

## *Interactive comment on* "Benthic C fixation and cycling in diffuse hydrothermal and background sediments in the Bransfield Strait, Antarctica" by Clare Woulds et al.

## Anonymous Referee #3

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General comments: This manuscript investigates the uptake of 13C via two substrate types 1) freeze dried algal cells and 2) bicarbonate into ~1000 m depth sediments with two sites being sedimented hydrothermal vents with different levels of diffuse venting and an off-vent control from a similar depth. The study finds that respiration was the dominant pathway for processing of algal material and that chemosynthesis was non-zero in all sites for both bacteria and fauna, but higher in the non-vent control. Chemosynthesis was subsequently confirmed through uptake of 13C into PLFAs for all sites, with non-vent uptake largely mirroring the patterns found for % incorporation of 13C. The authors thoroughly acknowledge that due to the limited replication and temperature differences between in situ and lab, these rates represent a confirma-

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tion of non-zero rates and are not sufficient to provide further comparisons between sites for fixation of C. The authors have provided a study, that despite the acknowledged limitations, demonstrates the dominate pathway for heterotrophy for particulate OM and confirm chemosynthesis (both background and HSV) without overreaching in their interpretations. I find the manuscript clearly written, but overly concise in the methodology. The results presented adequately support the authors interpretations, with context presented for the limitations present in the study. Specific comments: LN 83: "inorganic substrates" to bicarbonate (H13CO3-) LN 103: A brief description here of the background macrofauna would be appropriate. LN 112: Product number for your labeled algal material is needed, CIL does not appear to sell a marine algal detritus that I could find by searching their site. Be careful with that description too, as it implies a bit of possible reworking given that you are working at 1000 m depth. If the material is the dual labeled lyophilized algal cells then it is really fresh algal material for application at a relatively deep site; you should discuss this if it is the case. An estimate of what portion of the annual flux this application represents would be appropriate to give more context to the amount of material be applied. LN113: Context for the relative amount of application versus what is already there and available would be useful so the reader can gauge how large the applications are versus in situ backgrounds for C and N. LN 116: Describe the sampling intervals, this will help to indicate how many measurement points your rates are determined off of. LN 206: Provide a range of PLFA or organic ‰ 13C enrichments to support this claim. It will be more convincing to readers when presented in that manner. LN 212: I appreciate the candid nature of this statement, it shows a realistic interpretation of the data given the limited replication built into the study. LN 216: Provide a reference about the chemosynthetic endosymbionts, also an indication as to the nature of the symbionts, methane oxidizers or sulfur oxidizers would be appropriate. LN 235: "important aspect" Is this because it is minor, but potentially widespread? Vague as written. LN 244: So, this study likely represents minimum rates for chemosynthesis. The authors should phrase it that way and provide context of what the addition represented in comparison to normally available substrates. Better

to focus on what your study has actually shown than to speculate that rates would have been higher if in situ temps were maintained. LN 250 & 252: 0.24-1.02 and 1.29 both need mg in front of C m-2 d-1, respectively LN 263: Would it be worth trying to isolate polar lipids from archaeal components given their slow metabolism and the relatively short time frame of this study? LN 268-277: Thank you for addressing the variability observed during tracer studies relying on bacterial mediation of a substrate! Is it worth talking about reasons for potential hotspots for both heterotrophy and chemosynthetic processes that are occurring in this system? I would expect variations in vent flows and sporadic availability of resources to give rise to a community that readily adapts to changing conditions. LN 294: Provide percentages from the other studies here so the reader can directly compare these studies. LN 316: Does the time period involved in this incubation matter here? Transfer into symbiont and then into tube worm may take a bit more time and require a stronger signal to show up as the tracer is sequentially diluted through the two carbon pools? Figures: Figure 1: state that depth is in meters in figure caption. Figure 2 & 3: remove blue outline on bars. Considering the low uptake rates, consider converting into  $\mu$ g to limit the decimal places. But, you are consistent throughout currently. Figure 4: Format the letters for the figures into the actual graphs, hard to interpret as laid out presently. Also resulted in the splitting of the figure between page 24 and 25. Both substrates should be on the same y axis scale to aid in interpretation and comparison (both 60% max).

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