



**Using the tea bag index to compare litter decomposition in the
sub-littoral zone of urban and vegetated coastlines in the Baltic
Sea around Helsinki, Finland**

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<i>Tiivistelmä — Referat — Abstract</i> <p>As the world urbanizes at a rapid rate, urban encroachment into coastal waters has the potential to reshape global carbon cycles by modifying organic matter decomposition processes. Litter decomposition is regulated by the litter quality, environmental conditions, and the decomposer community. This study aims to investigate if different coastline characteristics (urban/hardened versus natural/vegetated) have localized differences in decomposition rates and litter stabilization. To test this, the Tea Bag Index (TBI) has been applied to aquatic systems by including a “leaching factor” to initial masses. By using uniform litter in aquatic systems, the litter quality and moisture conditions are fixed so that other environmental conditions and the decomposer community can be considered for their impact on the rate of decomposition. Three pairs (urban and natural) of sites were selected around Helsinki, Finland in the brackish coastal water of the Baltic Sea in the summer of 2021. At each site, five green and rooibos tea bags were placed with a temperature logger, and on days 15, 29, 43, 60, and 84 a tea bag of each type was removed, dried, and weighed. Additionally, water quality measurements were collected using a YSI multiparameter sonde. There was no significant difference in the decomposition rate nor stabilization factor between urban and natural sites, although, the rate calculated according to the TBI-methodology differed from the rate determined by fitting a model to the observations. Despite no significant difference, the stabilization factors were higher than average in similar environments and are indicative of efficient litter decomposition in the waters around Helsinki. Water temperature was significantly higher, and turbidity was significantly lower at natural sites. Therefore, the cooler waters and inferred higher hydrodynamic forces at urban sites may have counteracted their individual effects on the rate of litter decomposition. In fact, since the decomposer community at natural sites is suggested to be more diverse, the rooibos tea at natural sites may have begun to stabilize, thus, implying that the study period was too long. Overall, this study found that coastlines currently have no clear localized effects on litter decomposition, but in the future, this may begin to change.</p>			
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1. Introduction

1.1 Urbanization of Coastal Waters

Currently, more than half of the world's population lives in an urban area, and it is expected to grow to greater than two-thirds by 2050 (United Nations 2019). As urban populations outgrow that of rural, environmental pressures will extend and intensify (Grimm et al. 2008). Notably, the effects are not confined to terrestrial land-use changes, but also encroach into aquatic systems. Around 40% of the global population lives in a coastal area and the number of coastal cities has continued to grow since the late 20th century (Barragán & de Andrés 2015; United Nations 2017). Consequently, humans have not only urbanized the coastal terrain around cities, but also acknowledged the peripheral footprint of urbanizing adjacent to aquatic systems. Marine urbanization includes, but is not limited, to the replacement of natural habitats with artificial structures, hardened shorelines, light and acoustic pollution, and polluted runoff of warmer freshwater.

A crucial contributor to climate change is the anthropogenic carbon dioxide (CO₂) input to the atmosphere. This flux is often concentrated in urban centers and can be assimilated into the mixing layer of water surfaces. Enhanced atmospheric CO₂ effluxes from cities has significant uptake into oceanic water up to 100 kilometers offshore, causing increased acidification (Northcott et al. 2019). As fossil fuel burning persists, greater ocean acidification is expected to occur, leaving lasting effects on marine biota (Doney et al. 2009). Furthermore, as coastal waters are increasingly net autotrophic, atmospheric CO₂ uptake and carbon burial are rising accordingly (Mackenzie, Lerman, & Andersson 2004; Najjar et al. 2018). The combined effects of physical and chemical coastline manipulation have also decreased the microbial community diversity in urban waters (Alter et al. 2021; Momota & Hosokowa 2021). Overall, the impression of humans in coastal systems may have lasting biogeochemical consequences compared to their natural state.

1.2 Litter Decomposition

Climate warming has enhanced and accelerated decomposition rates (Petraglia et al. 2018; Trevathan-Tackett, Broderson, & Macreadie 2020). Decomposition in ecosystems is the driver of biogeochemical cycles that recycle and transport carbon stored in organic matter. The rate and efficiency of decomposition is controlled by three aspects: litter quality, environmental conditions, and the decomposer community (Coûteaux, Bottner, & Berg 1995). First, plant litter quality is dependent on the physical and chemical composition, most notably, the lignin,

phosphorus, nitrogen, and tannin content (Webster & Benfield 1986). In aquatic systems, diverse litter input is necessary to maintaining macroinvertebrate decomposer diversity (Leroy & Marks 2006). Next, moisture and temperature account for up to 70% of remaining mass variation, and rates of decomposition are highest in warmer and moister environments (Berg et al. 1993; Parton et al. 2007; Petraglia et al. 2018). Third, decomposer community diversity is necessary for efficient decomposition because its complexity positively correlates with decomposition rates in aquatic systems (García-Palacios et al. 2016; Handa et al. 2014; Schindler & Gessner 2009). To separate the environmental and biological characteristics from the litter quality, litter decomposition has been standardized through the Tea Bag Index (TBI; Keuskamp et al. 2013). This index utilizes a uniform method of studying decomposition in various environments by removing the discrepancy of alien litter quality with, for instance, Lipton tea bags. By using two different tea types with different organic matter quality, one could determine a stabilization factor (S) and decomposition rate (k, constant) for said system (Keuskamp et al. 2013).

While the TBI was initially intended for studying terrestrial decomposition, it has since been applied in marshes (Mueller et al. 2018; Trevathan-Tackett et al. 2021), freshwater streams, (Hunter, Williamson, & Sarneel 2021), lakes (Seelen et al. 2019), and oceans (Mori et al. 2021). Terrestrial litter decomposition occurs in two phases; first, the easily degradable compounds are decomposed at a quick rate of mass-loss, then, in phase two, the recalcitrant part, such as lignified material, begins to break down at a slower rate compared to phase one (Coûteaux, Bottner, & Berg 1995). However, in aquatic systems these processes are preceded by the rapid leaching of the tea's water-soluble components. To account for the initial rapid loss by leaching, a "leaching factor" must be applied to the initial mass of the tea bags when using the TBI in aquatic systems (Blume-Werry et al. 2021; Edwartz 2018; Mori et al. 2021; Pouyat et al. 2017; Seelen et al. 2019). With that applied, Seelen et al. (2019) argues that the TBI is suitable for assessing aquatic decomposition.

Aquatic systems provide peculiar conditions to study decomposition because they eliminate the effect of variable moisture content between sites. This creates ideal circumstances to study the effects of other environmental conditions, such as temperature, acidity, and the microbial community on decomposition rates (Boyero et al. 2011). In significantly acidic waters and soils, both the decomposer activity and colonization of plant litter can decrease, which lowers the rate of decomposition (Batty & Younger 2007; McKinley & Vestal 1982). In contrast, research has found that decomposition rates increase in warmer environments (Ferreira & Chauvet 2011; Petraglia et al. 2018; Trevathan-Tackett, Broderson, & Macreadie

2020) and that the rate of degradation exhibits a Q_{10} (temperature coefficient) of approximately 2-5 based on litter lability (Conant et al. 2008; De Beer et al. 2019; Dossou-Yovo 2021; Kätterer et al. 1998). Lastly, diverse and complex aquatic microbial decomposer communities correlate to a higher decomposition rate compared to less diverse communities (García-Palacios et al. 2016; Santschi et al. 2018). Understanding aquatic litter decomposition is important in establishing the link between terrestrial organic matter and the marine carbon cycle.

1.3 Baltic Sea Characteristics

In the Baltic Sea, climate change and urbanization have affected the biogeochemistry of its waters. Since 1990, the sea surface temperature of the Baltic has risen more than 2 °C, and the pH has decreased by 0.15 units from pre-industrial era levels (Reckermann et al. 2014; Schneider et al. 2015). Although the high alkalinity of the Baltic Sea has offset large annual changes in pH, anticipated rises in atmospheric CO₂ will further decrease the seawater pH level (HELCOM 2021; Schneider et al. 2015). More specifically, the loss of vegetation in urban coastlines will lower pH levels by altering the relationship between respiration and primary production (Duarte et al. 2013). By the end of the 21st century, the Baltic Sea is expected to undergo further acidification, rises in temperature, and based on certain scenarios, increased nitrogen fluxes 2-72% the current input (Reckermann et al. 2014; Schneider et al. 2015). Around Helsinki in the Baltic Sea (Gulf of Finland), total dissolved nitrogen in coastal urban waters peaked and has begun to decrease following advancements in wastewater treatment (Weckström 2006). A long-term effect of the external nitrogen and phosphorus loading has been high phosphorus internal loading and a positive feedback loop of frequent algal blooms, further nitrogen removal, and hypoxic zone expansion (Vahtera et al. 2007). Furthermore, climate change has intensified resuspension events in the shallow waters of the Gulf of Finland, prompting fluxes of both dissolved inorganic and organic nutrients into water column (Niemistö & Lund-Hansen 2019). While planktonic diatom species richness has declined over the last two centuries in urban areas, general phytoplankton species richness has increased in the greater Baltic Sea and can be attributed to long-term environmental change (Olli et al. 2014; Weckström 2006). As climate change and urbanization intensify, the Baltic Sea will be prone to an amplification of increased temperature, increased variation in pH, and nutrient fluxes.

1.4 Purpose of Research & Hypotheses

In this study, I investigate how decomposition in the sublittoral zone compares between hardened artificial coastlines and naturally vegetated coastlines around Helsinki, Finland using the TBI adapted for aquatic systems. I hypothesize that a higher decomposition rate and stabilization factor will be found at urban sites along hardened coastlines because the water temperature is expected to be higher and have less other organic matter compared to vegetated coasts. Therefore, I expect that the decomposer community will be efficient at decomposing a high degree of the introduced organic matter. This is compared to the naturally vegetated sites which I suspect will have a lower decomposition rate due to an abundance of available organic matter and cooler temperatures. Additionally, a goal of this study is to assess the applicability of the TBI to study decomposition in coastal brackish systems.

2. Materials & Methods

2.1 Study Area

This study was conducted in the coastal area around Helsinki, Finland (60.14°N – 60.19°N, 24.82°E – 25.08°E) in the summer of 2021 (Fig. 1). Geographically, the area is an archipelago with around 300 islands and has a varying coastline with several larger bays of restricted water exchange. Salinity typically varies between 0-6 PSU depending on distance from the coastline and water depth (Nyman 2022).

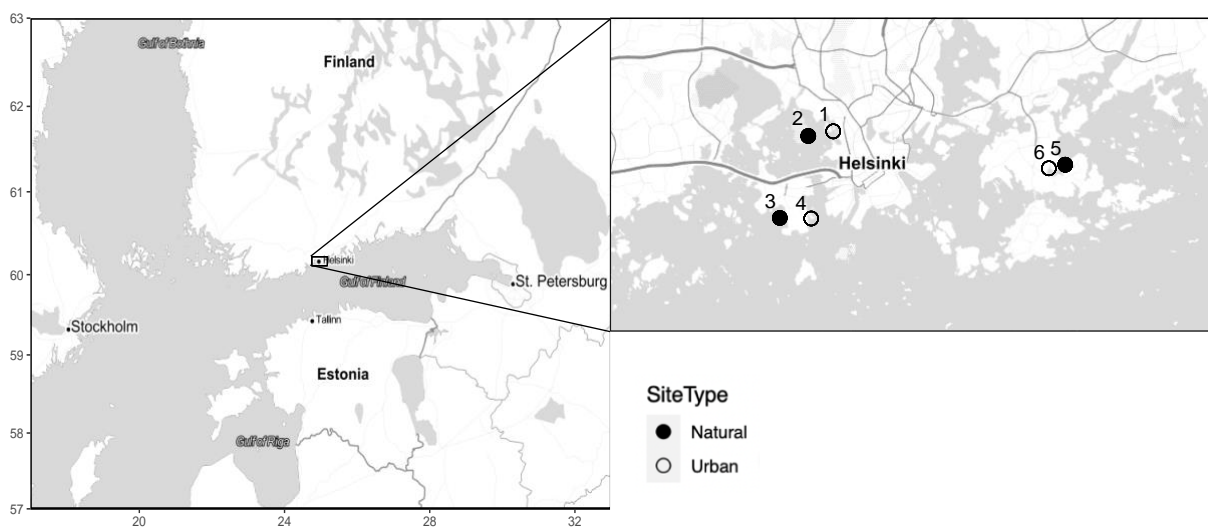


Figure 1. A map of the study area around Helsinki, Finland in the Gulf of Finland, Baltic Sea. Each point is labeled with the site number. Filled black circles indicate natural sites while empty circles indicate urban sites.

Initially, four pairs of sites were selected, but over the field season, one pair of sites was tampered with, and the samples were lost. The sites were selected visually, based on the composition of the nearby shoreline (built/urban versus natural/vegetated), and the pairs were placed within 1 km of each other. All sites were within 5 m of the shoreline and accessible via boat. No sites were in the outlet of the Vantaa River to avoid influence by the high output of organic matter.

2.2 Experimental Design

Synthetic Lipton Indonesian Sencha Green (EAN 87 22700 05552 5) and African Rooibos Infusion (EAN 87 11327 46168 0) pyramid tea bags were used to measure the rate and degree of decomposition (Keuskamp et al. 2013). Prior to placement, each tea bag was labelled, and its dry mass was recorded. At every site, five bags (non-rinsed) of both tea type were placed into an 11-gram fine mesh polyester drawstring bag (intended for produce, EAN

64 15716 79510 4) along with a Vemco Minilog-II-T temperature logger. The mesh bags were labeled with the appropriate site number, and were either directly attached to a stationary buoy or connected to a floating pier via a string extension. The bags were situated so that they were submerged into the water column approximately one meter below the surface. Initial deployment (day 0) was on June 24, 2021. On days 15, 29, 43, and 60 of the experiment, one tea bag of each type was removed from the site, and on day 84, the last tea bags and temperature logger were removed. During collection days, a YSI 6600 V2 water quality sonde, equipped with turbidity, chlorophyll-a, phycocyanin, temperature, and conductivity sensors was used to collect point measurements for each site. After each collection date, the tea bags were gently rinsed to remove any sediment on the outside of the bag, air dried for at least one week, then weighed to the nearest 0.01 gram. Throughout the field period, the labels on the tea bags began to rub off, so they were re-labelled using sharpie on masking tape around the string. The mass of the masking tape was subtracted from the final dry mass of the tea bags.

2.3 Calculations

2.3.1 Tea Bag Index

The purpose of using two different types of tea was to calculate both the decomposition constant, k , and the stabilization factor, S , according to Keuskamp et al. (2013). The calculations are designed for one time step, but this study completed a time series of the relative mass loss. Then, a comparison of the one-time-point method against constants derived by fitting regression equations to the data can be made. First, the green tea was used to calculate the stabilization factor using equation [1].

$$[1] \quad S = 1 - \frac{a_g}{H_g}$$

Where S is the stabilization factor, a_g is the decomposable fraction (final/initial weight), and H_g is the hydrolysable fraction of green tea. The hydrolysable fractions (mean \pm SD) for green (0.842 ± 0.023) and rooibos tea (0.552 ± 0.050) were based off the tea quality parameter assessment from Keuskamp et al. (2013). Using the stabilization factor from the green tea, I then calculated the decomposable fraction of the rooibos tea (a_r) using equation [2] since the labile material will still be decomposing at the time of removal.

$$[2] \quad a_r = H_r (1 - S)$$

Where a_r is the decomposable fraction of rooibos tea and H_r is the hydrolysable fraction of rooibos tea. This equation implies that S is the same for each tea. Then, with a_r and W_{rt} (the remaining litter mass in grams of rooibos tea at time t) known, the decomposition rate, k , for the rooibos tea can be calculated using equation [3; Hunter, Williamson, & Sarneel 2021].

$$[3] \quad k_{TBI} = \frac{\ln\left(\frac{a_r}{W_t}\right)}{t}$$

Where W_t is remaining litter mass in grams after time t , k is the decomposition rate constant, and a_r is the decomposable fraction calculated in equation [2].

2.3.2 Trophic Status

To compare trophic status effects on decomposition rates with Seelen et al. (2019), each site was assigned a trophic state based on the trophic state index (TSI; Carlson 1997). Although the TSI was designed for, and primarily used to measure the trophic status of inland waters (Carlson 1997), this provides a means to compare results with the previous work from Seelen et al. (2019). Since no direct Secchi disk depth (SD) measurements were made, the turbidity (FNU) measurements from the YSI were transformed into an equivalent SD (feet) using equation [4; USGS 2016].

$$[4] \quad SD = 11.123 * Turbidity^{-0.637}$$

Then, the calculated SD was converted to meters and used to estimate the TSI using equation [5].

$$[5] \quad TSI = 10 \left(6 - \frac{\ln SD}{\ln 2} \right)$$

Based on the TSI, a site was either classified as oligotrophic (<50) or eutrophic (>50; Seelen et al. 2019).

2.4 Leaching Factor

To accommodate for the rapid loss of initial mass, a leaching factor is needed to be applied to the tea bag masses. Multiple studies have previously assessed the need for a leaching factor to be applied to the mass of the tea bags (Blume-Werry et al. 2021; Edwartz 2018; Mori et al. 2021; Pouyat et al. 2017; Seelen et al. 2019). While Seelen et al. (2019) found that 21.8% and 9.0% of green tea and rooibos tea, respectively, was leached from the initial mass over 3 hours, the other studies noted higher numbers that are closer in relation to the estimation of the water soluble fraction for the tea types from Keuskamp et al. (2013). Therefore, I opted to use a leaching factor of 40% and 20% to apply to the masses of green and rooibos tea, respectively, as suggested by Blume-Werry et al. (2021), Edwartz (2018), Mori et al. (2021), and Pouyat et al. (2017). Choosing an appropriate leaching factor is necessary to gauge the correct mass loss that occurs prior to decomposition. If the leaching factor is underestimated, the rate of mass loss and stabilization factor would be larger than actuality, while an overestimated leaching factor would decrease the rate and stabilization values.

The leaching factor was applied as described by Seelen et al. (2019). First, the initial mass was multiplied by the leaching factor and subtracted from the initial mass. Then, the hydrolysable fraction for green (H_g) and rooibos (H_r) tea needed to be adjusted since the fraction is dependent on the initial mass. To do so, the leaching factor was subtracted from the hydrolysable fraction (from Keuskamp et al. 2013) for each tea type and then divided by 1-leaching factor. These adjusted values were then used in the final calculations.

2.5 Data Analyses

In this study, there were two independent variables (urban and natural sites) that were repeatedly sampled for their environmental data over the duration the study period. Therefore, a repeated measures ANOVA was used to compare data collected by the YSI over the sampling periods at urban and natural sites. Additionally, a t-test was used to compare means of the water temperature (from the logger) and the calculated decomposition rate and stabilization factor of the tea bags between urban and natural sites. To ensure the correct test was used, the YSI data were checked first for normality using the Shapiro-Wilk test followed by an outlier test to ensure there were no extreme outliers. The decomposition rate, stabilization factor, and logger water temperature were also checked for normality, but then followed by an F-test for variance. Where the data fit the requirements (i.e., normal distribution and same variances or no extreme outliers), a Two-Sample t-test or repeated measure ANOVA was performed. The YSI data that

did not fit the requirements (water temperature and turbidity) underwent a log transformation to meet normality. The decomposition and stabilization factor also did not meet normality due to the small sample size, so Wilcoxon Rank Sum Test was performed on those variables. All analyses and visualizations were carried out on RStudio version 1.3.1073 using the following packages: stats v4.0.2, rstatix v0.6.0, ggplot2 v3.3.5, ggmap v3.0.0 (Kahle et al. 2019, Kassambara 2021, R Core Team 2022, Wickham et al. 2021).

3. Results

3.1 Environmental Variables

The YSI water quality data analyzed resulted in two variables, turbidity (FNU) and TSI, having a significant higher value at urban compared to natural sites (Table 1; $p=0.002$). At the urban sites, the mean turbidity and TSI were $4.61 (\pm 1.54 \text{ SD})$ and $55.8 (\pm 6.43 \text{ SD})$, respectively, while the natural sites had means of $3.02 (\pm 2.11 \text{ SD})$ and $50.5 (\pm 3.60 \text{ SD})$, respectively (Table 1). Based on the TSI values, only one site, site 3 (natural), had a mean value within the oligotrophic range, while the other sites were above the eutrophic status threshold (Fig. 2). Additionally, the water temperature ($^{\circ}\text{C}$) recorded by the loggers obtained a significantly higher temperature at natural sites ($p<0.001$; Table 1; Fig. 3). The mean water temperature at urban sites was $17.4 (\pm 3.98 \text{ SD})$ while the natural sites had a mean temperature of $17.9 (\pm 4.02 \text{ SD})$; Table 1). The beginning of the summer of 2021 was exceptionally warm compared to previous years, and following day 20 (July 14, 2021), a rapid cooling period can be seen (Fig. 3).

Table 1. Comparison of environmental variables for urban ($n=3$) and natural ($n=3$) sites. Listed are the mean values \pm standard deviation and the p -values. Asterisks indicate significance level of the difference between site types (* $p<0.05$, ** $p<0.01$, *** $p<0.001$).

	Urban Sites	Natural Sites	<i>p</i> -value
	Mean \pm SD	Mean \pm SD	
Logger Water Temperature ($^{\circ}\text{C}$)	17.4 ± 3.98	17.9 ± 4.02	0.000***
YSI Water Temperature ($^{\circ}\text{C}$)	16.5 ± 4.28	16.2 ± 4.07	0.657
pH	7.96 ± 0.28	7.92 ± 0.35	0.477
Turbidity (FNU)	4.61 ± 1.54	3.02 ± 2.11	0.002**
Trophic State Index (TSI)	55.8 ± 6.43	50.5 ± 3.60	0.002**
DO (%)	92.0 ± 9.82	90.1 ± 12.6	0.459
DO (mg/l)	8.71 ± 0.77	8.63 ± 1.38	0.638
SPC ($\mu\text{g}/\text{cm}$)	9252 ± 309	9282 ± 314	0.749

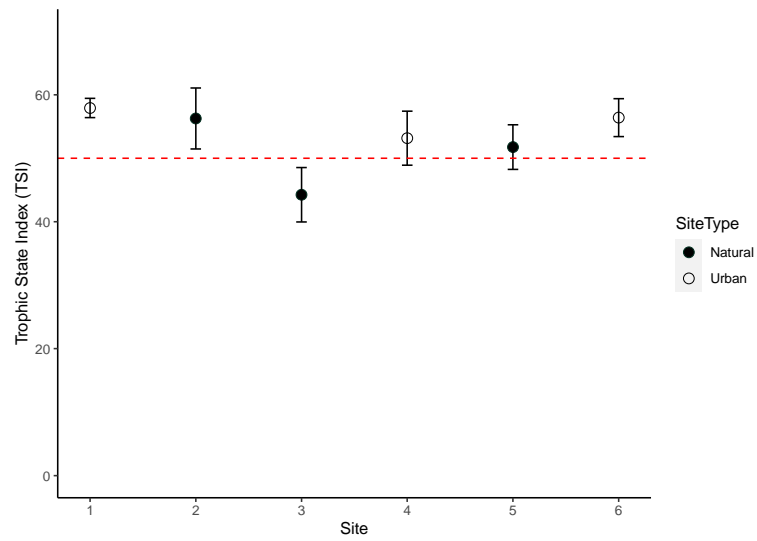


Figure 2. The trophic state index for natural (black points) and urban (empty points) sites. Below the dashed red line (TSI = 50) is considered an oligotrophic state and above the line is considered eutrophic. Site numbers are referring to figure 1. Error bars are standard deviation.

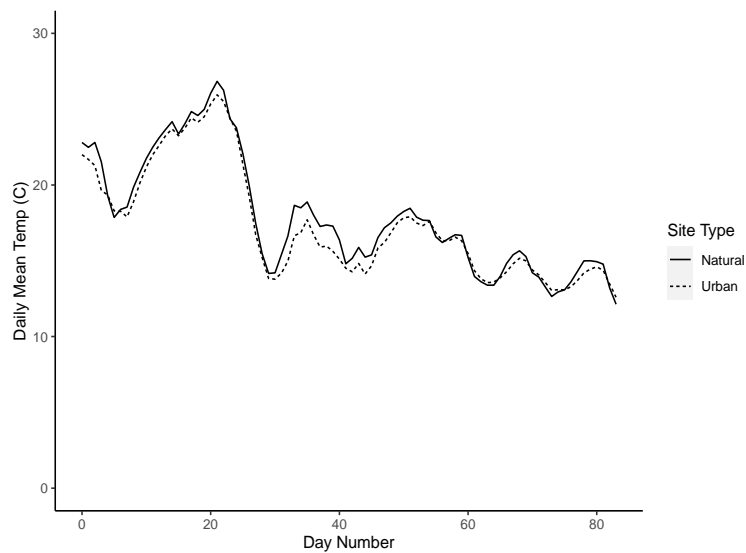


Figure 3. A line plot of the daily mean water temperature (°C) in natural sites (solid line) and urban sites (dashed line) over the duration of the experiment (84 days).

3.2 Tea Bag Decomposition

Without a leaching factor applied, both the rooibos and green tea underwent a rapid rate of initial mass loss before plateauing on day 20 at around 70% and 30% relative remaining mass, respectively (Fig. 4a). In comparison, the mass loss curves with the leaching factors applied depicts a stabilization of the green tea around 50% relative remaining mass on day 40 (Fig. 4b). The rooibos tea at urban sites decayed in a linear rate, while it appeared to stabilize around 85% relative remaining mass on day 40 at the natural sites (Fig. 4b).

The decomposition rates fit to the model with the leaching factor were not significantly different, yet the model prescribed a decomposition rate nearly ten times larger for natural sites (0.0363 ± 0.0215 SE) compared to urban sites (0.00382 ± 0.00369 SE; Table 2). There was also no significant difference between urban and natural sites when using the TBI calculation (eq 3), with a decomposition rate of $0.0180 (\pm 0.00028$ SE) and $0.0189 (\pm 0.00046$ SE), respectively (Table 2). Lastly, there was no significant difference between site types when comparing the calculated stabilization factor (Table 2).

Table 2. Both the calculated and fitted decomposition rates, as well as the calculated stabilization factor for urban and natural sites. Listed are the mean values \pm standard error and the p-values.

	Urban Sites	Natural Sites	
	Mean \pm SE	Mean \pm SE	<i>p</i> -value
Calculated Decomposition Rate ($k \cdot 10^3$)	18.0 \pm 0.28	18.9 \pm 0.46	0.166
Fitted Decomposition Rate ($k \cdot 10^3$)	3.82 \pm 3.69	36.3 \pm 21.5	0.078
Stabilization Factor (<i>S</i>)	0.39 \pm 0.02	0.39 \pm 0.03	0.773

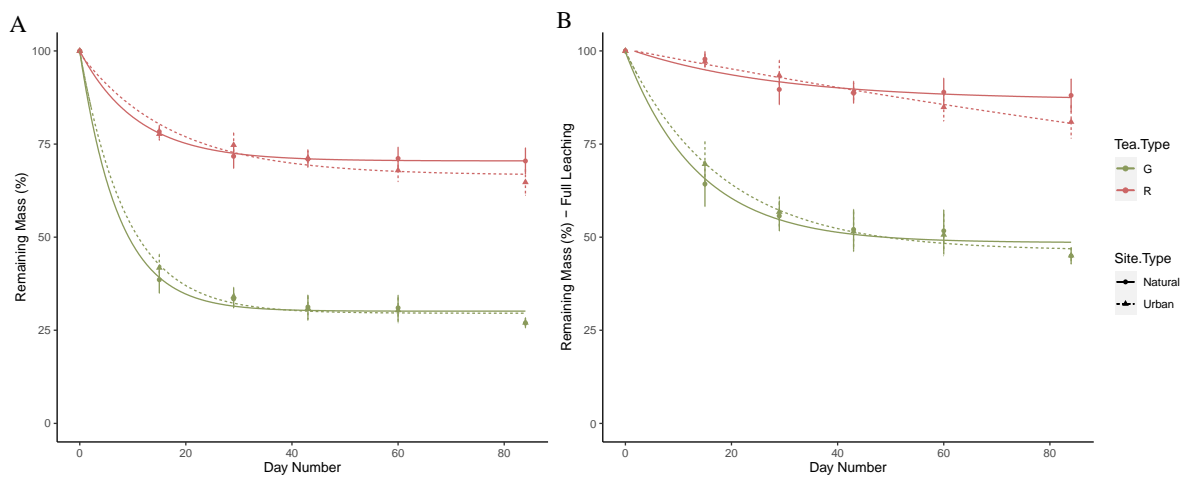


Figure 4. Plots of remaining mass (%) of green (green lines) and rooibos (red lines) tea bags in urban (triangles) and natural (circles) sites during the length of the experiment (84 days). (a) The remaining mass with no alterations to values. (b) The relative remaining mass after factoring out the leaching factor. Each point represents the day tea bags were removed. Vertical lines from each point are the standard deviation. The lines are fitted using eq. [3] with dashed lines representing urban sites and solid lines indicating natural sites.

4. Discussion

The findings of this study do not corroborate with the hypothesis because there was no significant difference in the decomposition rate of the tea bag litter between site type. Although not significant, the difference between the decomposition rate fit to the model at urban and natural sites is approaching significance (Table 2). While the natural sites do have significantly higher water temperature and significantly lower turbidity and trophic status, the site types do not exhibit different rates of decomposition or stabilization factors (Fig. 2; Fig. 3; Table 1). Hence, the reason for similarity between decomposition characteristics despite different environmental conditions is worth further investigation.

4.1 Tea Bag Index

The TBI has been applied in aquatic environments, albeit, with some dispute about the validity of the results (Mori et al. 2021; Seelen et al. 2019). First, in the current study there is a stark visual difference between the decomposition curves with and without the leaching factor applied (Fig. 4). Since leaching is an influential first step in aquatic litter decomposition, my results suggest that a leaching factor must be included in the calculations of the TBI to ratify the decomposition curve. Without the leaching, there is a stabilization in the rooibos tea for both site types, leaving the TBI calculations invalid (Fig. 4a). The large leaching factor discrepancy between Seelen et al. (2019) and other studies was most likely due to differences in their methods. Where Seelen et al. (2019) only conducted their leaching experiment for 3-hours, others either waited up to 72-hours until the rapid leaching period stabilized (Edwartz 2018) or until all the water-soluble mass was gone (Pouyat et al. 2017). Next, while Seelen et al. (2019) found the TBI was suitable for lake systems, Mori et al. (2021) disagreed with their sentiment in freshwater systems. However, decomposition in saline systems performed more appropriate with the TBI (Mori et al. 2021). In this study, I analyzed litter decomposition in a brackish water system, so would expect the results to fall between the freshwater and saline results of previous studies. Since the decomposition curve shows a stabilization of the green tea decomposing, but not the rooibos tea at urban sites (Fig. 4b), the requirements for the TBI are fulfilled. With that in mind, the decomposition curve demonstrates that this is a suitable method for studying litter decomposition in brackish coastal waters.

4.1.1 Stabilization Factor & Decomposition Rate

The stabilization factor and the decomposition rate give insight into how this system varies from others, and the effects of the environmental and biological conditions at each site type. Foremost, the stabilization factor from this experiment falls as an outlier compared to Keuskamp et al. (2013) and Seelen et al. (2019), but within the range of other studies (Table 2; Fig. 5; Mueller et al. 2018; Petraglia et al. 2018). Compared to European lakes and terrestrial systems, S in this study is approximately two times as large, indicating that a higher degree of the labile fraction was decomposed (Fig. 5). Only one location from Keuskamp et al. (2013) had a higher S than this study (Fig. 5, Ecosystem 8). However, compared to marsh ecosystems, snowbeds, grasslands, and deciduous and coniferous forests at similar temperatures as the study sites, the calculated S is only slightly above the average (Mueller et al. 2018; Petraglia et al. 2018). Since S specifies the degree of decomposition, it is inferred that decomposition of hydrolysable organic matter is efficient in the Helsinki coastal sites selected for this study. Thus, releasing a large proportion of the stored carbon. S has been negatively correlated to temperature (Keuskamp et al. 2013), so despite higher-than-average temperatures in Helsinki throughout the experiment, the S remaining high denotes an efficient system. The marine carbon reservoir is the largest on Earth, and unless decomposed first, coastal ecosystems are a gateway to burial for plant litter. This emphasizes the significance of the stabilization factor on global carbon budgets.

Next, k is also within the range of previous studies, but there is some discrepancy between the TBI calculations and the values derived from the model. The k values fall within the range of values from Keuskamp et al. (2013) and are less than what was found in European lakes by Seelen et al. (2019; Fig. 5). The TBI calculations of k show no significant difference between the site types, which supports the null hypothesis (Table 2; Fig. 5). However, the model fit to the time series data shows that the rate of decomposition at urban sites is slightly slower and more linear, while natural sites have a rapid initial loss (Fig. 4b). In fact, the rooibos tea at natural sites may have begun to stabilize, invalidating the calculations for that site type, and implying that the study period for the TBI in that environment needs to be shortened. This fact could only be seen with the full time series plotted, and it would have been missed with just the TBI's one-time-point calculation. Thus, bringing attention to a drawback of using only the calculation to estimate the decomposition rate. Assuming that the modelled k for natural sites is valid, it is approaching a significantly higher rate than urban sites. As previously mentioned, aquatic environments allow for ideal circumstances to study the effects of temperature on decomposition rates, and here we see that natural sites had both a significantly

higher temperature (Table 1), as well as a fitted decomposition rate nearly ten times higher than urban sites (Table 2).

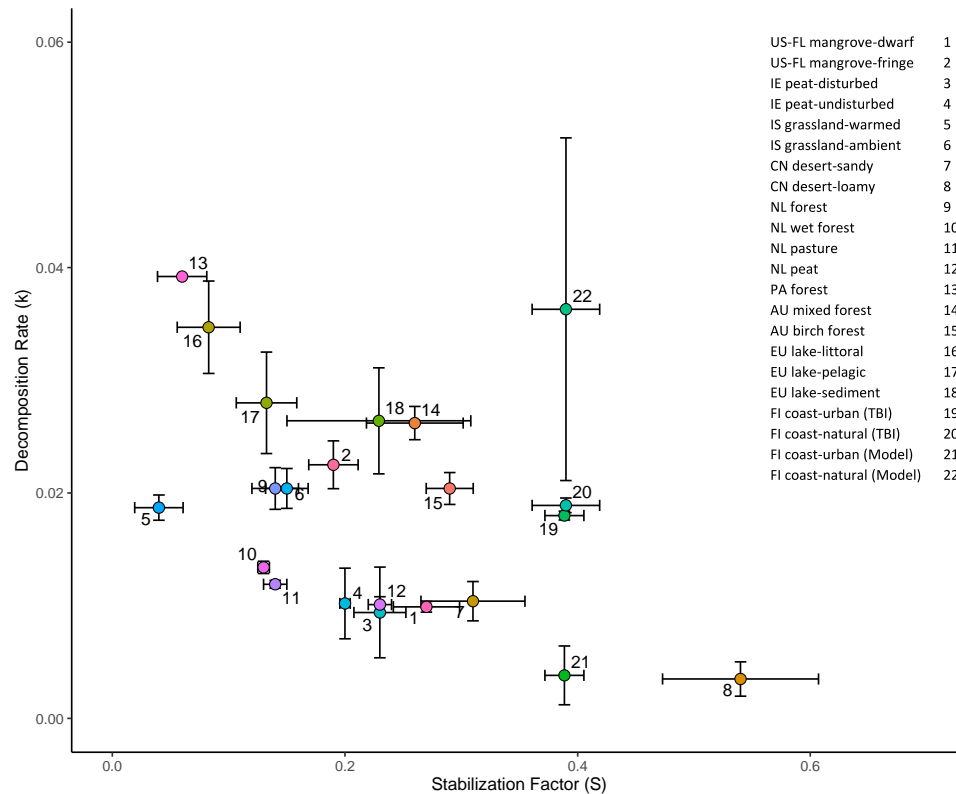


Figure 5. The decomposition rate (k) and stabilization factor (S) for terrestrial soil ecosystems (1-15, Keuskamp et al. 2013), lake ecosystems (16-18, Seelen et al. 2019), and brackish coastal ecosystems (the current study; TBI:19-20, Model:21-22). Error bars are standard error.

4.2 Environmental Conditions

Since the difference between the fitted decomposition rates at urban and natural sites is approaching significance, the environmental conditions likely explain this (Fig. 4b). The only environmental variables that were significantly different between site types were the water temperature, turbidity, and trophic state index (Table 1). Higher temperatures correlate to a higher litter decomposition rate in aquatic environments (Ferreira & Chauvet 2011; Follstad Shah et al. 2017) and temperature explains much of the variation in litter decomposition rates (Djukic et al. 2018; Mueller et al. 2018; Petraglia et al. 2018). Overall, both site types experienced higher water temperatures compared to previous years, and here a few reasons can be theorized why urban sites had a significantly lower temperature yet similar decomposition rate as natural sites. First, the urban sites have little protection from open ocean exposure (hydrodynamic forces) compared to the naturally vegetated sites. Exposure to the

hydrodynamic forces decreases water temperature, which has a negative effect on the rate of decomposition. Conversely, high levels of hydrodynamic forces were found to increase the decomposition rate of plant litter (Costa et al. 2019). Considering the linearity of the urban decomposition rate, I speculate that the effects from the hydrodynamic forces and lower water temperatures counteracted and resulted in a steady rate of mass loss insignificantly different from the natural sites.

Lastly, due to the often adverse environmental conditions, urban sites are surmised to have less diverse microbial communities (Alter et al. 2021; Momota & Hosokowa 2021). Due to the fine mesh of both the tea bags and the drawstring bags they were placed in, meso- and macrofauna were excluded from accessing the tea litter, so only the microbial decomposer community could contribute to the decomposition (Ferreira et al. 2016; García-Palacios et al. 2016). Since community diversity is another prominent factor in decomposition (García-Palacios et al. 2016; Santschi et al. 2018), a less diverse community and lower decomposer abundance would also negatively influence the decomposition rate. At the natural sites, the microbial community is assumed to be more diverse and abundant, which could explain the rapid initial decomposition rate. Combined with higher temperatures, it would make sense that the rooibos tea at natural sites stabilized rapidly. While it is not possible to pinpoint one specific causation, temperature and microbial community are likely the key contributing factors to the different shapes of the decomposition curves (Fig. 4b).

4.2.1 Trophic State Index (TSI)

Eutrophication has already exerted pressures on the biogeochemistry of the Baltic Sea (HELCOM 2021). The TSI for urban and natural sites supports the notion that urban sites show more signs of eutrophication and habitat disturbance than vegetated sites (Fig. 2; Table 1). Advancing eutrophication has been shown to increase the rate of decomposition (Deegan et al. 2012), especially for plant litter with lower initial nutrient content (Gulis, Ferreira & Graça 2006). Since this study found that urban sites have a significantly higher eutrophication status (as indicated by the TSI) than natural sites, it is expected that the decomposition rate at urban sites would have been higher as well. Although a significant difference between the TSI at urban and natural sites was observed, the mean of both site types falls within the eutrophic regime (Fig. 2). Seelen et al. (2019) found that eutrophic lakes, according to the TSI, have higher stabilization factors. However, in this study there was no significant difference between stabilization factors at urban and natural sites, despite urban sites having a significantly higher TSI (Table 1).

Since the eutrophication value was not determined through direct measurement of nutrients, rather turbidity, it must be treated with caution. Turbidity is a measure of sediment suspension deflecting rays of light, so in turbulent waters with high sediment content, the turbidity can be high despite a low availability of nutrients. A similar issue may have arisen in the urban waters of Helsinki. The exposure to hydrodynamic forces at urban sites results in resuspended sediments (Niemistö & Lund-Hansen 2019) which lowers vegetation and increases disturbances. On the contrary, at natural sites there is more root biomass to solidify the sediment, prevent resuspension, and foster photosynthesis. If the measurements using water turbidity are correctly indicative of eutrophication status, other factors (temperature and microbial community) must be more influential in determining the decomposition rate and stabilization factor. To check this, data from three surface water quality monitoring stations near each pair of sites (Nyman unpublished) were compared to this study. Sites 1 and 2 exhibited a mean TSI indicative of eutrophic status, while the other pairs of sites had an oligotrophic mean TSI. However, the TSIs ranged from approximately 45 to 55, a similar range to the results from this study. Ideally, direct nutrient measurements should be made in tandem with the tea bag removal to ensure proper interpretation of eutrophication status, but in this case, the conversion from turbidity to TSI was an adequate substitution.

5. Conclusion

In the future, climate change is expected to alter the chemical and physical attributes of water. By conducting this study in an urban landscape that is already under the stress of increased temperatures and nutrient loading (HELCOM 2021), I gained insight into how aquatic decomposition may respond to these future changes. First, the changes to litter decomposition have implications for the global carbon cycle. Organic matter that enters marine systems can either be decomposed and recycled, or enter the marine carbon reservoir (Macreadie et al. 2017). When plant material is decomposed, CO₂ is released, so significant changes to coastal environmental conditions and decomposer communities will alter the flux characteristics. At all the sites, I calculated higher than average stabilization factors (Fig. 5), which are indicative of higher CO₂ fluxes in the coastal waters and atmosphere. This, combined with the already elevated atmospheric CO₂ in urban centers, can further acidify urban coastal waters (Northcott et al. 2019) and potentially result in a positive feedback loop that acidifies waters, increases the rate and degree of decomposition, and then fluxes more CO₂ into the water and atmosphere. Additionally, continued urbanization of coastal waters will increase the level of hydrodynamic forces by removing vegetation that would typically provide refuge from the open ocean (Guichard, Bourget, & Robert 2001). Higher hydrodynamic forces (Costa et al. 2019), along with increases in temperature (Ferreira & Chauvet 2011; Macreadie et al. 2017; Trevathan-Tackett, Broderson, & Macreadie 2020), are expected to increase the rate of aquatic litter decomposition. Then, by removing vegetation, more sediment and nutrient fluxes into the water column could increase the trophic status, decomposition rates, and stabilization factors. However, the manipulation of urban waters will test the tolerance of the microbial community and decrease its diversity (Alter et al. 2021; Momota & Hosokawa 2021). If enough of the community is lost, the decomposition rate may slow, and more carbon will be buried. Albeit, reaching that tipping point may be too far into the future to predict.

This study found that the TBI, adapted with a leaching factor, can be an appropriate method to study plant litter decomposition in brackish water. The rate of decomposition between urban and natural site types is approaching a significance difference, primarily due to the cooler temperatures and lack of microbial diversity at urban sites. Overtime, the decomposition rates at urban and vegetated coastlines may differentiate further. Additionally, the stabilization factor was similar at each site type, indicating an overall high degree of decomposition and release of CO₂. In the future, we expect to see the effects of urbanization and climate change amplified in coastal waters. As a result, decomposition rates may increase

to a tipping point due to rising temperatures, nutrients, hydrodynamic forces, and acidity before declining again because of the lack of a robust and diverse decomposer community.

In future studies, I suggest deploying additional tea bags that will be collected in the first few days to aid in obtaining a more accurate initial decline of mass. Also, selecting more sites in a range of different regions and climates would shed light onto how urbanization globally is impacting micro- and macroscale coastal carbon dynamics and decomposition. The length of the study may be shortened to ensure that the rooibos tea does not stabilize before the completion of the experiment. Lastly, the collection of water samples would help to precisely measure water quality and eutrophication status.

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