

## ***Interactive comment on “Modern calibration of *Poa flabellata* (Tussac grass) as a new paleoclimate proxy in the South Atlantic” by Dulcinea V. Groff et al.***

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Dear Samuel Bodé,

We are grateful for your helpful comments, which have improved this manuscript. We have responded to your comments below. We also provide an explanation of the changes we intend to make in the manuscript.

Best wishes, Dulcinea Groff

\*I believe this is an interesting piece of work, as indeed more reliable paleoclimatic proxies for the southern Atlantic are needed, to increase our understanding of past cli-

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mate patterns. The author also collected a nice dataset. I do agree that *Poa flabellata* peat has promising potential to be the base of a good proxy, as it has a high accumulation rate in the peat, and is mainly present as the unique plant species. I do however not agree that the real proof of the power of the recorded isotopic signal as paleoclimate proxy has really been given in this manuscript. I have a couple of major concerns on the data treatment and interpretation and a large number of minor remarks.

First, the observed correlation of  $^{13}\text{C}$  and  $^{18}\text{O}$  of the leaf cellulose with RH and T is used as an indication of the power of the proxy for paleo climatic studies. The leaf samples were young leaves growing during the sampled year. The leaves start to grow vertically in the summer and get broader during the winter. The summer samples are thus systematically younger samples than the winter samples. It can not be excluded that the observed difference in  $^{18}\text{O}$  and  $^{13}\text{C}$  is related to the change in leaf phenology rather than a climatic response. As the entire leaves are collected in winter, the recorded isotopic signal is a combination of the entire growing season.

We believe the reviewer may have overlooked where we described our collection strategy. We collected the youngest leaves of a tussock plant each month to capture the most recent growth. We did not state in the manuscript that the leaves grow vertically in summer and broader during winter. Because we collected young inner leaf material we do not think the observed pattern is related to change in leaf phenology or systematically younger-older samples.

The time resolution of peat core reconstructions would average several years of accumulated plant matter in a single sample. When measuring  $^{13}\text{C}$  and  $^{18}\text{O}$  isotopes of cellulose from peat, any seasonal variation would be time averaged. Alternatively, the measured isotopes could suggest that past environmental conditions similar to or were from the seasonal relationship we observed in this study.

\*Further, it is important to note that this seasonal resolution will not be recorded in the peat record, as only mature leaves will contribute to the litter. A much better way of

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assessing the potential of the proxy for paleoclimatic reconstructions would of course be to sample a peat core, and correlate  $^{13}\text{C}$  and  $^{18}\text{O}$  signals of the core to recorded climate data. Therefore, it is needed to better frame the study, and rather use it as a background study on the physiological response of the Tussac grass and incorporation of atmospheric isotopic signal in the cellulose and only put it forward as a very first step toward the development of a paleo climatic proxy.

We would not assume that only mature leaves contribute to the litter. We think leaves are continuously growing and dying throughout the year.

Because of time averaging ( $\sim 20\text{-}30$  years per cm) and limits to historic weather data [dating back to 1874; Lister and Jones 2015], it is not possible to get enough samples to establish a linkage between weather and isotopic values – we would have only 4-5 peat samples. We agree that paleo data would support this work with a proof of concept, but we wanted to focus on establishing the validity of the proxy through modern calibration, and we feel that these actualistic studies are important.

\*Further comments: It is not always clear what is tested when statistical tests are performed. An example of this, is when the RH and T of the different locations are compared. It seems to me that the yearly average T and RH are compared using the individual days as replicates (i.e. SD computed on variation between days). This seems wrong to me as the variation in T and RH between individual days has no link with the uncertainty on the average T and RH of that location. To compare the RH and T of the measuring period for these locations only the measurement uncertainty (which is typically very small). Further to be able to say something on the difference in yearly average RH and T between sites in general, several years of observations are needed.

We are not trying to establish differences in weather conditions between sites, but rather we used multiple sites to test whether there was significant geographic variation in how tussac records T and RH. Our purpose was to show that these analyses can be done on cores from across the study region by establishing a link between T/RH and

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leaf isotopes. Solution: We will clarify that the space for time substitution used here demonstrates that the processes influencing isotope signals in leaf cellulose observed over a season scales to inter annual change. We demonstrate that the geographic and seasonal patterns give us good confidence in the scalability of the relationship between leaf cellulose signals and humidity and temperature.

\*The same problem occur when  $^{18}\text{O}$  and  $^2\text{H}$  in precipitation between seasons is compared, the monthly variability is not related to the uncertainty of the mean. On top of this, it is not so meaningful to compare the numerical average when comparing seasons, i.e. the weighted average should be used. Again only the measurement uncertainty is relevant when comparing the seasons. Using the weighed averages the average of the locations could be used to compare the different seasons, however, to me it doesn't seems right to use these different location as replicates.

It is unclear to us what the reviewer is requesting, so we are not sure how to respond. There may be some confusion about replicates; our sample sizes are similar between groups. Our use of multiple locations was to determine whether there was any spatial variation in stable isotope values, which would mean any paleoclimate work would require local sampling. That we found no such differences among site responses should be seen as a validation of this approach to the fossil record.

Further, it's important to note that our goal here is not to reconstruct seasonal changes, as peat records will include time-averaging of  $\sim 20\text{-}30$  years/cm.

\*as a final note on this seasons start and end the 21th of a month, while samples were taken per month, this should also be acknowledged.) When looking at the correlation of the  $^{13}\text{C}$  and  $^{18}\text{O}$  in leaf cellulose with RH and T, it is observed that they correlate with both. Beside the issue discussed above it is also important to note that RH and T are also strongly correlated (i.e. Drier (77 -85 %)/hotter (9-11C) summers and wetter (90-98%)/colder (1-5.5C) winters). For which, from the data it can not be concluded if the effect is a result of the RH or T or both. It would be interesting to give some insight

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on what effect might be prevailing here.

Thanks for taking note of this. We intend to acknowledge this. Solution: We intend to include a comment about the discretion between season (start/end on 21 of a month) and when samples were collected. For the second part of the comment, we would like to point out that we addressed this point on lines 270-274.

Smaller remarks: L75: it sounds quite contradictory to expect low WUE in water-limited environments.

Thank you for noticing this, it is an easy fix. Solution: Would be better if L75 read as, “environments where conservative water use strategies are common functional traits that allow. . .”

L80: It is a rather strange thing to say that  $\delta^{18}\text{O}$  of source water often correlate with temperature. Better to say (and this is also how it is described in the given reference) that the  $\delta^{18}\text{O}$  of precipitation correlates with temperature. (sure in the case of tussocks, source water is directly related to precipitation, but this cannot be claimed in general).

Agreed, better to revise this statement. Thank you. Solution: “Temperature of the environment is often correlated with  $\delta^{18}\text{O}$  of source water (Libby et al., 1976).

L128: It is not clear what ‘frozen for eight days’ exactly means, what happened after these 8 days? In fact the entire section is not very clear, what is the point of freezing them for 8 days if some where already stored at RT for 6 months? Please rewrite and clarify.

Agreed, we will clarify. Solution: “Samples collected between September 2015 to February 2016 were frozen in February 2016 and samples collected in March 2016 to August/September 2016 were frozen in August/September 2016. Samples were frozen for eight days at the Falkland Islands Department of Agriculture to comply with U.S. Department of Agriculture permitting to prevent the spread of pests.”

L144 and L150: When secondary reference material are used, the accepted value

used for it should be given (this do sometimes change over time).

Yes, we will add this. Solution: For L144: "...using USGS-42 (8.6 ‰ accepted value) and IAEA-601 cellulose (31.9‰ accepted value)...". For L150: "...included USGS-40 glutamic acid (-28.3‰ accepted value), USGS-41 glutamic acid (24.4 ‰ accepted value), and internal UWSIF  $\alpha$ -cellulose(-24.9 ‰ accepted value)."

L203: Paragraph is too long, to many irrelevant details are given for which the major lines get lost. I believe this paragraph should be shortened by c.a. 50%

Thank you for the suggestion. Solution: We will edit this paragraph.

L204: Why is this correlation analysis done on averaged values? It should be done using the individual data points.

Most correlation statistics assume independence of observations. Using individual data points instead of averaged values would violate this assumption when the goal is to compare seasonal variation across sites each month.

L13: replace 'investigate' by 'measured', delete 'plants'

Helpful suggestion, thank you. Solution: Rewrite as "Here, we measured the isotopic composition of *Poa flabellata* and..."

L14: I believe the author mean: '. . . . explore relationships with seasonal temperature and air humidity variations across 4. . ..'

Yes, thank you. Solution: We will change as described.

L16: Delete 'significantly' (if not you would not report it)

Good suggestion. Thank you.

L23: 'did not differ significantly' (there is no test to claim that 2 things do not differ, you can only claim that you could not see a significant difference.

Okay, good suggestion. We will change this. Solution: "No observed no significant

difference in the d18O values of monthly composite precipitation between seasons or study locations.

L32: ‘. . .resulted in an intensification and poleward. . .’

Sure, we will make this change. Solution: Change “correspond” to ‘resulted’.

L35: Sentence starting with ‘The inconsistency of. . .’ Is not totally clear, reformulate.

Thanks. We will fix by splitting into two sentences. Solution: “Meteorological measurements from the Falkland Islands date back to 1874 and are not continuous (Lister and Jones, 2015). This means we lack critical information on the long-term patterns and whether these are novel conditions.”

L47: Should it not be ‘. . .generate substantial amounts of peat. . .’ ?

We will change this.

L57: I believe it could be cited as. ‘Smith and Prince (1985) established radiocarbon. . .’

Yes, thank you, we will make this change.

L62: ‘. . .of any peatland, globally, P. . . . .’

Thanks. We will make this change.

L67: ‘. . .in this semi-arid habitat. . .’

The references used here do not describe this semi-arid habitat in the Falkland Islands, these studies are about other places in the world that are semi-arid but not the Falklands.

L69: a) I guess it is ‘up to 39 cm’ or ‘c.a. 39 cm’ b) ‘. . .year) while in winter an increase. . .’

We will make the recommended change. Solution: Change to “(~ 39 cm per year)

while in winter...”

L70: ‘. . .tiller at the base of. . .’

Thank you for making this correction. Solution: Change to “...production of new tillers at the base of a...”

L70: Sentence starting with ‘The climate signal. . .’ Is not clear, please reformulate.

Thank you, this sentence will be rewritten for clarity. Solution: Stable isotopes  $\delta^{18}\text{O}$ ,  $\delta\text{D}$  and  $\delta^{13}\text{C}$  in the cellulose of plant tissues (roots, shoots, and leaves) can reliably record the climate signal related to environmental growing conditions (Araguás-Araguás et al., 2000). Carbon isotopes. . .”

L73: ‘physiological responses such as changes in stomatal conductance and. . .’

Nice suggestion. Will change accordingly.

L77: ‘. . .information on. . .’

Okay, sounds okay either way.

L78: ‘. . .tissue was formed. . .’

Okay.

L79: ‘. . .humidity and plant physiology (. . .’

Great suggestion. Solution: Change to “...temperature, humidity, and plant physiology (...”

L80: ‘. . .often correlate. . .’

Good suggestion. Will change. Solution: “...source water often correlates with temperature...”

L81: ‘. . .cellulose can also. . .’

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Good suggestion. Solution: We will make this change. “...cellulose can also be influenced. . .”

L82: ‘. . .plant water pools.’

Thank you. Solution: We will make the suggested change “...and plant water pools.”

L86: rather use ‘%’

Good suggestion. Solution: This will be changed to be the first use of % and first defined (removed first use later in the text L115-116).

L97: If mean temperature is given, the time span of this mean should be given, same for

Good point. We will add this. Solution: Our source references worldclim.org and we will add that the time span ranges from 1922-1988.

L109: Why is the fact that they were first shipped to University of Maine mentioned? I don’t think the reader is interested in the postal route. . .

Okay. Solution: We will remove this, however it seems relevant to know how the samples were treated.

L113: It is ‘. . .were purified. . .’ or ‘Water was extracted out of precipitation sample. . .’

Good suggestion. Solution: We will change to ‘purified’.

L119: ‘. . .relative to VSMOW.’ No need to mention they are reported in % (this is visible in the results, and can lead to inconsistency).

Good suggestion. Solution: Will make this change (it was also removed in a previous comment).

L130: ‘. . .were used, fine roots. . .’

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Okay. We will remove the word “and” as suggested.

L133: Just out of personal curiosity, did the grinding of leaf material using a ball mill work? my experience is that this do not work very well with fibrous material.

The grinding of leaf material with the Retch ball mill worked very well. We used two balls in each canister to homogenize the fibrous material.

L137: a) What is an undetectable amount? Should give a detection limit here, if not it is meaningless. b) it is nitrogen and carbon content, not %nitrogen an %carbon

Agreed. Solution: This is how it could be re-written: “Further indicators of purity include low amounts of nitrogen content (0.13 % to 0.16 % N), and analysis of carbon content (42.1 % to 42.8 % C) in cellulose of four samples.”

L140: ‘varied by < 0.1 ‰ and 0.3 ‰ respectively’

Good suggestion. Solution: We will remove the plus/minus signs.

L143: ‘. . .relative to VSMOW (. . .’

Good suggestion. Thank you for helping to tighten this language. Solution: We could rewrite as mentioned.

L146 and 151: replace ranges by ‘c.a. 0.25 mg’ and ‘c.a. 2 mg’

Sure. We intend to add the targeted weight instead of the range of our weights. Solution: We will change this to reflect the target weights “c.a. 0.25” and “c.a. 2 mg”.

L148: Delete ‘; units are expressed. . . . .mil’)

Thanks for the suggestion. Solution: We will delete.

L150: Reformulate last sentence

L153: what is meant with ‘analysing the average’? I think the author means that for every site 3 to 4 leafs were used as replicates in every month.

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Our experimental unit was the average of 3-4 plant leaf samples (from new plants each time) at each site each month. We did not expect differences across sites. Solution: We will edit as suggested "...we averaged three to four plant leaf samples"

L184: what is the 'n = 344', summer only counts 90 days. . .

Thank you for pointing this out. We agree this could use clarification. Solution: Rewrite like this: "...using n = 344 individual trajectories. . .(n = 332 individual trajectories) were from the. . ."

L189: Simply say that September 2015 sample was missing for surf bay (we all know that a year has 12 months). Was that sample missing or was it not sampled, meaning that October 2015 is in fact September + October?

Thank you, this definitely needs clarification. Solution: Rewrite: "...which was not sampled in September 2015, and October 2015 represents September and October 2015."

L192: '. . . D values (. . .', 'monthly composite' is redundant with the first part of the sentence.

Great catch. Thank you, this will improve writing clarity. Solution: "D isotopes (y = . . .)."

L204: '. . .leaf 18O and 13C values. . .'

Thank you. We will make this change to the symbols. Solution: "...monthly average leaf  $\delta^{13}\text{C}$  and  $\delta^{18}\text{O}$  values for. . ."

L206: add p value after segregation between winter and summer values.

Thanks. We will move our reported p-value to the end of the sentence. Solution: "...positive correlation (Pearson's  $r = 0.877$ ,  $n = 24$ ; Fig. 3B) and segregation between winter and summer values ( $p < 0.001$ )."

L206: How could measurement have a significant correlation? I believe the author

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meant to write that the d18O value of precipitation did not correlate significantly. Further it would be more logical to write that d18O of leaf and root did not correlate with d18O of precipitation rather than the other way around (statistically it is the same thing, though it is not logical).

Sure. We will change to make it more logical, if that helps. Solution: “The  $\delta^{18}\text{O}_{\text{leaf}}$  or  $\delta^{18}\text{O}_{\text{root}}$  did not correlate with  $\delta^{18}\text{O}$  in precipitation across all. . .”

L212: ‘ $28.9 \pm 1.3 \text{ ‰}$

’idem at L214 etc. . .(change everywhere)

L213: I do not find it meaning full to add the range, as the distribution was normal, giving average  $\pm$  SD is enough (and really no reason to add also the range).

We think both the standard deviation and the range are useful. Reporting the range is valuable for understanding patterns in the data, e.g., boundaries.

L222: interaction is not an effect, what the author wanted to say is that ‘no significant interaction could be observed’.

Thanks for making this language clearer. We will make this change.

L237: Not clear if this is an own observation (nothing is mentioned about VPD in the result section) or something from literature.

Thanks for pointing out the need to clarify. Solution: We will clarify by writing “The observed negative correlation between d13C and relative humidity. . .conditions.” instead of “The negative correlation. . .(Ferrio and Voltas 2004).”

L241: ‘. . .ratio of CO<sub>2</sub> partial pressure in the leaf and that of the ambient air. . .’

Great, thanks. We like this suggestion and will rewrite. Solution: “...ratio of CO<sub>2</sub> partial pressure in the leaf and that of the ambient air. . .”

L241: what can be explained, the difference in parial pressure ratio or the effect of it

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on the 13C?

Thanks for noting this. We will clarify. Solution: We will rewrite like this “Variation in d13C is driven by changes...2000).”

L257: what are ‘cellulose isotopes’?

Thanks. Solution: We will remove the word “isotopes” and the sentence reads well.

L317: Quite strange to say ‘at least 12,500 14C years, while on line 64 it says that peatlands initiated between 12,500 and 5,500 14C years. . .

Thank you for pointing this out. This sentence could use clarification. Solution: We could be consistent and say “some peatlands initiated by 12,500 14C years. . .” instead of giving a range.

Figure 3b: Why is not the individual data presented, rather than averages?

We designed the experiment so that each replicate is one month at each site. In other words, each experimental unit is an average of 3-4 isotope measurements from one site (four total sites).

Table S1: if the it is given that longitude is south, the negative sign should not be used (sensu stricto a negative south latitude is a northern latitude). So remove the ‘(S)’ or the ‘-’ idem for long (W).

Great suggestion. Thank you for your attention to detail. Solution: We will remove the “-” from provided latitude/longitude.

Figure S3: Link did not work, until I found that the ‘-’ was not for a split for a line break (like my browser interpreted it when clicking on it), but a real hyphen. Probably better to use ‘<https://climatereanalyzer.org/>’

Okay, thank you. Solution: We will update with the new website name.

Figure S5: Add your data to this graph.

Good suggestion. Solution: We will add our data to this graph. However, this would mean the data would be presented twice: once in the main manuscript and once in the supplemental information.

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