Interactive comment on “Spatial gradients in soil-carbon character of a coastal forested floodplain are associated with abiotic features, but not microbial communities” by Aditi Sengupta et al.

Responses to Referee #1 comments are in blue

Anonymous Referee #1 Received and published: 20 June 2019

1. This paper attempts to identify associations between soil carbon chemistry (molecular composition of SOC fractions revealed by FT-ICR MS analysis) and microbial communities (analyzed by 16S rRNA) at the coastal terrestrial-aquatic interfaces (TAIs) influenced by salinity gradients along a small first order stream in the Washington Coast. These two high-resolution techniques generate tons of information on organic matter chemistry and microbial community composition, which allows detailed examination of their linkages. The introduction part nicely lays out the rationale and hypothesis of this study and the paper is overall well written. However, there are a few issues that need to be addressed.

We appreciate that the reviewer recognizes the value in the data we report. We have carefully considered all of the review comments and have provided responses.

2. First of all, the extracted fractions and analyzed molecules are only a small part of the SOC, which may (very likely) not reflect the overall chemistry of total soil organic matter. In this regard, the title and related descriptions should be clarifiedâA˘Tit is “chemical ˘ characteristics of soil carbon fractions” instead of “soil-carbon character”.

We will edit the title in the revised version to indicate this change and clarify in the text that we use “soil carbon character” in our text to indicate chemical characteristics of soil carbon fractions.

3. It should also be mentioned in the Methods how much SOC was extracted by the employed method.

The sequential extraction protocol is able to extract 2-15% of total organic carbon as per previous established protocols (Tfaily et al., 2015, 2017). *We will note this in the methods. The goal here is to get a representative sample of the water soluble and chloroform soluble pool, which the two references cited above prove. This is a well-established protocol and we are confident that the extractions represent both polar- and non-polar soil organic carbon fractions.*

4. Given the lability of WSOC, it is hence more likely to be influenced by microbial decomposition compared to bulk SOC, but it is also strongly influenced by direct inputs of low-molecular compounds from root exudates, etc.âA˘Tthis brings my second point. ˘ Despite the nicely formulated hypotheses for this paper, the authors seem to largely ignore (or underestimate) the influence of input processes on the molecular composition of extractable OC. Water- and solvent-extractable OC may derive from direct plant and algal inputs other than depolymerization of soil macromolecules by microbial-mediated enzyme attack. How would root exudates contribute to the thermodynamically less favorable C, for instance? Do you have an estimate of NPP (hence soil inputs) along the study gradient? The observed changes in C

chemistry may well be a combined result of decomposition and input processes. Similarly, how would photo-oxidation affect the signal?

Agreed that extractable OC is driven by inputs (plant and algal derived) and that the observed changes in C chemistry is a combined result of decomposition/input processes which we cannot separate out. We will add sentences in the introduction to indicate these.

While we agree that root exudates may impact the carbon signatures, this was not the focus of our study. However, we attempted to evaluate common root exudate composition from literature so that we could derive Gibbs Free Energy of the compounds, particularly focusing on compounds associated with vegetation found at our transects, but were unable to find any relevant information. We thank the reviewer for the suggestion and it indeed will be an interesting new study to see how root exudate chemistry varies across the salinity gradient.

Unfortunately, we do not have a good estimate of NPP for the field site at this time. Using MODIS NPP product is also not a viable option because MODIS is 1 km pixel scale while the Beaver Creek site itself is 3.8 km^2. However, we are in the process of collecting data to make such calculations for future studies focused on plant physiology at this same site. In the future we plan to examine changes in soil carbon chemistry as the floodplain soils becomes increasingly saline, and will include NPP information in our future efforts. Thank you for the recommendation.

We do not anticipate photo-oxidation at 10 cm and 19-30 cm soil depths.

5. Regarding the analysis and interpretation of the FT-ICR MS data, I am not convinced that the number of common/unique formulas is the best parameter to describe changes in OC chemistry.

We have limited our interpretation of common/unique formulas only from the perspective of similarity to compound classes at different sampling locations. We do not claim this to be the best parameter. We have looked at other features to describe changes in OC chemistry including Gibbs Free Energy, heteroatom content, and inferred biochemical transformations.

6. The relative abundance of these formulas should be considered.

We have provided relative peak abundances of compound classes in the water extracted organic carbon fraction (Table 1, Line 1048).

7. How representative are the unique formulas in the overall abundance of total MS peaks, for instance? How does the relative abundance of common formulas change with salinity gradient? Hemingway et al. 2017 GCA give a good example for such kind of analysis.

This is an interesting idea suggested by the reviewer but it is beyond the scope of our study. The analysis being suggested here is different from what we asked and evaluated. We did pairwise comparisons by grouping samples according to landscape position and depth (Lines 367-370), with common/unique features comparable between groups like Floodplain versus Inland, Floodplain versus Terrestrial, and Inland versus Terrestrial at two individual depths.

Comparing sample 1 to sample 2, and then sample 1 to sample 3, and so on to evaluate how common formulas change with salinity gradient will lead to results that will be difficult to interpret because the commonality/unique features as a fraction of common/unique peaks is not a property inherent to sample, but only emerges when compared to samples. Therefore, we did not evaluate representativeness of unique formulas in the overall peaks because the unique/common feature is relative and dependent on which groups are being compared. We specifically used the common/unique formulas to understand relative compound class similarity at transects, irrespective of the salinity gradient. Given that our system is in transition with variable tidal/terrestrial inputs, we used the common/unique features to understand the predominant compound class patterns in our field site.

8. Specific comments: Line 219: Why these two depths?

The two soil depths were chosen based on visual soil characteristics. The shallow depth was the organic-rich horizon, while the deeper depth was characterized by lighter colored, clay-rich soils. We did not go any deeper due to logistical constraints—during the time of sampling, the holes back-filled with water up to roughly the depth of the "deep" samples. The depth of distinct layers were fairly consistent across all floodplain sites and not as evident in the upland forest site, however we maintained consistency with how the floodplain soils were collected in this case.

9. Line 395: Relationship with what?

Relationship of compound-like peak abundances with specific conductivity. We will edit the manuscript to reflect this change.