Authors comments on Swann and Patwardhan

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We thank the reviewers for their comments and acknowledgement that this method is an important step in improving the quality of biogenic silica isotope/geochemical measurements. Below we have summarised our response to the reviewers questions and will incorporate these comments into the revised version of the manuscript.

Reviewer 1

We thank this reviewer for encouraging comments.

1) *...Unlike foraminiferal analysis, where tests can be picked individually, the authors correctly say that this is rarely possible for small diatom shells and phytoliths. I would, however, argue that individual picking is possible for sponge spicules (Hendry et al., 2010a)...*
2) *Previous methods for assessment of contamination are mentioned by the authors (lines 25-30, p 1631) including spot counts under light microscope/SEM and XRF. However, I would also add into this list dissolution experiments that selectively dissolve smaller particles (presumably contaminants) and surface coatings preferentially....*
3) *A caveat worth mentioning when discussing the use of XRF, or other methods of assessing contamination through Al analysis, is the wide range of Al content observed in diatom opal, especially at the surface-water interface...*

We agree with the comments 1-3 from the reviewer 1 and would be happy to accommodate them into a revised version of the manuscript.

4) *Have other methods of purity assessment been carried out on BFCmod and PS1772-8 (e.g. SEM spot counts, dissolution experiments)? It might be useful to have some further, independent measures of contamination if possible.*

In addition to those mentioned in the paper, the purity of both BFC_{mod} and PS1772-8 have also been assessed and confirmed as being contaminant free by semi/non-quantitative methods such as SEM, light microscopy, XRD and NMR analysis.

5) *When the authors prepare the "silt", is there any risk of leaching of clays by the NaOH? How concentrated is the NaOH? Perhaps the authors could try leaching in different molarities, and comparing the resulting curves to assess the possibility of lithogenic leaching.*

The "silt" end-members were prepared using a 0.5 M NaOH solution to remove all diatoms/biogenic silica (Morley et al., 2005). As the reviewer points out, there is a risk that even a mild alkaline solution could have leached the "silt". However, existing work has shown that the ratio of Si/Al leached from clays is constant over time and solution strength (Kamatani and Oku, 2000). Accordingly it is reasonable to assume that any leaching would not have preferentially removed one element over the other and so altered its geochemical composition/FTIR spectra. Although this situation is not ideal, in this instance it presents the only possibility for obtaining a pure end-member sample that is as representative as possible of the contaminants in the diatom samples.

6) *Do the authors have an idea of how the spectrum would compare for phytoliths and sponge spicules (given the range in omega values for diatoms and sponges)? For example, if diatom samples are contaminated with non-diatom opal, which is a particular problem for Si isotope analysis, would this be detected in the spectrum? This is touched upon in the conclusion, but it would be interesting to see some examples, if possible.*

Other authors have shown that both phytoliths and sponge spicules have similar spectra to diatoms (Gendron-Badou et al., 2002; Loucaides et al., 2010). Whilst the FTIR method is therefore suitable for distinguishing between biogenic silica and other contaminants, the technique is not sufficient for detecting

between different types of biogenic silica which will instead continue to require checking via microscopy.

7) *Is there evidence that a KBR pressed pellet results in material loss? (Line 13-14, p 1634).*

What is meant in the manuscript is that once a pressed KBr pellet is prepared for FTIR analysis, it is very difficult to retrieve the samples after measurements. In fact, it is not a common practice to recover samples from KBr pellet in order to avoid any contamination. On the other hand, the method employed in this manuscript – ATR-FTIR – allows not only the use of minimum amounts of sample but also sample recovery.

8) *Are there other ways the authors could identify the other contaminants? (p 1638). It seems reasonable to assume organic matter could be important – could the authors analyse the organic carbon content?*

The issue of organic matter in the sample is an important one, particularly as light microscopy shows that many of the high residuals in Figure 5 may be due to organic matter. Due to the large amounts of material needed for XRF analysis (>100 mg), insufficient material remains to carry out a proper organic carbon analysis. LOI values obtained during the preparation of a fused bead for XRF analysis would appear to confirm that higher levels of organic matter are present in these samples. However, distinguishing between organic matter, occluded water and other processes that may influence LOI such as diatom silica exchanges prevent an accurate assessment of organic matter content in this way (pg. 1639). FTIR, on the other hand, is a powerful technique for detecting and quantifying the presence of any organic matter and water content (for example see Patwardhan *et al.*, 2006). Furthermore, it can differentiate between proteins (which typically mediate biogenic silica formation) and polysaccharides (predominantly found as protective coatings around the diatom frustule).

9) *Q3 and Q4 bonds (line 29, p 1632) should be defined, and perhaps it would be useful to mention that the ratio of the two gives an indication of hydration (Leng et al., 2009).*

This will be added in the revised version, in summary Q_4 refers to Si-(O-Si)$_4$ bonds and Q_3 to HO-Si-(O-Si)$_3$ bonds

Reviewer 2

The reviewer suggests sections that need further discussion in the manuscript and these will be completed in our revised version.

1) *The analytical reproducibility of replicate FTIR analyses of small sample aliquots is expressed in terms of contamination (p1636, lines 11-13). While this is useful as overall description, it does not show where in the spectra the deviations occur and how strong they really are. Examples from end-members and samples would help to illustrate possible deviations within the analytical limitations.*

The variability in the FTIR spectra is apparent across all wavelengths. In cleaner samples it is focused around the main spectral peak at ~1100 cm^{-1} associated with -Si-O-Si bonds (see figure below). Conversely, in less clean samples the variability is centred over the main spectral "contaminant" peaks.

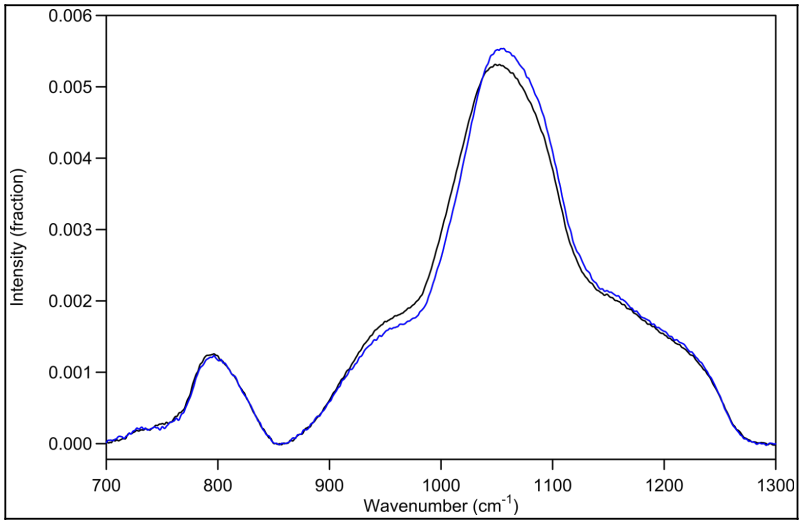

Figure 1: Replicate FTIR spectra for sample #10: FTIR estimates of contamination are 11.8% using the Baikal-"Silt" model with a reproducibility (1σ) of 3.3%.

2) *The definition of end-members is the crucial point of the approach. Two purified diatom standard materials are used as diatom end-members whose degree of contamination was assessed by light microscopy and XRF. In terms of Al2O3 content, contamination of BFCmod is 2.0 wt%, while for PS1772-8 it is given with 0.09% (p1637, line18-19). What is the effect of this contamination of the end-member on the quality of the model? Would it be possible to use a pure diatom opal, e.g. from culture experiments, as the "true end-member" to avoid this source of error. Given the low, but in terms of real samples clear difference in the contamination of the diatom end-members, why does this not lead to larger differences in the estimates of FTIR contamination from these standards (p1637, line 20).*

The presence of small but varying amounts of Al_2O_3 in our diatom end-members are suggested to be an indication of contamination. This is not necessarily the case as diatoms naturally contain Al_2O_3 within their frustules, the amount of which can vary between individual sites (Koning et al., 2007). As mentioned in our reply to reviewer 1 (point 4), the purity of these diatom end-members was assessed and confirmed using other techniques including SEM, light microscopy, XRD and NMR analysis (Chapligin et al., In prep).

3) *How accurate is the definition of contamination by the Al2O3 content as determined by XRF. I am aware of the problem here, but we know that Al is incorporated into the diatom frustule in amounts that could be of relevance for contamination assessment based on this criterion. Beside inorganic contamination, that is the target of Al2O3 analyses, diatom frustules can contain organic matter encapsulated in their frustules. How would this impact the FTIR spectra? Furthermore, at least lacustrine sediments have others sources of biogenic opal like sponge spicules and phytoliths that are not captured by the end-members used but would have to be seen as contamination. Are there alternative methods to quantify inorganic contamination of diatom samples independently to show the accuracy of the definition by Al2O3 content of XRF for selected samples.*
4) *Residuals between the best-fit model and the observed FTIR spectra for any endmember model used are between 2.8 % and 19.4 % (p 1738, line 9). This might be an effect not only of the accuracy of the silt end-member definition but also of the contamination of the diatom end-members. What is the effect of high residuals on the determined degree of contamination in the samples and up to what level of deviation can a sound estimate be given. Even low residuals indicate that the end-members can not completely explain the measured sample FTIR spectra and could possibly indicate also contamination.*

The use of Al_2O_3 and XRF for assessing contamination is becoming increasingly widespread. On the one hand using Al_2O_3 as a tracer is a valid choice as contaminants are often aluminosilicates. On the other this approach is flawed if carbonates or organic matter are present, although high organic matter/carbonate concentrations may be detectable by examining the LOI and Ca data provided by XRF analyses. The FTIR method presented here is, we believe, the first attempt to quantitatively advance this issue and get around this problem, although the method will fail to distinguish between different types of biogenic silica (see response to Reviewer 1 comment 6).

The reviewer suggests that the FTIR residuals are primarily a function of contamination in the diatom end-members. Given the above responses, we argue against this. Instead we suggest that the large variability in residuals between different diatom end-member models almost certainly reflect the fact (particularly for BFC_{mod}) that not all diatoms are fully representative of those found in the Lake Baikal sediment record. This appears to be confirmed when using a Baikal diatom end-member which produces the lowest residuals, implying that the FTIR method works best when using a sites specific end-member [pg. 1638-9]. Where residuals remain even after using the Baikal diatom end-member, we attributed this to the "silt" end-member not being fully representative of the contamination matrix in all samples. Residuals may also be increased by the poor analytical reproducibility for moderately to heavily contaminated samples (Section 3.1). Whilst, as highlighted by the reviewer, these uncertainties may limit the ability to precisely quantify the degree of contamination in an unclean sample when residuals are greater than 7%, the method remains more than capable of distinguishing between clean or unclean material for which analytical reproducibility is less than 1% contamination and residual <5%.

With regards to organic matter, as mentioned to in our response to reviewer 1 (point 8) FTIR is a powerful technique for detecting the presence of organic matter and water content. Diatom intra-cellular organic matter is present in all diatoms and so is accounted for by the diatom end-member used in the FTIR models. However, the absence of an organic matter end-member to account for material external to the frustule may be further contributing to the high residuals in some samples.

References

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