G., Trevathan-Tackett, S., Apichanangkool, P., & Macreadie, P.I. (2017). Sediment Sampling in Estuaries: Site Selection and Sampling Techniques. In: Weckström K., Saunders K., Gell P., Skilbeck C. Applications of Paleoenvironmental Techniques in Estuarine Studies. Developments in Paleoenvironmental Research, 20. Springer, Dordrecht. Available at <<u>https://doi.org/10.1007/978-94-024-0990-1\_5</u>>
- Smeaton, C., Barlow, N.L., & Austin, W.E. (2020). Coring and compaction: Best practice in blue carbon stock and burial estimations. *Geoderma*, 364, 114180. Available at <<u>https://doi.org/10.1016/j.geoderma.2020.114180</u>>
- Smith, S.V. (1981). Marine macrophytes as global carbon sink. *Science, 211(4484)*, pp.838-840. Available at <<u>https://doi.org/10.1126/science.211.4484.838</u>>
- Trevathan-Tackett, S. M., Kelleway, J. I., Macreadie, P., Beardall, J., Ralph, P., & Bellgrove, A. (2015). Comparison of marine macrophytes for their contributions to blue carbon sequestration. *Ecology*, 96(11), 150511125256001. Available at <<u>https://doi.org/10.6084/m9.figshare.c.3308118.v1</u>>
- Schiewer, U. (2008). Chapter 2 The Baltic Coastal Zones. *Ecology of baltic coastal waters*, pp.23-33. Springer Berlin Heidelberg. Available at <<u>https://link.springer.com/book/10.1007/978-3-540-73524-3</u>>
- Ulyanova, M., Sivkov, V., Kanapatskij, T., & Pimenov, N. (2014). Seasonal variations in methane concentrations and diffusive fluxes in the Curonian and Vistula lagoons, Baltic Sea. *Geo-Marine Letters*, *34*, pp.231–240. Available at <<u>https://doi.org/10.1007/s00367-013-0352-0</u>>
- Vaasma, Tiit. (2008). Grain-size analysis of lacustrine sediments: A comparison of pre-treatment methods. *Estonian Journal of Ecology*, *57*, pp. 231–243. Available at <<u>https://doi.org/10.3176/eco.2008.4.01</u>>

# Appendixes

**Appendix 1. Maps of the investigated bays, categorized with corresponding openness index (Ea).** The maps were made from Google Maps, using QGIS Software (v3.20 Odense; QGIS Development Team 2021).



# 39

## Semi-enclosed bays



Ea= 0.057

Ea= 0.101

# Open bays



Ea= 0.259





Ea= 0.591

#### <u>Legend</u>



**Appendix 2. Classification of sediment for all samples.** The results are displayed for each core, depending on the depth section and water depth, in terms of shallow or deep core. The terms were attributed depending on the proportions of gravel, sand and mud content in each sample. It follows the Folk's classification system (1968), and the Wentworth grain size classification scale (1922).

Bay	Core #	Deenness	Sediment depth interval (cm)							
Day	0010 #	Deepiless	0 - 5	5 - 12,5	>12,5					
	1	shallow	sandy mud	mud	mud					
Östra	2	shallow	sandy mud	mud	mud					
Myttingeviken	3	deep	mud	mud	mud					
	4	deep	mud	mud	slightly gravelly mud					
Eke fjärd	1	shallow	sandy mud	sandy mud	sandy mud					
	3	deep	slightly gravelly sandy mud	slightly gravelly sandy mud	mud					
	4	deep	slightly gravelly sandy mud	sandy mud	mud					
långbroviken	1	shallow	mud	sandy mud	slightly gravelly mud					
	2	shallow	mud	sandy mud	mud					
Langbrowken	3	deep	mud	sandy mud	mud					
	4	deep	gravelly mud	gravelly mud	mud					
	1	shallow	gravelly mud	gravelly mud	gravelly mud					
Söderfladen	2	shallow	mud	mud	mud					
	3	deep	mud	mud	mud					
	4	deep	mud	mud	gravelly muddy sand					
	1	shallow	slightly gravelly sandy mud	slightly gravelly mud	slightly gravelly mud					
	2	shallow	sandy mud	mud	slightly gravelly mud					
Handlatet	3	deep	sandy mud	mud	mud					
	4	octor inDespination0 - 51shallowsandy mud2shallowsandy mud3deepmud4deepmud1shallowsandy mud3deepslightly gravelly sandy4deepslightly gravelly sandy1shallowmud2shallowmud3deepmud4deepgravelly mud3deepmud4deepgravelly mud3deepmud4deepmud1shallowslightly gravelly sandy2shallowslightly gravelly sandy3deepmud4deepmud1shallowsandy mud3deepmud4deepmud1shallowsandy mud2shallowsandy mud3deepmud4deepsandy mud1shallowsandy gravel2shallowslightly gravelly sandy3deepmud4deepslightly gravelly sandy3deepslightly gravelly sandy3deepslightly gravelly sandy4deepslightly gravelly sandy3deepslightly gravelly sandy4deepslightly gravelly sandy4deepslightly gravelly sandy3deepslightly gravelly	mud	mud	mud					
	1	shallow	sandy mud	slightly gravelly sandy mud	mud					
C ≜rt-slass #sslass	2	shallow	mud	slightly gravelly mud	mud					
Famoimstiaden	3	deep	mud	sandy mud	slightly gravelly mud					
	3       deep       mud         4       deep       mud         1       shallow       sandy mud         3       deep       slightly gravelly sandy mud       slig         4       deep       slightly gravelly sandy mud       slig         1       shallow       mud       mud         2       shallow       mud       mud         3       deep       mud       mud         4       deep       gravelly mud       gravelly mud         2       shallow       mud       gravelly mud         2       shallow       mud       gravelly mud         2       shallow       gravelly mud       gravelly mud         3       deep       mud       gravelly mud         4       deep       mud       gravelly mud         3       deep       mud       gravelly sandy mud         4       deep       mud       gravelly sandy mud         3       deep       mud       gravelly sandy mud         4       deep       mud       gravelly sandy mud         3       deep       mud       gravelly sandy mud         4       deep       mud       gravelly	mud	mud							
	1	shallow	sandy gravel	gravelly mud	slightly gravelly mud					
Västra	2	shallow	slightly gravelly sandy mud	gravelly muddy sand	muddy sandy gravel					
Lermaren	3	deep	mud	mud	slightly gravelly mud					
	4	deepmudmudslightlshallowsandy mudslightly gravelly sandy mudslightly gravelly sandy mudslightly gravelly sandy muddeepslightly gravelly sandy mudslightly gravelly sandy mudslightly gravelly sandy mudslightly gravelly sandy mudshallowmudsandy mudsandy mudslightlyshallowmudsandy mudsandy muddeepgravelly mudgravelly mudgravelly mudgravelly muddeepmudmudmudgravellyshallowgravelly mudmudgravelly mudgravellydeepmudmudmudslightlydeepmudmudgravellyslightlyshallowslightly gravelly sandy mudslightlyslightlydeepmudmudmudslightlyshallowsandy mudslightly gravelly sandy mudslightlydeepmudslightly gravelly sandy mudslightlyshallowsandy mudslightly gravelly mudslightlyshallowsandy gravelgravelly mudslightlyshallowsandy gravelgravelly mudslightlyshallowsandy gravelly sandy mudslightlyslightlyshallowslightly gravelly sandy mudslightlyslightlyshallowslightly gravelly sandy mudslightlyslightlyshallowslightly gravelly gravelly muddy sandslightlyslightlyshallowslightly gravelly muddy sandsligh	mud							
	1	shallow	slightly gravelly muddy sand	slightly gravelly muddy sand	slightly gravelly sandy mud					
<b></b>	2	shallow	slightly gravelly muddy sand	sandy mud	slightly gravelly mud					
Siviken	3	deep	slightly gravelly sandy mud	sandy mud	sandy mud					
	4	deep	slightly gravelly sandy mud	slightly gravelly sandy mud	sandy mud					
	1	shallow	slightly gravelly sandy mud	slightly gravelly sandy mud	slightly gravelly sandy mud					
<b>F</b> 4 1	2	shallow	slightly gravelly muddy sand	slightly gravelly sandy mud	slightly gravelly sandy mud					
Edvassviken	3	deep	slightly gravelly sandy mud	slightly gravelly sandy mud	sandy mud					
	4	deep	slightly gravelly sandy mud	slightly gravelly sandy mud	slightly gravelly mud					
			1							

**Appendix 3. List of vegetation species presence per bay.** This table shows the records of species identified by bays, ordered from most enclosed to most open bays. The taxa recorded are a combination of results from the laboratory identification of the quantitative samples (X), sight from the boat (S) and the snorkeling surveys on the field (F). Sightings of species on the field are displayed in this table because they come from stations where no sample was collected, due to a vegetation cover estimated from 1% to 5%. The bays in bold (Söderfladen, Handfatet, Fårholmsfladen) have a higher sampling effort, since they include the syringe sampling stations. All the species identified over the syringe sampling area were also present in the other quantitative samples of the same bay, thus they haven't been specifically highlighted here.

	Östra Myttingeviken	Eke fjärd	Långbroviken	Söderfladen	Handfatet	Fårholmsfladen	Västra Lermaren	Siviken	Edvassviken
Angiosperms									
Ceratophyllum demersum	Х	Х	Х		Х	Х	F	F	
Najas marina	Х	Х	Х	Х	Х	Х		Х	
Myriophyllum spicatum		F			Х	F	F	F	Х
Phragmites australis	S	S	S	S	S	S	S	S	S
Potamogeton perfoliatus									Х
Ruppia maritima								Х	
Stuckenia pectinata	Х			X	Х	Х		Х	F
Callitriche hermaphroditica							F		
Macro-algae									
Chara baltica				X					
Chara horrida				X					
Chara tomentosa				X	Х	Х			
Chara virgata					Х				
Filamentous algae									
Cladphora sp.	Х	Х	Х	Х	Х	Х		Х	
Spirogyra sp.					Х	Х			
Ulothrix sp. (subflaccida)	Х		Х		Х	Х			
Cyanobacteria									
Lyngbya sp.	Х					Х			
Moss									
Fontinalis antipyretica			Х						

Appendix 4. Summary of sediment carbon pool values for seagrass meadows in the Baltic sea and the Skagerrak-Kattegat area. Values are presented as means over the upper 25 cm sediment layer ( $\pm$  SE for all variables; except %C<sub>org</sub> in Dahl *et al.* (2016), which is presented with  $\pm$  SD).

Locations		percent C <sub>org</sub> (% Corg)	Corg stocks (gCorg.m-2)	References
Eastern Baltic Sea	(average)	0.3 ± 0.0	578 ± 43	Röhr <i>et al.</i> (2018)
	Finland	0.24 ± 0.033	627 ± 25	Röhr <i>et al.</i> (2016)
	Sweden	0.18 ± 0.01	500 ± 50 **	Dahl <i>et al.</i> (2016)
Kattegat- Skagerrak	(average)	2.5 ± 0.6	4862 ± 741	Röhr <i>et al.</i> (2018)
	Denmark - Skagerrak	1 75 + 0 562	2644 ± 207	Döbr et el (2016)
	Denmark - Kattegat	$1.75 \pm 0.505$	6005 ± 1127	Rolli <i>et al.</i> (2010)
	Sweden	2.79 ± 0.50	3500 ± 410 **	Dahl <i>et al.</i> (2016)

\*\* values were given in gCorg.cm<sup>-2</sup> in the study of Dahl et al. (2016)

Appendix 5. Bay size and related total projected amount of carbon stored in the bay's sediment. Bays are displayed from smallest to the largest. Values are presented as means  $(\pm SD)$  over the 25 cm layer.

Вау	Area (ha)	Corg stocks (gCorg.m-2)	Projected Corg stocks per hectare (MgC.ha)	Total projected Corg stocks (MgC)	Type of bay
Handfatet	3	1413.49 ± 116.28	35.0 ± 2.3	106 ± 7	semi-enclosed
Edvassviken	4	1580.82 ± 239.76	40.1 ± 5.4	169 ± 23	open
Östra Myttingeviken	6	957.71 ± 47.43	23.2 ± 1.0	142 ± 6	enclosed
Söderfladen	7	1299.22 ± 121.46	32.4 ± 3.4	232 ± 25	enclosed
Fårholmsfladen	7	1188.51 ± 345.26	29.0 ± 8.0	209 ± 58	semi-enclosed
Siviken	8	1659.19 ± 222.11	43.4 ± 6.2	363 ± 52	open
Långbroviken	10	909.86 ± 157.90	24.4 ± 3.2	254 ± 33	enclosed
Västra Lermaren	13	1278.99 ± 558.66	33.1 ± 12.2	435 ± 160	open
Eke fjärd	19	1430.21 ± 126.99	36.6 ± 3.6	712 ± 69	enclosed

Appendix 6. Organic carbon density along the sediment profile. Bays are displayed from the most enclosed to the most open. Values are presented as means ( $\pm$  SD) of gC<sub>org</sub>.cm<sup>-3</sup> by depth sections. Depth sections represent the upper layer (0 - 5 cm), intermediate layer (5 -12.5 cm) and bottom layer (12.5 - 25 cm).



Appendix 7. Sediment and vegetation characteristics per bay. The boxplots summarize the sediment characteristics over the upper 25 cm and the above-ground characteristics, for each bay. (a) DBD (g.cm<sup>-3</sup>), (b) porosity (%), (c) organic matter (%), (d) gravel content (%), (e) sand content (%), (f) mud content (%), (g) vegetation cover (%) and (h) vegetation biomass (g). The bays are ordered from the most enclosed to the most open:

Ö.M (Östra Myttingeviken), E.K (Eke Fjärd), Lå (Långbroviken), Sö (Söderfladen),

Ha (Handfatet), Få (Fårholmsfladen), V.L (Västra Lermaren), Si (Siviken), Ed (Edvassviken).



Appendix 8.1. Correlation matrix between  $\C_{org}$  and the sediment predictor variables. The values of the correlation between the individual variables are presented on the left panels. The bivariate scatterplots are presented with a fitted line on the right panels. The histogram distribution of each variable is presented on the diagonal panels.



Appendix 8.2. Correlation matrix between  $gC_{org} cm^{-2}$  and the sediment predictor variables. The values of the correlation between the individual variables are presented on the left panels. The bivariate scatterplots are presented with a fitted line on the right panels. The histogram distribution of each variable is presented on the diagonal panels.



Appendix 9. Comparison of the regression models used to test the relationship of carbon content and sediment characteristics. The results for  $%C_{org}$  are presented in table (A) and for  $gC_{org}$ .cm<sup>-2</sup> in table (B). The regression models tested here are Linear Mixed-Effect Models (LMEM) for linear relationships and Generalized Additive Mixed Model (GAMM) with a single predictor variable, for non-linear relationships. The regressions were tested on the entire dataset (32 observations) and on the dataset excluding three outliers. The graphical results for all observations can be found in Appendix 11, while Figure 5 shows the graphical results excluding the outliers. "DF" is the degrees of freedom for LMEM and "edf" stands for the estimated degrees of freedom for GAMM. Significant p-values are written in bold.

All the samples

(n=32)

A	% Corg					% Corg						
	Predictor variable	Regression model	DF/edf	F	p-value	R-squared (adj)	R-squared	DF/edf	F	p-value	R-squared (adj)	R-squared
	DBD	LMEM	22		0		0.65	19		0		0.75
	DBD	GAMM	2.939	11.21	< 2e-16	0.83		3.251	11.360	<2e-16	0.82	
	Deresity	LMEM	22		0.0098		0.46	19		0.0635		0.32
	Porosity	GAMM	0.921	0.258	0.0108	0.30		0.724	0.124	0.0776	0.17	
	N4. d a surfacet	LMEM	22		0.0038		0.32	19		0.0128		0.17
	widd content	GAMM	0.396	0.396	0.0027	0.22		0.899	0.290	0.0103	0.13	

Outliers excluded

(n=29)

в	g Corg.cm-2					g Corg.cm-2						
	Predictor variable	Regression model	DF/edf	F	p-value	R-squared (adj)	R-squared	DF/edf	F	p-value	R-squared (adj)	R-squared
		LMEM	22		0.8981		0.03	19		0		0.71
	DBD	GAMM	2.964	8604	7.41e-06	0.57		2.126	31796	<2e-16	0.73	
	Dorocity	LMEM	22		0.6692		0.04	19		0.0195		0.35
	POIOSILY	GAMM	2.89e-06	0	1	-9.35e-08		1.054	964.2	0.0279	0.24	
		LMEM	22		0.9031		0.02	19		0.1846		0.18
	Mud content	GAMM	6.03e-06	0	1	-1.94e-07		0.534	116.9	0.1950	0.06	

Appendix 10. Relationships between carbon content (% $C_{org}$  and  $gC_{org}$ .cm<sup>-2</sup>) and the sediments characteristics. Observations represent the core mean values of carbon content and predictor variables over the 25 cm upper sediment layer. The first line shows the relationships between % $C_{org}$  and (A) DBD (g.cm<sup>-3</sup>), (B) porosity (%), (C) mud content (%). The second line shows the relationships between  $gC_{org}$ .cm<sup>-2</sup> and the predictor variables:

(D) DBD (g.cm<sup>-3</sup>), (E) porosity (%), (F) mud content (%). In each graph, the observation with symbol " $\triangle$ " is one of Edvassviken's shallow cores, while the observations with symbol " $\doteqdot$ " are the two shallow cores of Västra Lermaren.



Appendix 11.  $\delta^{13}$ C along the sediment profiles. Bays are displayed from the most enclosed to the most open one. Values are presented as means (± SD) of  $\delta^{13}$ C (‰), by depth sections. Depth sections represent the upper layer (0 - 5 cm), intermediate layer (5 - 12.5 cm) and bottom layer (12.5 - 25 cm).



Stockholms universitet/Stockholm University SE-106 91 Stockholm Telefon/Phone: 08 - 16 20 00 www.su.se

