Point-by-point response to comments

Title: Fatty acid carbon isotopes: a new indicator of marine Antarctic paleoproductivity?
Journal: Organic Geochemistry

Reviewer 1:
I was not one of the prior reviewers but it appears that the authors have thoroughly revised in response to the prior reviewers' comments.

While reservations remain about the generalist biomarkers and their inferences, the work is carefully reported and merits publication, as it usefully adds data to the conditions and questions posed.

A few minor issues are noted:
Line 127-53 – please attend to superscripts (13) and subscripts (FA and FAME) and italicization (n) in the track changes text this has been incompletely formatted, and elsewhere in the ms (line 359-361)

Response 1: Amended as suggested.

Line 160 – fully extract. The method was sonication, the HBIs are unlikely to be fully extracted. Please remove word 'fully'. The IS doesn't replicate analyte, but reporting its extraction efficiency would be appropriate. Query: As noted on lines 188 the standard was added after extraction for FAs, was it added before extraction for HBIs, i.e. were these separate extractions as stated or is this incorrectly stated? Please check and correct if needed.

Response 2: Removed the word 'fully' as suggested. FAs and HBIs were extracted separately for different PhD projects. We have added a line at 151 to clarify this.

Free-fatty acids only. As noted by a prior reviewer, bound FAs as well as free fatty acids would be pertinent to production and this is not satisfactorily resolved by simply stating the paper studies free FAs. I suggest adding a statement that the release of C16FAs is likely to be incomplete using the sonication approach, and there are references to cite on the likely extraction efficiency if this was not assessed here. Extraction of bound FAs by saponification would likely release more C16FA. Please include a brief indication of the limitations of the methodology, based on what can be found in the literature, such as might inform future studies.

Response 3: We have modified from 'free' to 'freely extractable (using a standard solvent extraction protocol),' We feel our language in this regard is now appropriate. We are extracting the Fatty Acids that are freely extractable using standard solvent extraction protocols for paleoclimatology. We are fully aware that additional fatty acids can be released by more aggressive chemical treatments (or even further by using pyrolysis). We are also aware that more FAs could also be released from macromolecular material (kerogen, etc) that are not present in the cores. For these reasons, we believe this is not necessary to expand in this paper as it is not an organic geochemical method development paper and the audience understands this type of standard approach for Holocene paleoclimate.

Conclusion 'producers…would require further elucidation' – as the prior reviewer comments

indicate this claim is overworked, it's unlikely to be resolvable, or more-importantly generalizable, for such a non-specific compound. Rephrase to not suggest the information instead that can be gleaned from general biomarkers in combination with evidence for producers.

Response 4: We have amended the statement so that it now reads*:*

"Although we have made parsimonious interpretations, there are clearly uncertainties in interpreting the FA δ13C, and, as such various assumptions have been made. The primary producers of the C_{18} and especially the C_{24} FAs are a key source of uncertainty. Because these are general biomarkers, produced by many organisms, it is impossible to constrain entirely to one producer class. But with further work in the region, it could be possible further elucidate the most likely contributors. The possibility of inputs of FAs from multiple sources, in particular from organisms further up the food chain, has consequences for their interpretation since this could mean the δ13C FA is not fully reflecting just surface water conditions".

Such a statement fully acknowledges the uncertainties involved in our study and suggests that further studies like ours will help to better decipher the signal carried by the FAs.

Suggestions for revision or reasons for rejection (will be published if the paper is accepted for final publication)

The paper by Ashely et al presents a fairly high resolution record of fatty acid concentrations and delta 13C values of the most abundant ones (C18FA and C24FA), from offshore Antarctica, in an attempt to find out if this can be used to reconstruct changes in productivity. This productivity is important as a carbon sink, via the biological pump. Overall, the data are fine and should be published. The discussion also brings up valid arguments but appears a little limited, as are the presented data.
I have given more detailed comments in an annotated pdf, but here some main concerns:
1. It would be useful to also show the TOC profile, as well as bulk 13C, to get a sense of the carbon burial rate, and to place the FA results in a broader sedimentological framework. Expression of the FA concentration on a TOC basis would be useful.

Response 5: Unfortunately, the TOC was not analysed and the data is not available.

2. Have the authors considered measuring sterol concentrations, and sterol 13C contents, instead of FAs? Because sterols are clearly of phytoplankton original while FAs can also have a bacterial source. Please comment (we chose for FA, and not sterols, because...

Response 6: We thank the reviewer for their suggestion and agree this would be a good idea for a line of enquiry in theory. The first author studied this core and a Holocene IODP core from the Adelie drift for her PhD. She explored sterol concentrations and distributions. Sterols were abundant but highly coeluting. Because of this we found that suitable separation and isolation of sterol compounds for compound specific isotope analyses was not possible within the time-frame of the PhD.

3. A potential bacterial source of especially C18FA should be discussed (this would be an argument against FAs..)

Response 7: We thank the reviewer for this suggestion. We have added several additional references to Table S2 in the Supplementary which summarizes key references on potential sources of fatty acids, including both bacteria and additional references on marine phytoplankton sources:

Allen, E. E. & Bartlett, D. H. (2002) Structure and regulation of the omega-3 polyunsaturated fatty acid synthase genes from the deep-sea bacterium Photobacterium profundum strain SS9The GenBank accession numbers for the sequences reported in this paper are AF409100 and AF467805. Microbiology 148, pp. 1903-1913.

Allen, E. E., Facciotti, D. & Bartlett, D. H. (1999), Monounsaturated but Not Polyunsaturated Fatty Acids Are Required for Growth of the Deep-Sea Bacterium Photobacterium profundum SS9 at High Pressure and Low Temperature. Applied and Environmental Microbiology 65, 1710-1720, doi:10.1128/aem.65.4.1710-1720.1999.

Jónasdóttir, S. H. (2019) Fatty Acid Profiles and Production in Marine Phytoplankton. Marine Drugs 17, 151.

We have noted potential contributions of bacterial FAs to the main manuscript at lines 224-225, 265-266.

4. A discussion about the apparently low concentration of unsaturated FAs is in order.

Response 8: We do discuss this over two paragraphs, from line 242 to 265. Specifically:

"Many studies have shown that significant degradation of FAs occurs both within the water column and surface sediments as a result of microbial activity, and that there is preferential break down of both short-chained and unsaturated FA, compared to longer-chained and saturated FA (Haddad et al., 1992; Matsuda, 1978; Colombo et al., 1997)…
…. The complete lack of both unsaturated and short chained (fewer than 16 carbon atoms) FA compounds identified within DTGC2011 samples, even within the top layers, suggests that selective breakdown of compounds has already occurred within the water column and on the sea floor (before burial). Wakeham et al. (1984) assessed the loss of FAs with distance during their transport through the water column at a site in the equatorial Atlantic Ocean and estimated that only 0.4 to 2% of total FAs produced in the euphotic zone reached a depth of 389 m, and even less reaching more than 1,000 m depth, the vast majority of material being recycled in the upper water column. Their results also show a significant preference for degradation of both unsaturated and short chained compounds over saturated and longer chain length compounds. Although no studies into the fate of lipids within the water column exist for the Adélie region, the >1,000 m water depth at the core site would provide significant opportunity for these compounds to be broken down during transportation through the water column. It is likely, therefore, that the distribution of compounds preserved within the sediments will not be a direct reflection of production in the surface waters, and explains the preference for saturated FAs with carbon chain lengths of 16 and more."

So in the discussion above we do clearly discuss this issue. We don't really know how much more of a discussion is in order for compounds that we could not measure.

5. I find the apparent correlation between the C24 FA data and diatom species abundance not convincing; this needs to be made more clear.

Response 9: We discuss correlations between the SCFAs (R^2 values between 0.97 and 0.99), and LCFAs (0.88 and 0.95) and summarize this in Fig. 3. These correlations are significant and the distinct groupings suggest that compounds within each group (SCFAs and LCFAs) likely have a common precursor organism or group of organisms.

However, with the relationship of the C_{24} FA delta^{13}C data (presuming the reviewer is referring to the isotope data, they don't specify) we are careful not to talk of correlation. We feel that we do couch this part of our discussion in suitably cautious terms. For example we state at line 546: "*Comparison between $\delta^{13}C_{24FA}$ and the major diatom species abundances within the core (Fragilariopsis kerguelensis, Fragilariopsis curta, Fragilariopsis rhombica, Fragilariopsis cylindrus, Chaetoceros resting spores) shows a reasonably close coherence with Fragilariopsis kerguelensis, particularly since ~1800 C.E. (Fig. 6).*"

We are careful with our language and "Reasonably close coherence" is not the same as "correlation". We feel the readers can also judge visually through data comparison in Fig. 6. They are clearly some excursions which show coherence.

6. Please include also the other FA 13C data, if not for the discussion in this particular paper then for future reference for others who might measure these on other locations. The C22FA appears to be about equally abundant as the C24FA (looking at the GC trace in the supplement); the same is valid for the C16FA (which is of course a very generic lipid).

Response 10: Insufficient data of acceptable quality were measured on the C_{16} FA and C_{22} FA to be included in the study and manuscript.