

## Author's response to Reviewer#1 (anonymous)

### *General comments:*

The authors have worked with a variety of samples viz. vegetation, soil, suspended particulate matter, riverbed sediments from the Godavari region which is commendable.

**Response:** We thank the review for their positive assessment of the approach we used in this paper.

But I am curious as to why the authors chose isotopic analyses for the study. The authors must note that there are other much stronger techniques that can be applied for palaeovegetation reconstruction, such as geochemical biomarkers or compound specific isotopic analyses of normal alkanes. These provide much more detailed/spot on information without significantly less biases or overlaps. So the authors must signify and explain very clearly the selling point of this paper and strength of the technique that has been used.

**Response:** We agree with the reviewer that there are different techniques available for vegetation reconstructions, including plant-specific markers and/or compound-specific isotopic analysis, which are often applied in settings of marine sediments where a core is analysed that can give information about the past of the river basin. For a high-resolution study like we did in the Godavari basin with analysis of many modern plants, soils, riverbed and suspended sediments, bulk carbon isotope measurement provide a low-cost and high-throughput approach that can be easily applied in large geographic areas to determine the contribution of C3 and C4 vegetation. This is particularly important in arid tropical and subtropical basins, where carbon and carbon-based biomarker concentrations in soils and sediments are generally low and highly susceptible to degradation due to the high prevailing temperatures. Furthermore, sewage and waste inputs into (sub-)tropical rivers may contaminate a biomarker signal. For instance, compound-specific n-alkane analysis in the Godavari basin was not feasible due to low concentrations in soils and sediments and minor fuel spillages altering the n-alkane signal.

→ We will include this motivation for bulk carbon isotopic analysis in the introduction.

I would also like to add that unless a journal's terms and conditions require so, it is generally not a good idea to combine "results and discussion". Separating the two makes it much clearer as to what your own data and results are depicting and the discussion would include clear explanations of your results. It is important for readers to identify the original data of your research and separate them from previous literature data and knowledge that come under discussion part.

**Response:** We are aware that there are different ways to organise results and/or discussion sections and Biogeosciences does not require a certain format. Given our extensive and diverse dataset (vegetation, soils, SPM, and riverbed sediments for both the wet and the dry season), we used a combined results and discussion section to keep the overview of which part of the dataset is being interpreted. We believe that our format helps to create a good flow in the manuscript. For example, we need the results and discussion in each subsection (3.1:  $\delta^{13}\text{C}$  in C3 and C4 plants and controls by MAP) to move into the next section (3.2:  $\delta^{13}\text{C}$  in soils and sediments and their controlling factors) to build up to the application of a mixing model for C3/C4 contributions (3.3).

→ We will check the manuscript and emphasise our results by including phrases like: our results/ Godavari plants or samples show.

*Specific comments:*

Page 6, Line 150-152: There is no mention in the introduction as to how microbial inputs or early diagenetic alterations and early decomposition of organic matter might affect the isotopic signatures. The present approach is highly one-dimensional primarily considering input of C3 vs. C4 vegetation in connection with wetter and drier conditions. Authors must take into account all the other factors, particularly those which significantly influence isotopic fractionation.

**Response:** We agree that a complex interplay of different inputs and processes affect the organic matter (and its  $^{13}\text{C}$  composition) in soils, including preferential degradation, carbon-containing contaminations, and differential fractionation by bacterial and fungal biomass. While the majority of soil organic matter derives from microbial processed plant residues, the actual size of the microbial biomass is difficult to ascertain and highly dependable on the availability of labile carbon sources and moisture levels. Other researchers estimated that (for conditions with sufficient moisture levels and easily available C) the microbial biomass itself contributes only 1-5% of the total soil organic matter (Kögel-Knabner, 2002; Simpson et al., 2007). Furthermore, there will be considerable spatial heterogeneity in microbial biomass distribution in field studies and techniques to accurately determine the microbial biomass size are challenging (Birge et al., 2015; Wiesmeier et al., 2019). Therefore, we consider determining the contribution of the microbial biomass in the Godavari basin beyond the scope of our study. We note that we sampled the Godavari soils at the end of the dry season, when moisture levels were very low and likely limited the size and activity of the microbial biomass.

→ We will include a note in the introduction on the different processes/inputs affecting soil organic carbon and its isotopic composition in soils, which becomes more positive due to microbial processing. In the discussion (3.2.1) on degradation processes in soils, we will expand on the role of the microbial biomass and include the issues raised above.

Page 8, Line 190: "aboveground plant parts" - Is any part of a plant other than leaves being sampled?

**Response:** Yes, for trees and large shrubs the leaves were sampled, and for grasses and herbs the leaves and stems were combined.

→ We will specify this in the MS.

Page 11: "Modern C3 and C4 plants in the Godavari basin and control by MAP" - Are there no CAM plants in the region?

**Response:** A literature search revealed that CAM plants are not wide-spread in India (Sankhla et al., 1975; Ziegler et al., 1981), and this corresponds to our field observations that succulents are not prevalent. Modern Indian plants on the Gangetic plains (Basu et al., 2015) and our study in the Godavari basin were all identified as species with C3 and C4 photosynthetic pathways.

→ We will include a note on the (limited) occurrence of CAM plants in India in the introduction.

Page 13, Line 306-308: "For C4 plants, the plants collected in the Godavari basin had significantly more negative  $\delta^{13}\text{C}$  values than the global average estimate ( $-14.0 \pm 0.2$  ‰ ( $\pm$ standard error: SE) vs.  $-12.0$  ‰;  $p \leq 0.001$ ), revealing a difference between the local and global average C4 end-members" - Can you explain this observation?

**Response:** We analysed 16 C4 plants in the Godavari basin which fell within a relatively narrow isotopic range, and were all sampled at relatively dry locations with MAP mainly  $< 1000$  mm/y. Basu et al. (2015) also found relatively low  $\delta^{13}\text{C}$  values in C4 plants from the

Gangetic plains at < 1000 mm MAP, which they attributed to a moisture control resulting in lower  $\delta^{13}\text{C}$  values in moisture-stressed C4 plants. However, most other studies found no correlation between C4 plant  $\delta^{13}\text{C}$  and MAP. Potentially, the sample size of 16 C4 plants in the Godavari basin over a limited MAP range was too limited to identify a moisture control. In addition, correction of the Godavari vegetation dataset for the Suess effect, as suggested by both reviewers, may reduce the offset with global C4 plant data.

→ In the revised MS we will expand our discussion about a potential moisture control on C4 carbon isotopic values and discuss potential implications of the sample size of Godavari C4 plants. We will also consider the impact of the Suess correction for the comparison of C4 plant data.

Page 14, Line 317-318: Was the  $\delta^{13}\text{C}$  corrected for Suess effect?

**Response:** The  $\delta^{13}\text{C}$  data have not been corrected for the Suess effect. We thank the reviewer (as well as Prof. Feakins) for notifying this. We will correct  $\delta^{13}\text{C}$  data in our manuscript where applicable. In this specific instance, the effect of MAP on C4 plant  $\delta^{13}\text{C}$  values was evaluated and discussed for contemporary plants in the Godavari basin. The other studies cited here that are used for comparison also investigated modern plants within a certain precipitation range. We, therefore, believe that the comparison we make here is reliable and that the conclusions will hold after Suess correction. Regardless, we will check throughout the MS where contemporary measurements are compared with data reported in older literature, and correct for the Suess effect when applicable.

A preliminary assessment of the effect of the Suess correction on the comparison between our measurements on Godavari plants and data reported in literature, as well as for the plant end-members used in the mixing model shows that the overestimation of %C4 due to moisture availability will become slightly smaller (6% vs 10%). Moreover, there is still a substantial underestimation of the %C4 plants when using global plant end-members compared to those based on Godavari vegetation (15 vs 19%). Hence, the effect of Suess correction on the outcomes is relatively small, and our main conclusions about the mixing model appear to be still valid. We will take care to include Suess-corrected data and our full re-analysis thereof in the revised MS.

Page 15, Line 334-335: "This difference suggests that the latter value, which is reportedly strongly biased towards dry ecosystems" - This is not clear.

**Response:** This line will be rephrased to clarify.

Page 15, Line 336-337: "was significantly less negative in the upper basin ( $-28.0 \pm 0.3$  ‰,  $n=32$ ) than in the lower basin ( $-28.8 \pm 0.2$  ‰,  $n=45$ ;  $p \leq 0.05$ )" - Does this difference in the value qualify as "significantly" less?

**Response:** The difference between the upper and lower basin is indeed small, but significant.

→ We will rephrase these lines accordingly.

Page 15, Line 337: "reflecting the gradient in MAP" - Is the difference enough to conclude a "gradient in MAP"?

**Response:** Yes, MAP ranges from ~400 to 2300 mm/y based on long-term data. The difference in MAP between the upper and lower basin was previously reported in section 2.4.

→ We will relocate the information MAP in the upper and lower basin to this section to clarify the gradient in MAP.

Page 15, Line 342: "Pearson's R = -0.34" - This is not even significant especially for such a small population.

**Response:** The effect of MAP on  $\delta^{13}\text{C}$  values of the individual C3 plants is indeed weak, but significant following an independent samples t-test. The significance is indicated by the two \*\*.

→ We will specify this in the revised MS and include an explanation of the level of significance in the caption of Fig. 4.

Page 15, Line 350-352: "Moreover, the Godavari C3 plants were not evenly distributed over the entire precipitation range. Together, this resulted in a relatively weak linear correlation with MAP for the individually measured C3 plants" - This contradicts the previous sentence on line 341. If the effect of MAP on isotopic values are significant, shouldn't the correlation be high?

**Response:** The Pearson's R provides information about the strength and direction of the correlation between MAP and  $\delta^{13}\text{C}$  values of the individual C3 plants, which is -0.34, due to the large variability we observe for the individual C3 plants at a certain level of MAP. Next, the linear correlation estimates the parameters that can be used to predict  $\delta^{13}\text{C}$  values based on MAP. The  $R^2$  represents the amount of variation in  $\delta^{13}\text{C}$  values that can be explained by MAP (which is ~12 % for the C3 plants). A significant correlation between  $\delta^{13}\text{C}$  values and MAP (as expressed by the significant Pearson's R) does not imply a high  $R^2$  (i.e. the factor that explains most of the variation) in the linear regression.

→ We will clarify in the revised MS that the  $R^2$  describes the amount of variation that can be explained by the independent variable (here MAP).

Page 16, Line 384-386: "This isotopic contrast corresponds with the vegetation distribution in the basin, with mixed C3 and C4 vegetation in the upper basin and more C3 plants in the lower basin" - Is the vegetational input only controlling factor for the isotopic values? What about any signatures of soil bacteria?

**Response:** Our observation of different soil  $\delta^{13}\text{C}$  values the upper and lower basin points toward a control by the overlying C3/C4 vegetation. As discussed in the response to the reviewer's comment on Page 6, Line 150-152, the contribution of the microbial biomass is likely small (i.e., 1-5%). We agree that part of the soil organic carbon will be microbially processed, and potential effects of this degradation process are discussed in section 3.2.1

→ We will expand (see response Page 6, Line 150-152), the discussion on microbial processing effects on soil  $\delta^{13}\text{C}$  values.

Page 18, Line 408-410: "However, C4-derived OC has also been shown to be preferentially incorporated into fine fractions where it is better protected against degradation, whereas C3-derived OC is preferentially added to the coarse fraction thus leaving it less protected" - What governs this affinity for the C4 plants towards finer fractions whereas C3 plants towards coarser fractions?

**Response:** The C4-derived OC has been shown to contain more labile, easy degradable compounds. The fine soil particles have presumably a higher ability to stabilise these labile, C4-derived compounds onto mineral surfaces than C3-derived OC that contains more difficult degradable compounds (partially derived from woody plant parts) that preferentially end up in the coarser soil fraction (Wynn, 2007; Wynn and Bird, 2007). In addition, the upper basin, where C4 contributions are highest, is characterized by soils that contain more clays than those in the lower basin, facilitating the enhanced association of C4 vegetation with mineral surfaces.

→ We will include information on the labile vs difficult degradable compounds contained in respectively C<sub>4</sub>- and C<sub>3</sub>-derived OC, resulting in a different stabilisation process in different soil fractions.

## References

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