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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 are grateful for the useful comments from Dr. Jesper Björklund. Our responses are listed below:

The manuscript by Wang et al presents a very interesting sample material for temperature reconstructions and examine how to best utilize this in conjunction with the popular and affordable BI technique. The paper is foremost dedicated to a very novel and clever de-staining experiment which I thoroughly enjoyed and have the potential to be highly cited in future BI studies. The second component was a careful comparison of LBI, DBI and MXD from parallel X-ray measurements to evaluate the performance

of the chemical de-staining and LBI and DBI parameters with MXD as reference. Although the authors conclude that the simple DBI was more successful in replicating the low-frequency variance of the MXD, they have made some very important discoveries in terms of de-staining of relict wood material. The DBI parameter appears to be quite successful, but has documented problems as the authors also mention in the final sentences. Therefore, all tools available for de-staining prior to DBI transformation must be considered of great value. I congratulate the authors to a fine, and from what I can tell labor intensive, experiment and I consider the manuscript suitable for publication following minor revisions and clarifications. I also look forward to learn more about the planned follow-up manuscript.

Detailed comments: L32 We would not say BI is recently developed anymore, it has been around almost 20 years now.

Response: Yes, we totally agree. We removed "recently developed".

L32-33 The BI technique is an alternative to the X-ray technique in producing proxy parameters such as MXD.

Response: We re-phrased the first sentence as: "The blue intensity (BI) technique is an alternative to the more expensive X-ray densitometric methodology in producing tree-ring proxy parameters such as maximum latewood density (MXD) for dendroclimatology." in Lines 32–33.

L37 -38 Consider changing to something like: "In contrast, BI is more affordable because of the utilization of commercial flatbed scanners to generate images of reflected blue light analyzed in potentially affordable image analysis software. . ."

Response: We re-phrased this sentence as "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." in Lines 37–39.

L38-42 Strange sentence, some of the studies encouraging more studies were made

later than the encouraged studies. Work a bit more on this sentence and consider also these references: Björklund et al., 2014, 2015; Dolgova, 2016; Fuentes et al., 2018; Kaczka et al., 2017; McCarroll et al., 2013; Rydval, Gunnarson, et al., 2017.

Response: Several of the suggested references are cited elsewhere in the manuscript. Here we only wish to cite studies specific to latewood BI rather than delta BI, because the topic of delta BI is detailed in the next paragraph. So we only added McCarrol et al., 2013. We improved the sentence to make it more fluent. "Excellent coherence was reported between the latewood BI (LBI) and MXD data measured from living-tree materials of a number of coniferous tree species across the northern hemisphere (Campbell et al., 2007; Kaczka et al., 2018; Österreicher et al., 2015; Rydval et al., 2014; Wilson et al., 2014), suggesting the potentials to use BI method in dendrocliamtic reconstructions (McCarrol et al., 2013; Rydval et al., 2017; Wilson et al., 2019)." in Lines 39–42. The later three references (McCarrol et al., 2013; Rydval et al., 2017; Wilson et al., 2019) are real reconstruction works.

L45 Should perhaps add something like: "..not accompanied by a similar difference in density.."

Response: I 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.

L83 newly exploited lake?

Response: This has been changed to "newly sampled lake".

L84 millennium-long?

Response: We think the meanings of "millennium-long" and "millennial" are very similar. We keep "millennial" here.

L96 What was the purpose of the weighing? Were the laths also weighed after the chemical analysis? Could not find any more use of these measurements in the

manuscript

Response: The weights of wood laths were used to calculate the Fe concentrations in the wood. This is because Fe concentrations in wood depend strongly on the weights of wood laths. The iron data shown in the original manuscript had already been adjusted by the wood weights. It was described at Line 113 of the revised manuscript, "data were adjusted to "milligram of Fe per gram of wood" according to the dilution and weight of the corresponding wood lath.". We only weighed a few post-treatment wood laths. This is because we set two control groups in addition to the chemical treatments, i.e. untreated stained samples and living tree samples. For these samples, we would not know their post-treatment weights. A consistent measure of weight from laths should be used, i.e. pre-treatment weight. Second, the amount of wood burrs contacted on the surface of laths might also affect the post-treatment weights. These burrs were often found detached from the laths after treatments (samples are placed in a tube on a shaker for at least 24 hrs). This mass loss varies tree by tree, and we cannot quantify the weight losses. For example, being similarly treated with MixC, one lath lost 0.2% of weight relative to a pre-treatment weight (0.2866g) while another lath could loss 1.03% of its weight (pre-treatment weight was 0.1935g).

L104 .., to identify the most effective. . .? Remove "(see results below)". The results are always be presented after the methods description.

Response: Ok, we removed "(see results below)".

L118 sensu Rydval et al., 2014?

Response: We added this reference here.

L118-119 Great initiative L121-122 Very strange statement. Real world observations? Do you mean: lower RGB values corresponds to lighter densities?

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 122–124.

Section 2.2, 2.3 and 2.4 Consider re-structuring here. Perhaps one section for chemical de-staining description. One section for BI and X-ray data development and one section for chronology development for climate analysis, and sample average RGB data?

Response: The manuscript comprises a de-staining experiment and a dendroclimatic assessment. Accordingly, we kept section 2.2 and merged sections 2.3, 2.4 and 2.5 to obtain one section for the de-stanning experiment (section 2.2) and one section for the dendroclimatic assessment (section 2.3)

L139 Did you use the full RGB spectrum or only the blue spectrum? If the latter, it is consistent with the use of BI based parameters. Same comment in L165.

Response: We extracted the red, green and blue intensity values from earlywood and latewood of each ring, then averaged values by colors and laths. Of course, for the LBI and DBI we considered only the blue spectrum. A revised version of Table 2 (in the supplementary reply letter) clarifies the measured parameters. We could not find the comment in L165.

L145 N.B. residuals are most often used for density related parameters. This is not a major problem here since you compare results from BI and X-ray, but may be important in pure climate reconstructions.

Response: Ok, we will think of calculating residuals in future reconstructions.

L168 "coherence" can also be a type of statistical analysis, perhaps change to the more general term of "agreement", or simply not explain correlation since more or less the entire readership is familiar with this.

Response: We changed it to "agreement".

Figure S4 Spelling of replication

Response: Corrected.

Figure s6 spelling of earlywood. It seems odd that the area of the 30% of the darkest pixels in the latewood are differently sized even though the latewood area is roughly the same (compare ring 4 and ring 5). Please check the definition you used and clarify why this is the case.

Response: Misspelling Corrected. We systematically used the 30% of the darkest latewood BI as LBI. However, the Figure S6b was generated using the densitometer function of CooRecorder (See Figure1 in the supplementary reply letter). We replaced this figure with the output of the actual LBI measurements (Figure2 in the supplementary reply letter).

L169-171 Would be great to have running Rbar or EPS, to evaluate the difference between the different parameters. Perhaps this can explain why the DBI perform so badly in the post 1960 period compared to LBI and MXD. Both in terms of trend and correlation.

Response: We added EPS values in the revised Figure 6 (in the supplementary reply letter). Some relevant discussions were made on the EPS data (at Line 273 in the revised manuscript). It is true that EPS of DBI was more unstable than LBI.

L182 spelling intensities

Response: Corrected.

Figs. S7-S8 Would be interesting to also present the Earlywood measurements. Would be even more interesting if you also presented Delta density and Earlywood density. It is puzzling why LBI and DBI has such similar trends in S7. Is there a HW/SW transition in these trees, if so why so weak in the earlywood? Are the rings in the post 1960 period very narrow? If so, I think that your measurement resolution is causing some problems here. Consider that the measurement resolution is affecting your latewood measurements more than your earlywood measurements. That is, your latewood BI

is deflated because of adjacent contamination of earlywood BI. Ergo the delta BI will be artificially lowered and similar in trend to LBI. Not completely relevant to your nice study, but could not resist :)

Response: We did not extract the earlywood BI and earlywood density data for this manuscript, which are in very raw forms, and consequently delta density data are not available. We responded to the rest of this comment below, under the L253–260 comment.

L196 check grammar

Response: We changed the sentence to "In addition, few differences were found between the untreated control DBI series and chemically treated DBI data, although colors of wood samples were visually distinct (Fig. 3a)" in Lines 204–205.

L208 combine to wood? Not clear, rephrase..

Response: We replaced "combine" with "bind".

L241-242 This is not surprising. If you would calculate delta density and correlate with delta BI you would probably find equally high correlation as between LBI and MXD. This is not needed in revision, I am merely pointing this out.

Response: OK. We deleted this sentence.

L253-260 I think you are right that the narrow ring widths are causing the problem here, but I would not say it is a healthy versus unhealthy tree problem. It is a problem of measurement resolution (see comment above for fig s7). Healthy tree can also have narrow rings.

Response: Yes. The L20 samples showed narrow rings and distinct sapwood-heartwood colors. This latter phenomenon is unusual for black spruce because this species usually does not show distinct boundary between earlywood and latewood. We speculate that this phenomenon was resulted from the unhealthy state of some

trees in this site because in the early stages of sampling, we collected trees as old as possible, regardless of their crown shape and growth rate. We agree that the cause of the divergence (in particular for the DBI which in theory is not affected by color) is most likely the narrow rings. We thus modified Lines 155–160 in the original manuscript to:

"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 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 263–267.

L262 yes interesting observation. Would be better underpinned if you also presented the rbar for all the parameters.

Response: We added moving EPS values in Figure 6 (see revised Fig. 6 in supplementary reply letter).

Please also note the supplement to this comment:
https://www.biogeosciences-discuss.net/bg-2020-102/bg-2020-102-AC2-supplement.pdf

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