

Reviewer 1

We thank Reviewer 1 for their comments and suggestions and feel that these edits have substantially strengthened our manuscript. Below, we include each of the reviewer's comments followed by our response in red font. All line numbers refer to where the edits would be found in a revised manuscript. We hope that these responses have adequately addressed the reviewer's questions and concerns.

Overall, this is an interesting article that investigates the decomposition rate changes of tea bags (TBI) and local plant litter at different soil depths and temperatures, while exploring the factors influencing litter decomposition rates. The writing is generally good, but some sections (such as Results and Discussion) need further refinement. Overall, I suggest major revisions to further enhance the quality of the article. I have several general questions and specific comments as follows:

General Comments:

(1) The title mentions "minerogenic salt marshes." How do these differ from organic marshes? Furthermore, the introduction and discussion sections do not extensively address or explore this distinction. Minerogenic marshes have specific characteristics that could potentially influence the decomposition rates at different soil depths. Could elaborate on this?

The reviewer raises a good point and in response we clarified the characteristics of our study system compared to organic marshes in section 2.1 (study site and design): "Total suspended sediment levels are high in the Altamaha River which feeds into the GCE-LTER domain and contributes to salt marsh vertical accretion (Langston et al. 2021; Mariotti et al. 2024). As a result, these minerogenic marshes have lower soil carbon content, porosity, and permeabilities but higher bulk densities compared to organic-rich marshes (Giblin and Howarth 1984)." (lines 136-140)

We also added the following sentence in section 4.3 of the Discussion to address how differences in soil properties between mineral and organic marshes might affect decay: "Similarly, parallel deployments in mineral and organic marshes with similar flooding regimes but different soil properties (e.g., bulk density, porosity, permeabilities, carbon content, etc.) could provide further insight into how the belowground environment affects decay." (lines 709-712)

(2) In Line 69, the authors discuss plant and animal effects. While the plant effects are covered, how do animals influence decomposition in this context? Specifically, Line 75 emphasizes animal burrows. How might animal burrows impact soil physicochemical properties and soil microorganisms?

To address the reviewer's question we added this sentence in section 1 (Introduction): "Bioturbation by animals can strongly affect decay rates in surface soil horizons, by altering redox conditions, digesting organic matter, and moving organic matter between oxic and anoxic layers (Kostka et al. 2002a; Kristensen et al 2012)." (lines 74-76).

(3) In Line 120, the authors mention using local plant detritus. Why is this important? How does comparing this with TBI enhance the study, and what specific questions or problems does it address? This should be clearly explained in the introduction.

Comparing decay rates estimated from the TBI with local plant detritus is important for determining the applicability of this approach to the target study system. Tea used in the TBI represents allochthonous organic matter with a different biochemical composition – and potentially different decay dynamics – than plant detritus produced locally in the wetland. Comparisons of decay rates based on local detritus between marshes can be complicated, however, due to differences in biochemical composition that reflect factors ranging from nutrient availability to plant species identity. The standardized litter approach of the TBI offers a way to evaluate environmental controls on decay without potential confounding factors due to differences in litter organic matter composition. But before applying decay rates estimated with the TBI approach it is important to verify that it produces rates that are comparable to natural local plant detritus.

In response, we added the following clarifying sentences and hypothesis to the introduction (section 1).

“Built into this is the assumption is that decay dynamics of very specific types of terrestrial organic matter (i.e., rooibos and green teas) reasonably approximate those of local plant detritus.” (line 109-110)

“Consequently, comparing decay rates estimated from the TBI with natural, local litter is important for determining the applicability of this approach to the target study system.” (lines 115-117)

“Lastly, we expected that decay rates of rooibos tea and local plant detritus would be comparable and slower than TBI rates.” (lines 129-130)

(4) Lines 128-141 contain detailed information about study locations. Would it be possible to include a map or illustrative figure to better present the experimental setup?

As suggested, an illustrative figure and aerial plot of the study site was added to the section, showing the 3 transects and their respective distance to the main channel and the tidal creek edge.

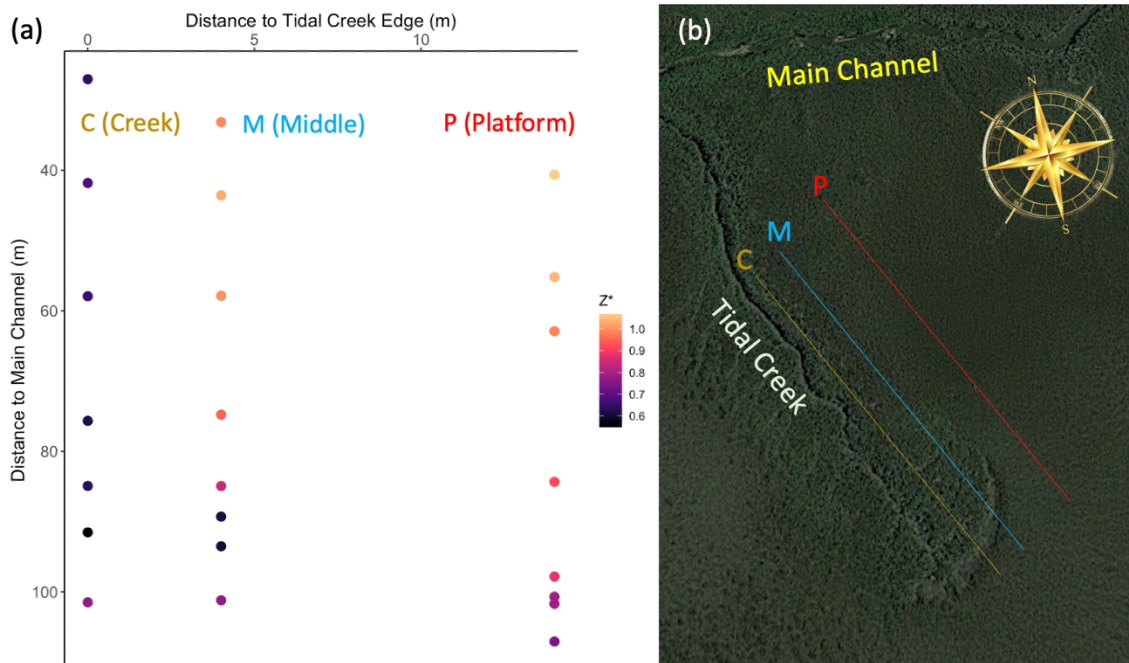


Figure 1. (a) Study plot orientation relative to the main channel, tidal creek, and relative marsh surface elevation (Z^* , see equation 1). Plots were distributed along the tidal creek (C, 0 m), in the marsh platform (P, 14 m), or in between (M, 4 m) across an elevation range of 0.55-1.13 m (NAVD 88). Plot color corresponds to Z^* or relative position in the tidal frame (m). Spacing between plots reflects Wu et al.'s (2022) goal of capturing marsh processes around the fan of a headward eroding creek. (b) Aerial photograph of study site from Google Earth with demarcated lines showing approximate plot distribution in respect to the headward eroding creek. Exact plot locations are described by Wu et al. (2022). Plots affected by the headward eroding creek were excluded from this study.

(5) Lines 241-254 raise several questions. The data used are from 2003-2004; could there be discrepancies with the current situation? Also, why use root litter for local litter experiments? The root litter used weighs 10g, whereas TBI uses approximately 1.6g. Does this affect the comparability of the experiments? Moreover, local litter was placed at -10cm and -20cm depths, but not at -50cm. How many replicates were there for TBI, and are they consistent with local litter experiments?

The reviewer is correct to note that the comparison between the 2003-2004 litter deployment and our TBI experiment is imperfect. However, one goal of our work was to assess potential bias of using a non-local organic matter (i.e., tea) to approximate decay of local marsh grass detritus and the dataset we present is the only one – published or not – to provide this comparison. Prior work on decay in this system used source specific isotopes and biomarkers, providing information on a much finer resolution than the bulk, mass loss approach of the TBI. Comparisons between compound class (or compound specific) rates with total mass loss rates can be difficult to interpret because plant litter is comprised of multiple compound classes with different reactivities. Roots deposit organic matter into anoxic soil horizons and this material is more likely to contribute to burial than aboveground shoots and leaves. Consequently, root litter is the best proxy for bulk soil organic matter decay. In response to the reviewers question we added the following clarifying sentence: Root detritus is a good proxy for evaluating soil organic matter dynamics since this material is deposited directly into anoxic horizons and contributes to soil accumulation (lines 284-286).

Both the litterbag technique and our empirical calculations of tea decay are based on relative mass loss. Because of this, it should not matter if the initial mass was 1 or 10 g. Root litterbags were buried between 10 and 20 cm, which is slightly deeper than the shallow tea bags (10 cm). Because the rhizosphere extends to ~30 cm we believe that the slight difference in burial depth between the root litterbags and the shallow tea bags should not affect decay rates. Recognizing that environmental conditions that could affect decay differ between surface and deeper horizons we only compared the root litterbag rates with the shallow (10 cm) horizon tea bags (e.g., lines 518-520: "... TBI rates at three months are 2.8 – 7.2 times faster than prior studies but that drops to roughly double over longer, 6-12 month periods (10 cm horizon only...)")

To address the concern about differences in environmental conditions between the litterbag experiment and the teabag deployments we compared monthly mean temperatures and precipitation between 2003 and 2019. This is reported at lines 289-291: "Environmental conditions including precipitation and temperature were similar ($p > 0.05$) between the litterbag study in the

summer of 2003-2004 ($7.7 \pm 1.4 \text{ cm yr}^{-1}$ and $20.4 \pm 2.1 \text{ }^\circ\text{C}$) and TBI experiment in the summer of 2019-2020 ($12.1 \pm 1.9 \text{ cm yr}^{-1}$ and $21.4 \pm 2.3 \text{ }^\circ\text{C}$).”

The goal of this aspect of the manuscript was to evaluate whether the TBI produces decay rates comparable to natural litter and we believe that slight differences in deployment depth and environmental conditions cannot account for the 120 % difference (TBI vs. natural litter) and 36 % difference (rooibos tea vs. natural litter) between these two approaches.

There were more replicates in our TBI experiment (23 per depth and time point across 3 transects) than in the litterbag experiment (4 reps per time point in the marsh levee and plain sites).

(6) Lines 255-293, Data Analysis, is overly detailed and needs to be condensed for clarity.

As suggested, we have made the Data Analysis section more succinct. The section now reads as follows at lines 294-329:

“Changes in belowground environmental conditions across marsh surface elevations, between soil depths, and over time were assessed with regression analyses and t-tests. Distance category identity (C, M, P; Fig. 1) was excluded from all statistical analyses because it was confounded with marsh elevation, which we predicted would be a key factor affecting environmental conditions and decay rates. Tidal flooding effects on soil porewater chemistry and temperature were tested by constructing regression models against relative elevation (Z^*). Porewater data were then aggregated by sampling event and two sample t-tests were used to detect differences between 10 cm and 50 cm depths. Correlations between Z^* and soil temperature were further tested by partitioning according to season (summer: 18/7/2019 to 22/9/2019; fall: 23/9/2019 to 22/12/2019; winter: 23/12/2019 to 19/1/2020) and periods of tidal inundation or exposure; differences between slope coefficients were evaluated based on Clogg et al., (1995). We tested whether soil temperatures differed between depths within each season using paired t-tests.

We tested whether decay rates (TBI k , empirical k_g , empirical k_r ; d^{-1}) and stabilization factors (TBI S , empirical S_g , empirical S_r) differed over time (3-, 6-, or 12-months) and between soil depths (10 vs 50 cm) by constructing linear mixed effect models using the nlme package for R (Pinheiro et al., 2016). The mixed models evaluated the fixed effects of time and depth and included plot number as a random factor. We then conducted paired t-tests to further explore how TBI and empirical decay rates, decomposable fractions (TBI and empirical a_g and a_r), and stabilization factors changed over time within a depth horizon.

Potential drivers of TBI k were evaluated by calculating Spearman rank correlation coefficients between rates and environmental conditions for the three time points (3, 6, or 12 months) and two soil depths (10 and 50 cm). Porewater data for the 3, 6, and 12 month periods were combined with data from previous time points (e.g., 2, 3, or 6 months) to better represent cumulative conditions. Temperature was excluded because the shared time series with TBI k violated assumptions of independence. The TBI k values correlated strongly with relative elevation (Z^*), *S. alterniflora* rhizome and aboveground biomass, and soil stiffness. We then evaluated interdependencies between these potential drivers by using subsequent single-factor regressions of relative elevation (Z^*) vs. aboveground biomass and soil stiffness. Correlations between these variables limited further hypothesis testing of decay drivers to plot position within the tidal frame (Z^*). We tested whether TBI k rates and S values changed with relative elevation (Z^*) using linear regression models and then evaluated differences between the resulting slope coefficients over time and with depth, as described by Clogg et al., (1995).

Data were tested for outliers using a 1.5 interquartile range cutoff and \log_{10} transformed as needed to meet assumptions of normality. Analyses were conducted using R software (R Development Core Team, 2023). Data are presented as means \pm standard error (SE) unless noted otherwise.”

(7) The results section presents extensive data and comparisons, thoroughly examining the data. However, this section should focus more on objectively describing data changes and significant differences, avoiding excessive interpretation and discussion. For instance, Lines 342-344, 370-375, 388-390, and 396-397 include speculative comments that are better suited for the discussion section.

Those sentences were removed from the results section, as suggested.

(8) In the results section, the authors used a non-traditional TBI index calculation, separately calculating the decomposition rate and stabilization factor for green tea and rooibos tea. However, the terminology must be consistent throughout (e.g., kg, kr, Sg, Sr). Phrases like $S=S_g$ and TBI decay appear in the text. The same issue exists in the discussion section. Please ensure consistent terminology.

In response, we further clarified terminology used to distinguish the TBI and empirical approaches (copied below) and thoroughly edited the manuscript to ensure consistency throughout.

“From here forward, rates and variables calculated using Keuskamp et al. (2013) or the first order decay approach are referred to TBI and empirical, respectively. For the TBI, this includes TBI k (eq. 3), TBI a_g (fraction mass loss of green tea), TBI a_r (eq. 5), and TBI S (eq. 4) to denote decay rate, the decomposable fractions of green and rooibos teas, and the stabilization factor, respectively. The empirical calculations include k_g and k_r (eq. 6), a_g and a_r (mass fractions lost of each tea type), and S_g (eq. 4) and S_r (eq. 7) where g and r refer to green or rooibos teas, respectively. Importantly, there are two commonalities between these approaches: TBI a_g is the same as empirical a_g , and TBI S is the same as empirical S_g .” (lines 267-274)

(9) Lines 509-525 commendably summarize the discussion on TBI results and the relationship between k and S . However, the discussion would benefit from a clearer connection to the study's findings and implications.

We appreciate the reviewers positive feedback and in response have made the connections to our study clearer. The goal of this section of the manuscript was to contextualize our TBI decay rates relative to natural litter decay in the same study system and TBI rates in wetlands globally. These comparisons are important for interpretation and extrapolation of TBI rates as well as for using the main strength of the TBI approach – uniformity of litter composition – to test large scale ecological theories, such as the Metabolic Theory of Ecology.

Lines 530-537: We expected TBI rates to be faster in our study in subtropical Georgia, USA, compared to higher latitudes based on the metabolic theory of ecology, which predicts that decay rates increase with rising temperatures (Yvon-Durocher et al., 2010), and observations that warming accelerates loss of labile compounds in soils (Conant et al., 2011; Melillo et al., 2002). To test this, we compared our rates with those from 7 other salt marsh TBI studies that encompass 11 countries and span a latitudinal gradient of 93.7° (-37.7° to 56° ; Fig. 5, SI table 2) (Mueller et al.,

2018, Puppini et al., 2023, Marley et al., 2019, Alsafran et al., 2017, Yousefi Lalimi et al., 2018, Sanderman and Eagle, unpub, Tang et al., 2023).

Lines 546-548: “Our Sapelo Island, GA, 3-month rates were similar to salt marshes in North Carolina, Virginia, Maryland, California, and Massachusetts, USA and Zeijhong Province (ZJ), China (Fig. 5).”

Lines 568-573: “In our experiment on Sapelo Island (GA, USA), the highest S values coincided with the fastest decay rates in the first 3 months (Fig. 5). Stabilization values then decreased between 3- and 6- months but there was no overall temporal trend because values increased at 12-months. This variability is not unique to our site; TBI S values decreased and increased at East Lothian (ELN), Scotland and Schleswig-Holstein (SH), Germany, respectively, between 3- and 12- months (Fig. 5).”

Lines 579-581: “This interpretation, though, is caveated our experimental findings and by Mori et al.’s (2022) discussion that green and rooibos teas do not share S values which violates assumptions of the TBI method (Table 3).”

Lines 582-584: “Faster decay rates estimated in this study using the TBI method relative to more conventional litterbag and laboratory experiments suggest that these approaches are not interchangeable.”

Lines 590-592: “Few studies like ours have directly compared decay rates from the TBI, its components, and more conventional approaches but this would be useful in assessing whether Keuskamp et al.’s (2013) method can be applied broadly, in dry and saturated soils”

Specific Comments:

(1) Line 32: Replace "Tea BI rate" with "TBI."

As suggested, we replaced “Tea BI” with “TBI”.

(2) Line 59: The authors mention that effects on soil organic matter decay are "less well understood." What specific scientific questions or reasons contribute to this lack of understanding? Please clarify.

This sentence was deleting during editing of the introduction.

(3) Line 102: When discussing the advantages of TBI, "inexpensive" lacks professionalism. Consider using a more precise term.

In response, we have changed the term “inexpensive” to “cost effective”.

(4) Line 137: Why were tea bags placed at -50 cm, and how was soil disturbance minimized during placement? How many experimental replicates were there?

Tea bags were placed at 50 cm to see how being in a more stable environment with fewer effects from aboveground abiotic and biotic variables (plant roots, crab burrows, tidal flooding) could affect decomposition. We added the following clarifying sentence at lines 222-223: “Burial depths were chosen to assess decay rates within the more dynamic rhizosphere (10 cm) compared to more stable, deeper horizons (50 cm).”

Disturbance at the time of deployment was minimized by inserting a trenching shovel to 50 cm depth and pushing it forward ~5 cm to create a small wedge of space in the soil column. Then a PVC pole with green and rooibos teabags attached with cable ties at 10 and 50 cm was placed into the soil. Because no material was removed, the soil column squeezed back together following shovel removal. Three replicate poles and attached tea bags were placed at each plot and collected at different time periods (3, 6, or 12 months). Recognizing that these actions disrupt soil porewaters and redox conditions we did not collect samples for 2 (porewater) or 3 (tea bags) months.

The experimental design is described at lines 141-144: "Study plots were established in summer 2019 along a tidal creek, with a total of 23 plots placed at 3 distances from the creekbank edge (creek: 0 m, 7 plots; middle: 4 m, 8 plots; platform: 14 m, 8 plots) that captured a range of marsh surface elevations, from 0.55 to 1.13 m (North American Vertical Datum of 1988, NAVD 88; Fig. 1)."

Teabag deployment is described at lines 220-222: "Tea bags were dried at 60 °C to constant mass prior to deployment, during which triplicate bags of each tea type were buried in every plot at 10 and 50 cm depth in July 2019 (initial tea weights: rooibos: 2.01±0.004 g; green: 2.17±0.004 g)."

Consequently for each time point there were 23 replicates at 10 and 50 cm depth, distributed across the 3 distance categories from the creekbank edge (creek, middle, and platform).

(5) Line 208: Specify the initial weight of the tea bags.

We added the initial weights of the tea bags, "Rooibos: 2.01±0.004 g; Green: 2.17±0.004 g" at line 222.

(6) Line 228-229: Why does the methods section introduce potential results and discussion points instead of presenting them in the results section?

We included these sentences as justification for the following description of model selection. Because this information is necessary to understand the data analyses we feel that it is important to include in section 2.6. For these reasons no changes were made.

(7) Line 195: How were the crabs and snails measured or quantified?

In response, we added this information: "The densities of burrows (>0.5 cm diameter, all species pooled) and snails (>0.3 cm spire height) were recorded in 0.5 × 0.5 m quadrats at each plot as individuals m⁻²." (Lines 210-211)

(8) Line 288: Correct the citation format to "Clogg et al., (2009)."

As suggested, the citation formatting was corrected.

(9) Previous comments may have already addressed this, but I remain curious. This study focuses on minerogenic marshes. Are the articles discussed in the discussion section based on minerogenic marshes, or do they include other types of wetlands as well?

Few studies distinguish between minerogenic and organic marshes. We only referenced other salt marsh studies but because of this reporting gap we did not differentiate according to whether they were minerogenic or organic. However, we feel that knowing our study site is minerogenic is

important for interpreting and extrapolating the results. The changes made to the text in response to this comment are the same as those for Reviewer 1's first general comment (above).