Dendrochronologia 69 (2021) 125880

Contents lists available at ScienceDirect

Dendrochronologia

journal homepage: www.elsevier.com/locate/dendro

ORIGINAL ARTICLE

The utility of bulk wood density for tree-ring research

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ARTICLE INFO

Keywords:
Gravimetric-volumetric wood density
Dendroclimatology
Biomass
Blue intensity
X-ray density

ABSTRACT

Bulk wood density measurements are recognized for their utility in ecology, industry, and biomass estimations. In tree-ring research, microdensitometric techniques are widely used, but their ability to determine the correct central tendency has been questioned. Though rarely used, it may be possible to use bulk wood density as a tool to check the accuracy of and even correct microdensitometric measurements. Since measuring bulk wood density in parallel with X-ray densitometry is quickly and easily done, we suspect that its omission is largely due to a lack of awareness of the procedure and/or its importance. In this study, we describe a simple protocol for measuring bulk wood density tailored for tree-ring researchers and demonstrate a few possible applications. To implement real-world examples of the applications, we used a sample of existing X-ray and Blue Intensity (BI) measurements from 127 living and dead *Pinus sylvestris* trees from northern Sweden to produce new measurements of bulk wood density.

We can confirm that the central tendency in this sample material is offset using X-ray densitometry and that the diagnosis and correction of X-ray density is easily done using bulk wood density in linear transfer functions. However, this approach was not suitable for our BI measurements due to heavy discoloration. Nevertheless, we were able to use bulk wood density to diagnose and improve the use of deltaBI (latewood BI – earlywood BI) with regard to its overall trends and multi-centennial variability in a dendroclimatological application. Moreover, we experimented with percent of latewood width, scaled with bulk wood density, as a time- and cost-effective proxy for annual ring density. Although our reconstruction only explains about half of the variation in ring density, it is most likely superior to using fixed literature values of density in allometric equations aimed at biomass estimations.

With this study, we hope to raise new awareness regarding the versatility and importance of bulk wood density for dendrochronology by demonstrating its simplicity, relevance, and applicability.

1. Introduction

Bulk wood density measurements are recognized for their utility in determining optimal end-uses of wood such as for construction or pulp (e.g., Barnett and Jeronimidis, 2003), as multiplier to determine woody biomass from volume estimations of trees and forests (Saranpää, 2003; Pretzsch et al., 2018), and for understanding the functional and competitive traits of tree species (e.g., Chave et al., 2009). Yet, its utility could potentially be expanded. In tree-ring research, microdensitometric techniques are widely used (Björklund et al., 2019). For example, maximum latewood density (MXD) has become a popular measurement parameter due to its close relationship with past temperatures (George and Esper, 2019), and total ring density (TRD) is increasingly used when deriving annual biomass growth estimates from tree rings (e.g., Babst et al., 2014; Bouriaud et al., 2015; Vannoppen et al., 2017; Zeller et al., 2017). However, the ability of microdensitometric techniques to reflect the correct central tendency of both TRD and MXD has been questioned (e.g., Björklund et al., 2019; Zeller et al., 2017). If mean values are

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https://doi.org/10.1016/j.dendro.2021.125880
Received 5 May 2021; Received in revised form 19 August 2021; Accepted 29 August 2021
Available online 2 September 2021

inaccurate, this would have serious implications for MXD-based climate reconstructions made up of differently sourced materials (e.g., Melvin et al., 2013; Zhang et al., 2015), and obviously bias annual biomass estimations using TRD (e.g., Pretzsch et al., 2018). For these applications, bulk wood density may serve as a simple diagnostic tool (*sensu* Zeller et al., 2017). Bulk wood density, here defined as gravimetric/volumetric wood density i.e., weight divided by volume at a given moisture content, or specific gravity (e.g., Williamson and Wiemann, 2010), is easy to measure and fast and affordable; it is not, however, capable of diagnosing the density of specific rings, nor of sub-annual parameters such as MXD (but see Ifju et al., 1965). Independent validation of X-ray density is largely overlooked in dendroclimatic studies (but see Lenz et al., 1976), although often included implicitly or explicitly in tree-ring-resolved studies of wood biomass and wood quality (*sensu* De Ridder et al., 2011; Evans, 1994; Zeller et al., 2017). Because measuring bulk wood density in parallel with X-ray densitometry is so simple and fast, we suspect that its omission is largely due to a lack of awareness regarding the protocol and/or its importance. With this study, we hope to raise awareness and to demonstrate novel ways of using bulk wood density in conjunction with related dendrochronological applications.

The microdensitometric technique of Blue Intensity or Blue Reflectance (BI; McCarroll et al., 2002) is becoming increasingly popular in dendroclimatic studies. The BI technique has repeatedly been shown to produce information that is nearly identical to that of MXD using corresponding parameters such as latewood BI (LBI) at inter-annual scales (e.g., Björklund et al., 2021; Campbell et al., 2007; Kaczka et al., 2018; Rydval et al., 2014). However, at multi-decadal and multi-centennial scales, LBI is challenged by a multitude of factors: systematic differences in reflection between heartwood and sapwood, areas with “sapstain” or “bluestain fungus”, areas with poor surface preparation, reflectance differences between dead and living material due to preservation conditions of dead wood, etc. (Björklund et al., 2014; Rydval et al., 2017; Wang et al., 2020; Wilson et al., 2017). Thus, diagnostic tools to evaluate the biasing effects of these factors should be welcome. A commonly used approach to mitigate localized within-sample discolorations is to derive the deltaBI (e.g., Björklund et al., 2014; Cao et al., 2020; Buckley et al., 2018; Tsvetanov et al., 2020; Reid and Wilson, 2020; Seftigen et al., 2020; Wilson et al., 2017). This parameter relies on the hypothesis that the discoloration in the earlywood and latewood of every ring is similar. Thus, subtracting the earlywood BI from the latewood BI cancels out the discoloration, but not the desired signal of the latewood. This has proved successful to some degree. However, it has limitations when applied to sample material with distinct color differences between dead wood and living wood (Björklund et al., 2014), and where signals are similar in earlywood and latewood (e.g., Blake et al., 2020). DeltaBI trend corrections are sometimes found to overcompensate as compared to their X-ray-based delta density (deltaXD) counterparts, and further adjustments have been proposed with encouraging results (Björklund et al., 2015; Fuentes et al., 2018; Linderholm et al., 2015). However, the modified deltaBI protocol has not been widely adopted by the dendroclimatic community, perhaps because it appears to be too elaborate and because every new species and climatic growth-limitation may need corresponding X-ray measurements for fine-tuning and validation. A simple diagnostic tool used in conjunction with the deltaBI parameter, such as bulk wood density measurements, would likely be more appealing if it was able to independently constrain the lower-most frequencies.

Denser spatial networks and greater replication of trees are needed to create robust data for studies of the biomass accumulation of forests than are needed for climate calibration exercises (Babst et al., 2018; Nehrbass-Ahles et al., 2014; Pretzsch et al., 2014). If TRD is a meaningful modulator of annual wood volume estimations into biomass, TRD quantifications would benefit from being fast and affordable. In fact, TRD is strongly related to latewood percent (LWpct) both within a tree, as well as within a tree ring (Pretzsch et al., 2018). The strong relationship stems from the fact that latewood and earlywood most often have starkly different densities (Saranpää, 2003). The inter-annual variability in both latewood and earlywood density are relatively modest in comparison. By clarifying the relationship between bulk wood density and LWpct, it should be possible to obtain proxy estimates of TRD. This approach could be cross-validated by performing X-ray densitometry on the same sample material. Moreover, BI measurements are traditionally not calibrated into wood density because such transformations are questionable due to the biasing influence of discoloration. For annual ring BI to be considered a proxy for TRD, there must be independent diagnostics to evaluate the relevant frequencies where BI commonly fails. This could possibly be done with bulk wood density. If successful, estimates from this approach could also be used as a proxy for TRD.

With this study, we would like to emphasize the simplicity, relevance, and applicability of bulk wood density measurements for tree-ring research. 1) We will demonstrate its simple assessment by providing an illustrated example of a bulk-density measurement protocol. 2) The relevance will be addressed by exploring uncertainties in mean levels of MXD and TRD. 3) The applicability will be demonstrated by i) outlining a bulk-density-based correction of TRD and MXD, ii) exploring the diagnostic capability of bulk wood density for deltaBI datasets, and iii) show-casing the potential of using calibrated LWpct or annual ring BI data as proxies of TRD. The demonstrations and experiments are conducted using an >800-year-long *Pinus sylvestris* L. dataset comprised of both living and dead trees from northern Sweden.

2. Materials and methods

2.1. Wood material and tree-ring densitometric data

Wood material from 36 living and 91 dead Scots pines (*Pinus sylvestris*) sampled in 2011 (first presented in Björklund et al., 2014) was revisited for additional analyses in this study. We chose this sample material because it has previously been analyzed for both X-ray-based and BI-based measurements (on the same samples) (Björklund et al., 2014).

The trees were sampled at a temperature-limited site in northern Sweden, just below the tree line at 500−600 m a.s.l. (Lat. 66.2 ° N, Lon. 18.1 °E). The full dataset covers 897–2010 CE, but here we analyze data for the period 1200–2010 CE because the sample depth during this period consistently yields EPS values above 0.85 (Wigley et al., 1984). The X-ray- and BI-based microdensitometry measurements were performed on the same core samples (one sample per tree). A twin-blade saw was employed to create approx. 1.2 mm thick laths for the X-ray analysis. The leftover wood pieces were subsequently sanded for BI analysis. The X-ray-based density measurements were produced using an Itrax multiscanner from Cox Analytical Systems (www.coxsys.se). The samples were prepared according to standard techniques (Schweingruber et al., 1978) following the protocol outlined in Gunnarson et al. (2011). The BI data were produced following the protocol from Campbell et al. (2011), with some modification. Digital images retained for BI analysis were produced with a flatbed scanner with a retailer-specified resolution of 1600 dpi (Epson Perfection V600 Series) calibrated with SilverFast Ai professional scan software using an IT8.7/2 color calibration target. Both the X-ray images and BI images were analyzed with the software WinDendro using an analysis track width of exactly 1 mm for each sample. For this study, we retained the parameters of MXD, earlywood density, TRD, and deltaXD (the difference between MXD and earlywood density), as well as LBI, earlywood BI, annual ring BI, and deltaBI (the difference between LBI and earlywood BI). Note that we always refer to the absorbed BI values that are positively correlated with wood density. Moreover, annual ring density and annual ring BI were integrated over the entire cores into X-ray-based sample mean densities and sample mean BIs, respectively. For more information about the tree-ring material and data production, we refer to Björklund et al. (2014). Before the analyses, the X-ray and BI data were compiled into chronologies as arithmetic means of raw, unstandardized data, or after treatment with regional curve standardization (RCS; Briffa et al., 1992). RCS was only used when chronologies were compared in a dendroclimatological context. RCS is an approach intended to preserve multi-centennial-scale variability in tree-ring chronologies. In this study, we created the regional mean curves by fitting cubic smoothing splines with a 50 % frequency response cut-off at 100 years (Cook and Peters, 1981) to the raw average of the age-aligned sample measurements. This function was subsequently subtracted from each raw data series to derive indices that were averaged based on calendar dates into chronologies.

2.2. Determination of bulk wood density

The bulk wood density measurements were derived from the laths (127 in total) that were previously used for the X-ray analysis (see Fig. 1 for the protocol used in this study). We were thus able to conduct new bulk wood density determinations on exactly the same laths as were used for the X-ray analyses. However, if samples were partly degraded by fungi or insects, these pieces were cut off with a scalpel. The number of rings retained for bulk wood density was roughly matched with the rings analyzed for X-ray and BI; that is, there may be some minor discrepancies in ring counts, on the order of 0–2 % fewer rings for the bulk wood density measurements. Moreover, laths were roughly 10 mm in tangential width (measured along the tree ring), but deviations (±3 mm) from this frequently occur even within one sample. If laths were fractured or sectioned into smaller pieces, all pieces were analyzed together. The total individual tree lath volumes ranged from 0.2 to 2 cm^3, with a mean *and* median of 1.0 cm^3. Both the weights and the volumes were determined for samples acclimatized at approx. 50 % relative humidity and at a temperature of approx. 20 °C. Note that literature specifies a range of different configurations for sample conditions (Williamson and Wiemann, 2010). It is important to always specify sample conditions because results may vary if they are altered.

Following the protocol in Fig. 1 and documentation and calculation example in Table 1, we derived measurements of bulk wood density. First the weights of the samples were determined with a weighing scale to the nearest centigram (Fig. 1b). Note that the water beaker can be on the scale at all times as long as the Tare function is used. The bulk volume, defined as the volume of the solids in each sample *and* the voids within the sample by the American Society for Testing and Materials, was subsequently determined using liquid displacement (Hill and Papadopoulos, 2001), here with regular cold tap water (Fig. 1c-e). Distilled water at a temperature of 4 °C (1.00000 g/cm^3) would be ideal but is not strictly necessary in this context, as the use of water at 21 °C (0.99802 g/cm^3) results in a minor underestimation of 2‰. Considering that tap water in Birmensdorf, Switzerland, where the measurements were done, contains dissolved minerals that increases the density, the overall discrepancy should be even smaller. Nevertheless, to fully remove these uncertainties, we recommend the use of cold distilled water when available. The volume measurement is accurate if done with

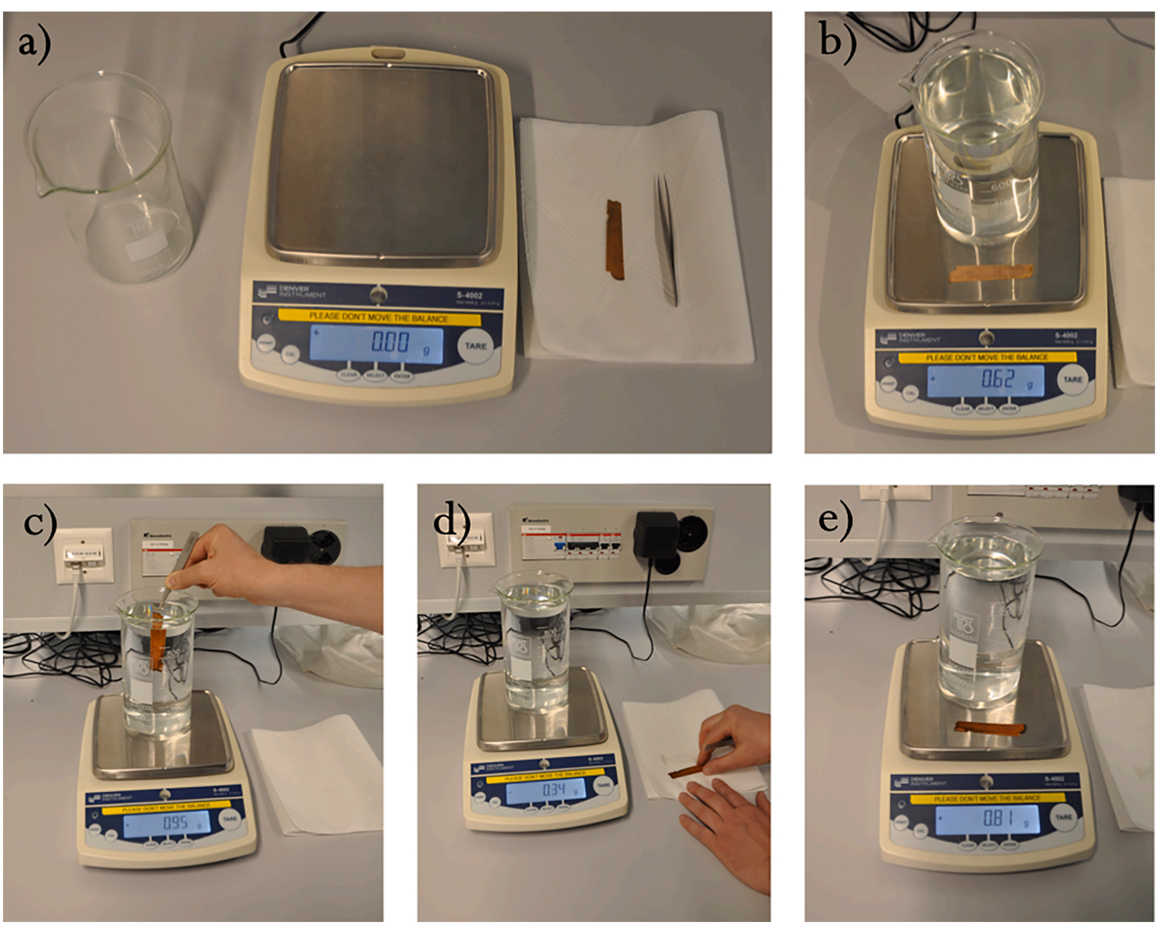

Fig. 1. Series of photographs illustrating a simple protocol for the determination of bulk wood density a.k.a. gravimetric/volumetric wood density. a) All hardware needed (from the left: beaker, scale, paper towel, sample, and tweezers). b) Place the sample on the scale and make a reading of the weight (remember to always tare the scale before weighing the sample). c) Tare and fully submerge the sample in the water using tweezers or a needle, shake off air bubbles, and make a reading of the weight. d) Take the sample out of the water and place it on the paper towel. Quickly wipe excess water off the sample. e) Tare before replacing the wet sample on the scale and make the final reading.

Table 1
Example of a spreadsheet for the determination of bulk wood density complemented with X-ray density and BI for the same sample. Note the sample ID in column A. B: Record the scale reading of the sample dry weight (Fig. 1b). C: Note the weight of the sample when fully submerged in water (Fig. 1c). This is equal to the displaced volume of water, since water ideally has a density of 1. D: Note the weight of the wet sample (Fig. 1e). E: The amount of water absorbed during the submersion is obtained by subtracting the dry sample weight from the wet sample weight. F: The sample volume F is then equal to the displaced water volume plus the absorbed water volume. Remember to also subtract the immersed volume of the tweezers or needle. Finally, the density can be calculated as the dry weight divided by the volume in G. Ideally, this value should correspond to the measurement obtained through X-ray densitometry in H. If sample discolorations are minor, the relationship between samples analyzed with BI (column I) should be in agreement with the relationship between samples analyzed for bulk wood density. If sample discolorations are severe, the relationship may be heavily affected.

A Sample ID	B Dry sample weight	C Displaced water	D Wet sample weight	E Absorbed water **D – B**	F Sample volume **C + E**	G Bulk wood density **B / F**	H X-ray density measurement	I BI measurement
Tree 1	0.35	0.72	0.69	0.34	1.06	0.33	0.365	86
Tree 2	…							

care, although precision diminishes with decreasing sample volumes. This is because the measurement error caused by air bubble formation, absorption, and the adhesion of water to the samples is relatively larger for small samples. Finally, the densities were calculated by dividing the weight by the volume (see Table 1 for bulk wood density calculations).

2.3. Using bulk wood density to diagnose and correct microdensitometric measurements

Lenz et al. (1976) explored the accuracy of the mean levels of density derived from the X-ray system later known as the Walesch technique (Eschbach et al., 1995). They regressed integrated measurements of annual ring density against bulk wood density and found a strong linear relationship that is nearly parallel to the 1:1 slope, but slightly offset. The more advanced and spatially explicit, albeit indirect, X-ray technique was consequently verified using bulk wood density with so-called "correction factors" (Lenz et al., 1976). In this study, we similarly regressed sample X-ray density against bulk wood density and used the Pearson correlation coefficient, slope, and intercept as diagnostics of the X-ray measurements. If the correlation coefficients are high enough to make a correction worthwhile, we, in contrast to Lenz et al. (1976), use the slope and intercept in a transfer function to correct mean values of all TRD and MXD values. If the correlation is low, it is still possible to obtain corrected values on average, but the uncertainty in each corrected value will be substantial and the variance of all values will be underestimated. We also subjected the BI-based measurements to this analysis. Because we expected the correlation between sample BI and bulk wood density to be poor, we identified cohorts in which sample BIs were presumably more different from each other than from corresponding cohorts of bulk wood density data. Visually, the dead wood material in this study appeared to be darker in color than the more recent material (standing dead and living material). However, we had no reason to suspect that this represented a similarly pronounced difference in density. Fig. 2 exhibits four different wood samples of roughly similar densities despite their color differences. Using the mean sample BI as a point of separation, we split the samples into one dark and one light cohort. This division represents the division of dead wood/recent wood quite well, with some exceptions. In total, the split resulted in 74 dark samples and 53 light samples. We then compared the relative differences between the sample BIs and bulk densities of the dark and light cohorts. The relative difference of the sample BIs and bulk densities were derived by normalizing the complete datasets before dividing into the cohorts. We then drew random sets of 50 samples (with replacement) from the light and dark BI cohorts and calculated the difference. This was repeated for the bulk wood density data. We then performed a student's *t*-test to determine if the difference between the light and dark BI samples was statistically different from the difference between the bulk densities from light and dark samples. We repeated this process 1000 times, then summed all the iterations for which the null hypothesis could be rejected to determine if the difference was significant ($p < 0.01$ corresponds to >990 rejections of the null hypothesis).

If the difference between the bulk densities of the two cohorts is statistically different from the difference between the sample BIs of the same cohorts, we are obliged to use two independent RCS in the following analyses of all BI parameters. The different cohorts could then be spliced together by adjusting the mean of the dark segment to the mean of the light segment over their common overlap (here the overlap averages were calculated for periods for which both dark and light chronologies had a replication of >10 trees). That is, the *overall* means of the different cohorts do not necessarily have to be the same, thus overcoming the segment length curse (Cook et al., 1995).

2.4. Potential use of latewood percent and annual ring BI as proxy estimates of TRD

If sample BI measurements are highly correlated with bulk wood density, it should be possible to use the regression slope and intercept to transform BI measurements into wood density and thus create a proxy for TRD that could be used to scale volume estimates into biomass. However, if the relationship is weak, then the bulk wood density

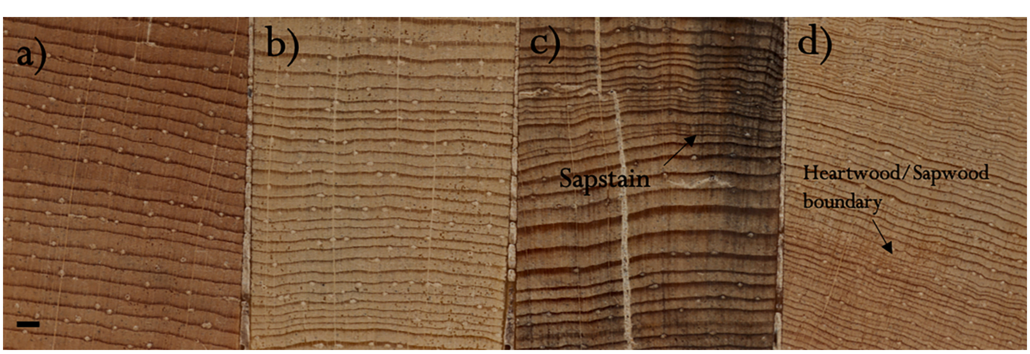

Fig. 2. Photographs of wood samples with similar overall densities but large differences in BI. Each sample is roughly 1 cm wide in the tangential direction (scale bar is 1 mm). a) Dead wood samples are generally darker in color; that is, they have higher absorbed BI than b) living wood samples, but do not necessarily have higher densities. c) Dead wood sample with fungal staining, resulting in high values of absorbed BI locally. d) Living tree sample with the typically light sapwood and slightly darker heartwood but no substantial difference in density between the two elements.

measurements have been useful to advise against this transformation. In exactly the same way, it is possible to regress sample LWpct against bulk wood density to explore their relationship and LWpct as a proxy for TRD. In this study, the transition from earlywood to latewood was defined as the halfway point between minimum and maximum density in each ring (the default option in WinDendro). The latewood width was then divided by the full ring width into LWpct per ring, but also integrated to LWpct per sample. The transformations of LWpct and annual ring BI into TRD were done depending on the robustness of the correlations with bulk wood density.

3. Results

The relationship between bulk wood density and X-ray-based sample density is robust and linear at $r = 0.9$ (Fig. 3a). However, the X-ray-based density measurements are consistently denser than the bulk wood density measurements. The overestimation in the raw X-ray data is about 10 %. Fortunately, the strong linear relationship between bulk wood density and sample X-ray density allows for a correction (Fig. 4). To correct the X-ray data, we applied the transfer function with the regression coefficients from Fig. 3 (*sensu* Björklund et al., 2019). The relationship between bulk wood density and sample BI data is highly significant but much weaker at $r = 0.4$ (Fig. 3b), indicating that BI relative density measurements are influenced by biased sample cell wall colors. Transforming annual ring BI into ring density using the transfer function in Fig. 3b is therefore not recommended for this discolored sample material.

A qualitative evaluation of the BI samples suggest that dead wood samples are generally darker than samples from living, or recently living, trees (Fig. 2). Using the dark and light cohorts, which largely represent dead wood and living wood, the difference in BI is significantly different from the difference in bulk wood density using the same cohorts according to a Student's *t*-test ($p < 0.01$; see also Fig. 5a for their relative distributions). Fig. 5b displays the less distinct distributions of bulk densities from light and dark samples. Because of these results, it is important to treat BI parameters from dark wood separately from light wood in further data analyses. However, even when we separated light and dark measurements of sample BIs, the correlation of sample BI and bulk wood density stayed low ($r < 0.4$ for both light and dark wood; results not shown). Sample LBI scattered against sample means of MXD also exhibit low correlation ($r = 0.43$) for the dark material but higher correlation ($r = 0.72$) for the light material (Fig. 5c). Minor discolorations still distort the relationship between LBI and MXD at the sample level, particularly for the darker cohort. However, if we transform the LBI and MXD parameters to deltaBI and deltaXD parameters, transformations that are designed to neutralize minor color/density differences not related to cell wall dimensions within each wood sample, the correlation coefficients rise for both dark and light wood at the sample level to $r > 0.8$ (Fig. 5d). Thus, LBI will still be biased from sample to sample, even if dark and light materials are separated, but deltaBI much less so. Note, however, that deltaBI from light samples still show tendencies to cluster at higher deltaBI values, whereas deltaXD for the same samples do not exhibit such a strong tendency. It is thus appropriate, even necessary, to keep deltaBI measurements for light and dark wood separated when employing RCS-like standardization techniques.

In Fig. 6a, we show that LBI and MXD have starkly different overall and centennial trends if standardized with one regional curve each. This is due to the discoloration of the dead wood compared to the more recent wood, but also because of smaller discoloration differences within the dead wood and more recent wood (Fig. 2). If we separate the dark and light material and standardize them with two separate regional curves, the overall trend similarity of LBI to MXD is much improved and the decadal to centennial variations are similar (Fig. 6b). The minor discrepancies on multi-decadal scales are likely related to the biased sample average of LBI, predominantly for the darker material (Fig. 5c). There are also within-sample variations due to e.g., heartwood/sapwood differences (Fig. 2). The overall trends are again different when we transform LBI and MXD into deltaBI and deltaXD, respectively (Fig. 6c). The overcompensation of the deltaBI parameter is related to the artificially reduced difference between latewood and earlywood BI in darker wood, described in Björklund et al. (2015). Applying separate regional curves to the dark and light material removes the overcompensation and yields a deltaBI chronology similar in trend and multidecadal scale variations to those of the deltaXD chronology (Fig. 6d). Moreover, deltaBI and LBI treated with two RCSs are similar to a proximal state-of-the-art multi-proxy temperature reconstruction for northern Fennoscandia (McCarroll et al., 2013), which is comprised of data sampled north (Lat. 68–69 °N, Lon. 16–34 °E) of the sample site of this study (Fig. 6e and f). The relationship between the deltaBI chronology and the McCarroll et al. reconstruction is slightly stronger than that between the reconstruction and the LBI. The correlation coefficients are consistently higher regardless if high-pass filtered, unfiltered, or low-pass filtered data are used (Fig. 6e and f). Moreover, the increase in temperatures from 1900 to 2010 CE is not fully captured in the LBI record, likely due to the dominance of the lighter sapwood in the recent material, which offsets the expected positive trend.

The relationship between sample LWpct and bulk wood density is highly significant, where LWpct explains 43 % of the variation in sample density (Fig. 7a). When the regression coefficients from Fig. 7a are used in a transfer function to reconstruct annual ring density, LWpct explains 47 % of the variation over 800 years (Fig. 7b). Although transformed LWpct only explain roughly half of the variance of TRD, this approach should be slightly better than applying a fixed wood density value from the literature because it captures some interannual variation *and* the overall trend with a realistic variability.

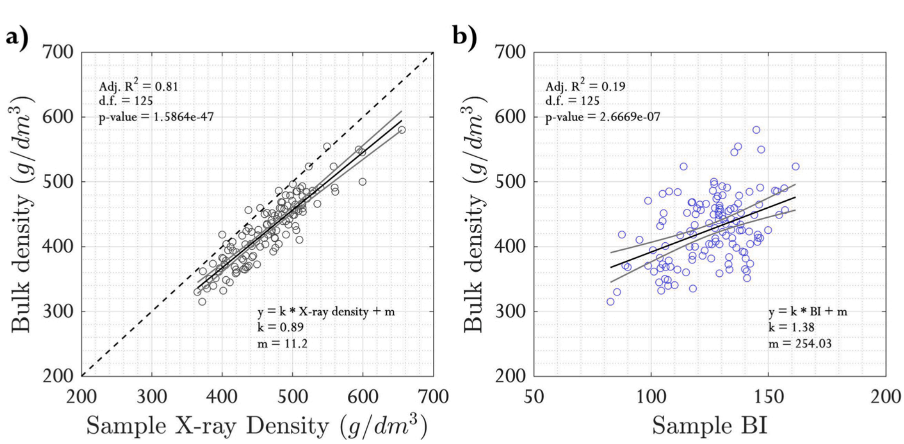

Fig. 3. a) Scatterplots of bulk wood density versus sample X-ray density and b) bulk wood density versus sample BI. Using the regression equations provided in a), it is possible to insert parameter values of X-ray density to obtain bulk wood density corrected annual values of TRD and MXD or whichever density parameter has been derived and is of interest. Theoretically, this could also be done for BI using the regression equation in b), but in this case, the result would not be meaningful because of the high uncertainty in each corrected value and grossly suppressed variance compared to the original bulk wood density.

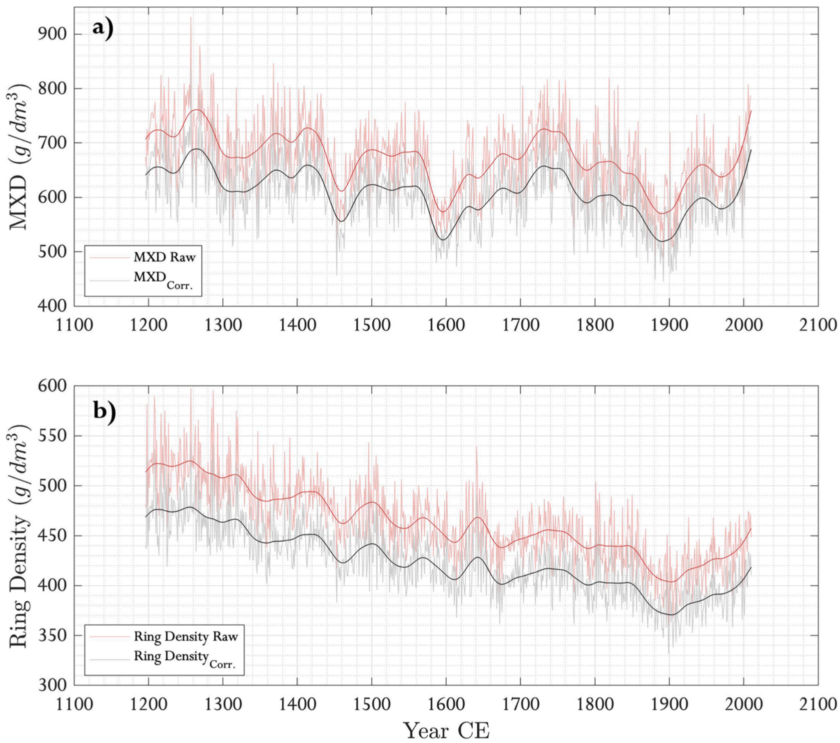

Fig. 4. Correction of a) X-ray MXD and b) ring density using the slope (k) and intercept (m) of the regression in Fig. 3a. The pale red lines represent raw unstandardized chronologies of MXD and ring density, respectively. The distinct red lines represent low-pass filtered, raw, and unstandardized MXD and ring-density chronologies (using cubic smoothing splines with a 50 % frequency response cut-off at 30 years) (Cook and Peters, 1981). The pale gray lines represent unstandardized data corrected with the transfer function y = 0.89 * x + 11.2. The distinct gray lines represent low-pass filtered, unstandardized, and corrected data. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

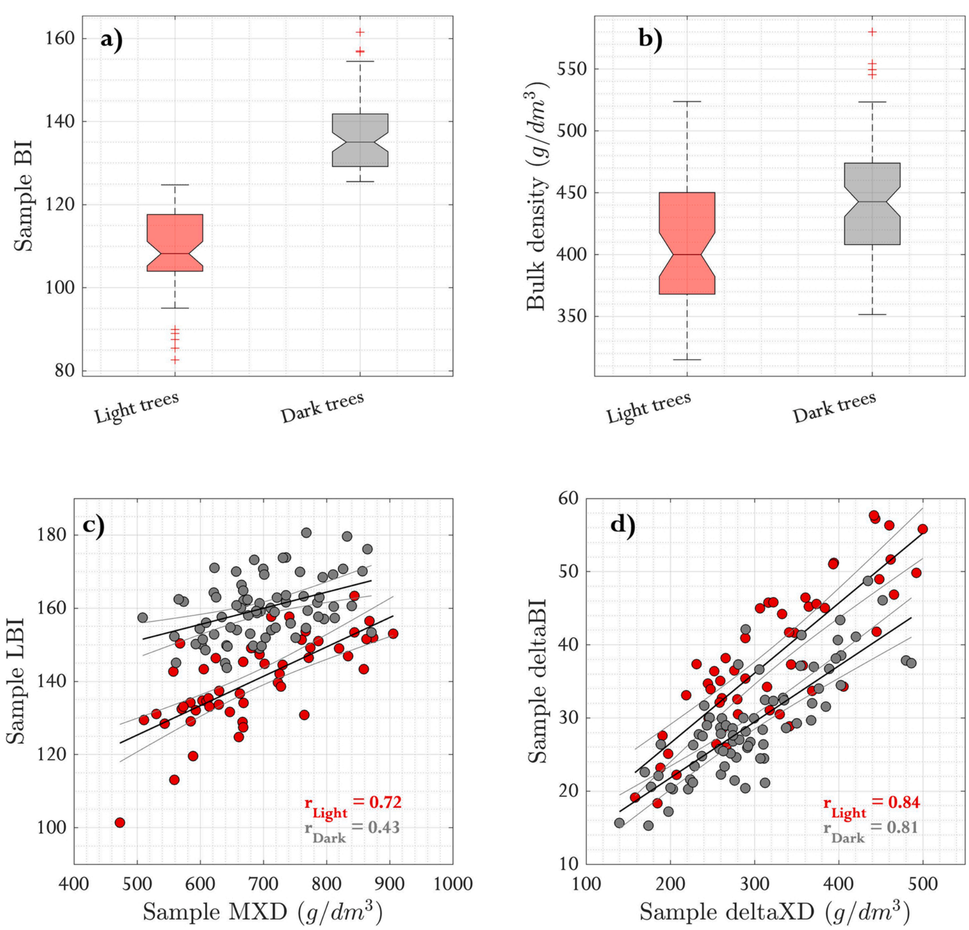

Fig. 5. Comparison of relationships between dark and light wood in terms of a) BI and b) bulk wood density. The difference between dark and light BI is statistically different from the difference between the bulk densities of light and dark wood according to a Student's *t*-test (a = 0.01). Based on the division of light and dark wood, c) displays the relationship between X-ray-based MXD and LBI at the sample level. d) same as in c) but for the deltaBI and deltaXD parameters. All correlations in c) and d) are significant with p-values < 0.001.

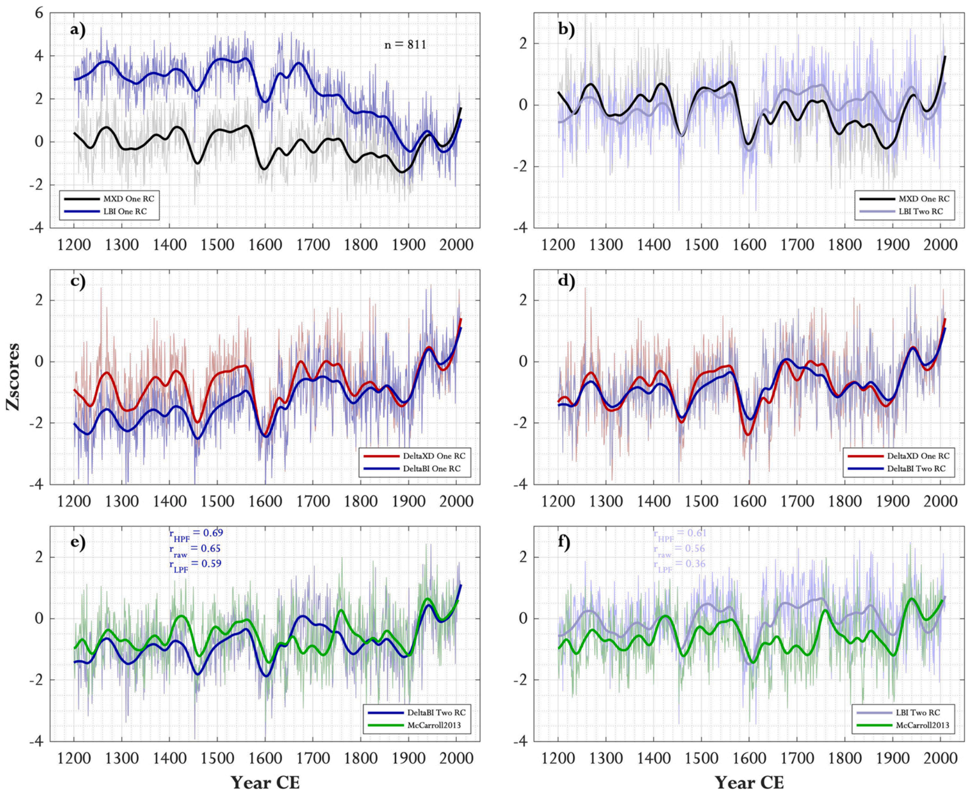

Fig. 6. Comparison of RCS standardized chronologies using various parameters and standardization implementation. a) MXD versus LBI using single RCS standardization (one RC) with low-pass filtered chronologies using cubic smoothing splines with 50 % frequency cut-off at 50 years (Cook and Peters, 1981) superimposed. b) Same as in a) but for LBI using two regional curves (two RC) for light and dark material, respectively. c) Same as in a) but for deltaBI and deltaXD. d) Same as in c) but for deltaBI using two regional curves. e) Double RCS deltaBI versus the McCarroll et al. (2013) multi-proxy reconstruction of summer temperatures. f) Double RCS LBI versus the McCarroll et al. (2013) reconstruction. In e) and f), correlations (r) between the chronologies are also displayed for 1st differenced data (HPF), unfiltered data (raw), and low-pass filtered data (LPF). All zscores were calculated using the means and standard deviations from the period 1900–2010 CE.

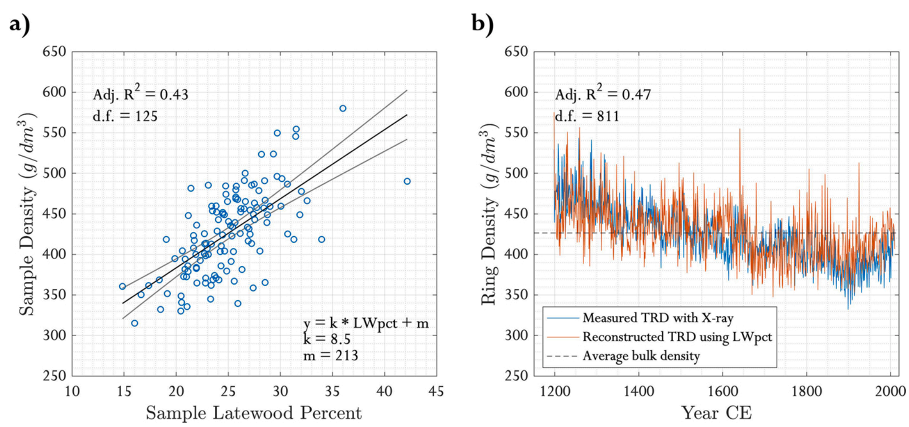

Fig. 7. a) Relationship between LWpct and bulk wood density. Tree-ring density reconstructed from LWpct and measured and corrected TRD using the X-ray technique in b).

4. Discussion

4.1. Reflecting on the bulk wood density protocol

In this study, we have described a simple measurement protocol for conducting bulk wood density measurements that is tailored for tree-ring researchers. Although the samples sometimes had small volumes, the high correlation with X-ray density reveals that the samples were accurately measured regardless of volume. However, at this stage, we do not recommend using samples of less than 0.2 cm^3, largely because we did not test samples this small. Recall that bias theoretically increases with decreasing sample size. For a chronology with approx. 100 samples, it takes roughly one workday to conduct bulk wood density measurements when a protocol is established, all equipment is available, and the samples are suitable. The suitability of the samples depends on the presence of degradation, an even tangential representation of all rings, and reasonably smooth surfaces. It is possible to use increment cores, laths, or even blocks from dead wood as long as there is no glue or residue from other analyses or treatments. Even compared to rapid BI measurements, bulk wood density measurements using this protocol require a marginal time investment. Considering the time spent inventorying samples for analyses and gluing them to wooden supports, it would be more convenient to conduct the bulk wood density measurements as a planned step prior to or in conjunction with BI or X-ray analyses. Bulk wood density measurements can also be conducted years later, as this study can confirm. However, considering the relevance of bulk wood density measurements, particularly for verifying X-ray density and BI measurements, we highly recommend conducting them in parallel to other analyses. In doing so, bulk wood density measurements can be used as a diagnostic tool and possibly for mean value corrections.

4.2. Relevance and applications of bulk wood density for X-ray densitometry

We established, with a high degree of confidence, that the X-ray-based density measurements of Björklund et al. (2014) had a mean value offset of approx. + 10 %. If the MXD data is updated or complemented by measurements done at a later time or with a different device, it is thus important to *not* pool the data prior to standardization procedures aimed at preserving the long-term variance, such as RCS (also cautioned in Helama et al., 2012; Melvin et al., 2013; Zhang et al., 2015). This is because the mean value offset can change with time or with a different device (e.g., Klesse et al., 2015; Björklund et al., 2019). However, even if differently analyzed MXD data from the same site are corrected with bulk wood density, it is not advisable to pool because variation in device-specific measurement resolutions can add bias that affects sub-annual parameters such as MXD and minimum density (Polge, 1978; Lenz et al., 1976; Björklund et al., 2019). The point of using bulk wood density in parallel to producing MXD is not to correct data for pooling with data produced with different devices, but to be able to pool data produced with the same device, or similar devices with the same resolution, at different time periods. This is because it may be preferable to pool new updates with older chronologies in order to produce robust RCS chronologies. Fitting separate regional curves to the presumably short and modern updates can lead to a loss of the climate signal in the update, which should be avoided if possible (Briffa and Melvin, 2011).

Moreover, if ring density is used to scale annual volume estimates into biomass, important biases will arise when density is not accurately measured. If a 10 % error in density is applied, this translates to a 10 % error in aboveground biomass because allometric equations usually scale volume estimates with density measurements (Zianis et al., 2005). Fortunately, ring density is far less susceptible to the resolution bias (Jacquin et al., 2017) and adequate values from different devices can most likely be obtained when corrected with bulk wood density (Zeller et al., 2017). The correction of ring density for biomass estimations thus appears to be more essential than correcting MXD data for dendroclimatological purposes. This balance is also reflected in the literature where, to our knowledge, only two studies have used bulk wood density in a dendroclimatological context (Björklund et al., 2019; Lenz et al., 1976). However, it is frequently utilized in non-dendroclimatic literature (e.g., De Ridder et al., 2011; Evans, 1994; Zeller et al., 2017). Finally, the use of bulk wood density in studies using micro-densitometry would be useful not only for individual studies but also for fostering a deeper understanding of species and X-ray-device specific discrepancies between bulk wood density and X-ray-based measurements across a wide range of wood types if used as part of a standard protocol.

4.3. Novel applications of bulk wood density for BI densitometry

In this sample material, the BI measurements had limited potential for direct correction using bulk wood density. However, bulk wood density clearly revealed the weakness of BI in terms of biased cell wall color. Using bulk wood density as a diagnostic tool to detect bias in BI is thus important to prevent the uncritical use of LBI in dendroclimatic studies and annual ring BI in potential studies of biomass. Bulk wood density could also be useful for establishing whether *other* sample materials are less biased and therefore directly comparable to X-ray products.

If color corrections, such as deltaBI, are used in dendroclimatic studies, bulk wood density can be used to indicate how to separate material to create less biased RCS chronologies. Defining cohorts of samples with starkly different colors but similar densities would improve the deltaBI protocol because the deltaBI is artificially suppressed in heavily discolored samples compared to less discolored samples (Björklund et al., 2015). If a “bulk-density-separated multiple-RCS deltaBI-chronology” differs from its “single-RCS deltaBI-chronology” counterpart in long-term trends, this should be taken as a strong indication that the deltaBI protocol is not sufficient on its own. If the trends are similar, however, this could be interpreted as a mark of quality. As BI is becoming more popular, deltaBI, bulk wood density, and the like become increasingly important diagnostic tools to determine the reliability of BI-based datasets.

4.4. Reconstructing ring density with bulk wood density and latewood percent

In the wood industry, the relative amount of latewood, here LWpct, is often taken as a proxy for wood density, which is well correlated with the quality of timber, such as impact strength, shear strength, bending strength, tensile strength, hardness, etc. (e.g., Barnett and Jeronimidis, 2003). To our knowledge, LWpct has never been explicitly tested or used as a predictor for annual ring density (but see Pretzsch et al., 2018). In our study, LWpct captured some inter-annual variation and the overall trend of TRD, as well as the correct mean value. Mean values are essential, but trends in density may also bias estimations of aboveground biomass (e.g., Pretzsch et al., 2018). Reconstructed annual ring density using LWpct is at least arguably better than using a fixed literature value for density in aboveground biomass estimations. In studies of annualized biomass, ring-width measurements are usually transformed into volume using various allometric equations (e.g., Zianis et al., 2005). The additional time expenditure required to measure earlywood and latewood widths and bulk wood density is minimal; it therefore makes sense to replace fixed literature values of density with these measurements. Moreover, the speed, affordability, and simplicity of measurements of earlywood and latewood widths are far superior to the more precise X-ray technique. If large amounts of sample material are to be analyzed, a literature value or LWpct-based ring density are the most practical options for representing density, as X-ray density measurements are simply too time-consuming and expensive.

4.5. Concluding remarks

We have confirmed that potential mean level offsets are prevalent in X-ray densitometry and emphasize that the diagnosis and correction of X-ray density is easily done using bulk wood density measurements. Although BI is less directly linked to wood density than X-ray densitometry, it would in theory be possible to use similar approaches for BI as for X-ray densitometry. However, bulk wood density can also be used to objectively advise against a transformation into wood density, which is also valuable. Even if bulk wood density is used to disqualify direct transformations of BI into density, it can still be used to diagnose and improve the use of deltaBI with regard to its overall trends and multi-centennial variability. Bulk wood density can also be used as a time- and cost-effective metric to help reconstruct annual ring density from latewood percent. Although our reconstruction only explains about half of the variation in ring density, it is most likely superior to using fixed literature values of density in allometric equations aimed at biomass estimations.

By demonstrating the simplicity, relevance, and applicability of bulk wood density, we hope to have raised new awareness of this versatile and important metric. We encourage tree-ring researchers to use it as part of standard protocols, as well as to continue to experiment with other relevant tree-ring applications.

Declaration of Competing Interest

The authors report no declarations of interest.

Acknowledgements

We acknowledge funding from the Swiss National Science Foundation (Project XELLCLIM no. 200021_182398) awarded to G.v.A. We thank Björn Gunnarson for help with the X-ray densitometric measurements back in 2012. We also thank Hans Linderholm, Peng Zhang, and Christian Axelsson for help with field missions during 2010-2011.

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