

Interactive comment on “Chemical de-staining and the delta correction for blue intensity measurements of stained lake subfossil trees” by Feng Wang et al.

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We greatly appreciate the comments from Dr. Miloš Rydval and his kind line-by-line suggestions to improve the English expressions. Our responses are listed below:

General comments: The manuscript addresses a highly topical issue related to the dendroclimatic utilization of reflected light (blue intensity) from lake subfossil wood material, specifically a discoloration bias related to staining, which is primarily attributed to Fe oxidation. The study investigates the application of a range of chemical treatment techniques in order to improve light reflectance data by reducing staining bias and also provides additional validation for the applicability of the delta BI correction procedure

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in the context of using samples affected by staining.

Overall, it is a nice, relevant and focused paper building on previous work on this topic. The study is designed and performed in a methodical manner and the manuscript is generally well organized and logically structured. Although I do not have any major comments on the methods used or analysis performed, the (mostly minor) comments detailed below will hopefully help to further refine this work. I would also recommend checking the manuscript to make minor language improvements (for example in order to clarify the meaning of certain statements) and I include specific suggestions (under minor comments) to indicate parts of the text where I believe most improvements could be made.

Specific comments:

L83: what is meant by 'new lake'? as in 'newly sampled lake' (i.e. data from samples from this lake were not analyzed / published before)? – please clarify

Response: We corrected “new lake” to “newly sampled lake”.

L106: Why were the treatment times different for MixC? Please explain briefly.

Response: MixC was the last, but the most efficient and active reagent we used to treat samples. We had known that reactions of other (less active) reagents would terminate in 12–24hrs before using the MixC reagent. We thus used a shorter reaction time (24hrs) for MixC treatments, while we sampled the solutions of MixC more intensively (at 0.5hrs after treatments; Figure S2) in order to capture the change point of reaction (although finally this figure was only placed in the supplementary material). We added a sentence to explain the different time settings “Treatment and sampling times were set shorter for MixC since it was the most active reagent.” in Lines 106–107 in the revised manuscript.

L117-118: Please add relevant reference(s) here.

Response: We added reference to (Rydval et al., 2014).

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L135-138: Please include information about the measurement resolution of the photo sensor used (i.e. the step size along the density measurement profile) – e.g. $10\mu\text{m}$ or variable?

Response: The resolution of the density profile is $10\mu\text{m}$. This information was added at Line138.

L147-148: In what way were they affected – structurally, their color? If this is true for the unstained samples, could this play a role in affecting the properties of stained samples to some degree as well? Undoubtedly, the treatments lead to improvement, but it would also be worth discussing if there are (or could be) some undesirable effects as well that may perhaps limit the observed improvement?

Response: The major effects are related to colors. As we can see from the unstained+living tree group on the right panel of Figure S7 (in the revised supplementary material), raw LBI, DBI, and MXD measurements were similar for MixA, MixB, and Control treatments, while MixC altered the LBI and DBI but not the MXD measurements. We think the bleaching effect of MixC is weak on the subfossil wood since DBI of stained LSTs did not diverge while DBI of living trees and unstained trees diverged. We added a few sentences at the end of the Result section 3.1 (Line 184–187) to discuss this effect. “MixC, the only reagent with bleaching function (Table 1), could have an additional bleaching effect on the wood, resulting in smaller LBI and DBI values in living and unstained trees compared to untreated control (Fig. S7). However, DBI of the stained LSTs was only slightly modified by the MixC treatment (Fig. S7), indicating that the bleaching effect of MixC is weak for the stained samples.”

L150: Was there any apparent difference in the sapwood / heartwood of the subfossil samples and if so, could that then potentially also have some effect on the results?

Response: The sapwood portion of subfossil samples is generally decayed due to their long stay in water/sediments. We thus did not observe such apparent sapwood-heartwood difference on the subfossil samples. In addition, black spruce in general

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does not show apparent color difference between the sapwood and heartwood. Living-tree samples of L20 may represent a special case where this color issue occurred.

L152-153: Just to clarify - 'regional chronologies' here represents different parameters / treatment methods and not the two sites (since those were pooled together)? A slight re-wording might help to ensure that this is clear.

Response: We changed the sentence to "Regional chronologies for each tree-ring parameter (LBI, DBI, and MXD) and treatment (MixA, MixB, MixC, and Control) were generated by pooling standardized series from both sites using the Tukey's bi-weight robust mean." in Lines 153–154.

L263: Slight reformulation is required here since the higher replication is needed to obtain a robust and representative (DBI) chronology rather than a chronology with an equivalent (or similarly strong) climatic signal to MXD (although it is true that these two things usually go hand in hand).

Response: We changed the sentence to "Firstly, a higher tree replication is often needed for DBI than MXD data, in order to obtain a robust chronology (Rydval et al., 2014; Wilson et al., 2019)." in Lines 270–271.

L264: At the same time, DBI appears to calibrate more weakly compared to LWB over the instrumental period. Could you suggest a possible explanation for this? Could you also add some moving window EPS statistics somewhere (e.g. in Figure 6) for the final chronologies used for the reconstruction in order to get a better idea which parts of the chronologies might be stronger / weaker.

Response: We added the moving EPS to Figure 6e in the revised manuscript (See Figure in the supplementary reply letter). We also discussed the more variable EPS of DBI in contrast to LBI and MXD at Line 273.

L278-279: I think this sentence could be reformulated somewhat and expressed more clearly.

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Response: We changed the sentence to “The chemical de-staining experiments, though not satisfactory regarding the robustness of LBI data, suggest that the post-sampling chemical Fe oxidation most likely result in the staining issue.” in Lines 286–287.

Table 2: It would be helpful to clarify a few things. To avoid any possible misinterpretation, it would help to specify in the caption that RGB refers to separate (R, G, and B) color channels rather than for example a full color RGB light values. Also, some more details should be included in relation to no. 3 and 5 to clarify the difference between these two terms – presumably delta RGB refers to all of the color channels whereas DBI only refers to the blue channel? Or is there some difference when it comes to how delta B (in RGB) was calculated compared to DBI? Unless I am mistaken, the settings for calculating DBI (i.e. how LBI and EBI are determined) in Coorecorder are adjustable – the main settings for this calculation should probably also be stated here.

Response: We clarified each parameter in the Table 2 and its caption (see supplementary reply letter). In short, the main difference between No. 1–3 and No. 4–6 is that No. 1–3 provide one R, G, and B value for each measured lath since all tree-ring measurements of each type (R, G, and B) were averaged by lath. Another difference between No.3 and No.5 is that No.3 is based on differencing R, G, and B intensities measured from 100% of pixels in latewood and earlywood, while DBI, i.e. No.5, is based on differencing LBI (measured from 30% of darkest pixels) and earlywood BI (measured from 100% of the pixels).

Figure 1 caption: ‘The gray shades in (a) correspond to . . .’ / ‘The gray shading in (a) corresponds to . . .’. Also, maybe simply use the term ‘replication’ instead of ‘distribution range’ here?

Response: The shading in a) shows the range distribution of black spruce, rather than the replication of trees. We corrected “shade” to “shading” and added the source of the tree distribution map.

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Figure 2: Certainly in the main text and maybe also here (e.g. in the figure caption) define MP-AES. For text in right-middle box consider: ‘Residual iron content per gram of wood . . .’. Also, it may be more suitable to use past tense in the text boxes: e.g. ‘lath1 digested using . . .’; ‘lath 2 rinsed in de-ionized water for 2 hrs, then air dried and sanded’, etc.

Response: We added the full name of MP-AES in the text (at Line 112) and figure caption. The past tense was used in Figure 2.

Figure 3 caption: The description for (a) should make it clear that these are multiple laths from one LST - for example: ‘(a) shows examples of differently treated laths from one LST and one living tree sample (last row)’. When referring to panels (b) and (c), it would be clearer to write it using the following format: (b) description, and (c) description – (instead of (b), and (c) description)

Response: We modified the Figure 3 caption: “Figure 3. Residual iron (Fe) and wood RGB intensities (see definitions in Table 2) of LST laths treated with seven chemical reagents. (a) shows examples of treated laths from one LST sample and one living tree sample (last row). The gray outer part of the example LST is discolored due to decay. (b) shows the mean wood RGB intensities according to treatment. (c) shows the mean concentrations of residual Fe according to treatment. Error bars in (b) and (c) refer to standard deviations of corresponding group. Percentages in (c) refer to the Fe removed by de-staining treatments relative to the Fe concentrations of untreated stained LSTs. (d)–(f) show the linear regressions of earlywood, latewood and delta RGB intensities against the log of residual Fe. Regressions are based only on the LST data (circles). Living-tree data are plotted as triangles but are excluded from the regressions.”.

Figure 6: Consider also adding some statistics in the panels such as the full period calibration. In the caption, maybe also specify the type of filter used.

Response: Done. See the revised Figure 6 in the supplementary reply letter.

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Minor comments: L36: Consider changing to e.g. ‘. . . high cost of X-ray densitometric equipment’ . . . unless this is meant to refer to the relatively high costs associated with processing samples at a ‘facility’ that is equipped to perform X-ray densitometry. In any case, a small edit is needed.

Response: Corrected.

L37: It may be more accurate to state that the production of BI is ‘relatively cheap’ rather than ‘cheap’.

Response: Corrected to “In contrast, BI is more affordable because it uses commercial flatbed scanners and image analysis software to measure the blue light reflectance of tree rings (Rydval et al., 2014)” in Lines 37–39.

L39: ‘coherence’ instead of ‘coherences’

Response: Corrected.

L43: Perhaps be a bit more specific here by changing ‘color issues’ for example to something like ‘color inconsistencies’ or ‘inconsistent color properties’?

Response: Corrected to “heterogeneous colors”.

L44/45: As this is a property of the wood, ‘leading to’ is not really appropriate here – instead perhaps ‘. . . exhibiting darker heartwood than sapwood’

Response: Yes, that is true. We corrected this sentence to “The best-known issue is the sapwood-heartwood color difference of several tree species such as pine and larch, which does not co-vary with density” in Lines 44–45.

L47: ‘occurs’ instead of ‘happens’

Response: Corrected.

L48: ‘LSTs are . . .’?

Response: Corrected.

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L49: just 'replication'

Response: Corrected.

L53: The second part of this sentence could be improved for example along the lines of '. . . to realize the potential of the promising BI technique . . .'

Response: We changed this sentence to "Therefore, it is critical to develop unbiased BI data from a variety of wood materials, in particular from the LSTs, to make the promising BI technique widely applicable in future dendroclimatic reconstructions." in Lines 52–53.

L58: 'consists of' instead of 'consists in'; the word 'delta' can be removed from this part of the sentence

Response: Corrected.

L59: Consider changing 'DBI is suitable to recover . . .' for example to 'DBI suitably represents . . .' or 'DBI corrects for . . .'

Response: Corrected.

L64: The last part of the sentence should be re-phrased e.g. '. . . without utilizing low frequency information of the more temperature-sensitive BI data' / '. . . without benefitting from / exploiting any potential improvements in the low frequency domain from the more temperature-sensitive BI data'

Response: Corrected.

L69/70: Consider changing the last part of the sentence to something along the lines of '. . . to a standard comparable to MXD-based reconstructions'

Response: Corrected.

L80/81: minor change needed here, e.g. '. . . LSTs after falling into the water and eventually becoming buried in lake sediments'

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Response: Corrected.

L88: 'fungal' rather than 'fungi'? Also, is there a better word that could replace 'invasion' here? Perhaps simply 'fungal discoloration'?

Response: Corrected.

L89: 'before the year'

Response: Corrected.

L94: Should this be 'radius' instead of 'radii' if laths are cut along a single radius, otherwise specify the number of radii

Response: Corrected.

L96: This should probably be 'weighed' instead of 'weighted'

Response: Corrected.

L102: I would recommend presenting the figures in the order that they are first mentioned in the text (Fig. S1 is first mentioned here while Fig. S2 is first mentioned in L131).

Response: We have corrected the order of all figures and tables in the revised manuscript.

L108: This could probably be improved slightly – e.g. 'one lath from each pair of laths'?

Response: Corrected.

L111: Please mention first that MP-AES stands for "microwave plasma-atomic emission spectrometer"

Response: Added at Lines 112.

L112: '. . . weight of the corresponding . . .'

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Response: Corrected.

L113: The meaning of this sentence was not immediately clear to me. Consider the following minor revision for the sake of clarity: ‘. . . represent the combined total . . . cannot be separately distinguished by MP-AES’

Response: Corrected to “Fe concentrations in this study represent the total amount of ferrous and ferric Fe, because MP-AES does not distinguish the type of Fe ions.” in Lines 114–115.

L115: ‘grit’ rather than ‘grits’

Response: Corrected.

L118: ‘interference’ instead of ‘inferences’

Response: Corrected.

L122: I think this could be explained a bit better – in a more specific way. Also, ‘consistent with’ rather than ‘consistent to’

Response: We changed the expression to “Because high RGB values represent light colors (i.e. high brightness), they were subtracted from a value of 256 such that smaller RGB values are associated with lighter colors” in Lines 122–123.

L141: perhaps ‘(i.e. each lath pair)’?

Response: Corrected.

L143: ‘An age-dependent spline . . .’

Response: Corrected.

L148/L150: Also in relation to Fig. S8 / S7, see earlier comment about sequential order of figures mentioned above.

Response: We have corrected the order of all figures and tables in the revised

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manuscript.

L151: maybe 'poor (tree) health' would be better than 'unhealthy tree growth'

Response: Corrected.

L154: 'averaged into'? Also, maybe consider showing the individual MXD chronologies in the SI.

Response: Added to SI material, Fig. S9. See Fig. S9 in the supplementary reply letter.

L161: This could use a bit of re-wording.

Response: We changed this sentence to "The reconstructions were based on the scaling method (Esper et al., 2005; Rydval et al., 2017) by adjusting means and standard deviations of the chronologies to those of the temperature target over the 1901–2015 time interval" in Lines 161–163.

L165: maybe slightly re-word – e.g. ' . . . in order to assess the role of Fe in the staining issue'

Response: We changed this to "in order to assess the roles of Fe in the staining issue" in Line 166–167.

L167: Perhaps just briefly specify the advantage of applying this specific filter in the context of high-pass / low-pass filtering.

Response: We used this method because it is the default option of the dplR package, not because of a specific advantage. The filtered series were very similar between using the Butterworth filter and Chebyshev filter.

L168: 'Performance of the reconstructions was assessed ...'

Response: Corrected.

L170: ' . . . while the 1961-2015 period . . . '

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Response: Corrected.

L178-179: Just as a general point, I wonder what the reason for this might be? Could it be related to the ‘color properties’ of the staining caused by Fe?

Response: Yes, this is correct. This is related to the properties of Fe oxides that stained the wood. In general, Fe oxides have stronger red light reflectance. From this aspect, our conclusions are also consistent with this general phenomenon. We feel slightly confused by the real-world colors of the stain on wood, which look more blue than red, however such “blue” stain reflect more red color according to our analysis (Fig. 3b).

L182: ‘intensities’

Response: Corrected.

L186-187: ‘. . . of treated earlywood and latewood’?

Response: Corrected.

L189: Please be more specific here and elaborate on what the difference is.

Response: We changed it to “Note that wood delta BI and DBI were not calculated exactly in the same way (see Table2; delta BI is the averaged difference between BI of entire latewood and earlywood from all tree rings in a sample, while DBI is a tree-ring parameter which presents the difference between LBI and BI of entire earlywood for each tree ring).” in Lines 194–196.

L191: Consider something along the lines of ‘. . . four treatments examined in more detail . . .’

Response: Sorry, but we could not understand this comment

L192: ‘prior to 1900 CE’ or ‘prior to the year 1900 CE’

Response: Corrected.

L193: ‘coherence’ rather than ‘coherences’

Response: Corrected.

L196: 'few differences' instead of 'little differences'

Response: We changed the sentence to "In addition, few differences were found between the control DBI series and chemically treated DBI data, although the colors of wood samples were visually distinct (Fig. 3a)." in Lines 203–204.

L203: 'Briefly' is probably not needed here

Response: We prefer to keep it. The term is suggested by one of the co-author working in geochemistry, because the chemistry of iron is complex.

L208: I would recommend re-wording 'combine to wood' – 'combine with wood' or maybe 'bind to wood'?

Response: We corrected it to "bind".

L225: just 'etc.' instead of 'and etc.'

Response: Corrected.

L228: maybe 'in our samples' rather than 'from our samples'?

Response: Corrected.

L229-230: Minor edit needed here: e.g. 'is not sensitive to sulfur and phosphorous' / 'is not designed to detect sulfur and phosphorous' or 'is not sufficiently sensitive to detect . . .?'

Response: We made some changes to clarify this. "However, the MP-AES instrument is not sufficiently sensitive to verify our hypothesis regarding those Fe complexes (detection limits are ~6500 ppb and 125 ppb for dissolved sulfur and phosphorus, respectively, compared to ~4.6 ppb for dissolved Fe)" in Lines 236–238.

L234: '. . . very little residual Fe . . .'

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Response: Corrected.

L236: probably replace 'such' with 'the'

Response: Corrected.

L240: maybe something like '. . . when staining is present in subfossil wood' would be better

Response: Corrected.

L242: 'a' should be removed

Response: Corrected.

L247: something is missing here – e.g. '. . . for example as with the most efficient MixC protocol' or something similar

Response: Corrected.

L250-251: 'This evidence suggests that DBI is not only an excellent solution to resolving ...'

Response: Corrected.

L251: 'but also efficiently resolves ...'

Response: Corrected.

L256-257: The last part of this sentence could be re-worded and expanded a bit to clarify what is meant by this.

Response: We made some corrections. "Although DBI is theoretically sufficient to solve the sapwood-heartwood color issue (Björklund et al., 2014), in our case it could only partially correct this problem (Fig. S8). Old living trees were collected from lakeshore forests at the L20 site and they often displayed declining ring widths compared to healthy trees sampled later at the same site (not shown). DBI of L20 is likely

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influenced by these narrow tree rings (Björklund et al., 2019) because DBI of black spruce is not only correlated to MXD but also to the ring-width data (Wang et al., submitted). We thus speculate the divergence of DBI reflects mostly a specific issue related to the declining growth of unhealthy trees.” in Lines 262–267.

L258: replace ‘declined’ with ‘declining’

Response: Corrected.

L259: ‘decline’ instead of ‘declines’? Also, the reference Björklund et al., 2019 (Reviews of Geophysics) may also be relevant to this point.

Response: Corrected and reference added.

L261: ‘need to be’ rather than ‘need be’?

Response: Corrected.

L268-269: ‘. . . further attention / investigation is needed . . .’

Response: Corrected.

L281: should probably be ‘. . . used as part of Fe extraction protocols . . .’

Response: Corrected.

L282: ‘which also face / are also susceptible to / also suffer from’?

Response: Corrected.

Please also note the supplement to this comment:

<https://www.biogeosciences-discuss.net/bg-2020-102/bg-2020-102-AC1-supplement.pdf>

Interactive comment on Biogeosciences Discuss., <https://doi.org/10.5194/bg-2020-102>, 2020.

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