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ORIGINAL ARTICLE

Blue intensity for dendroclimatology: Should we have the blues? Experiments from Scotland

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ABSTRACT

Blue intensity (BI) has the potential to provide information on past summer temperatures of a similar quality to maximum latewood density (MXD), but at a substantially reduced cost. This paper provides a methodological guide to the generation of BI data using a new and affordable BI measurement system; CooRecorder. Focussing on four sites in the Scottish Highlands from a wider network of 42 sites developed for the Scottish Pine Project, BI and MXD data from Scots pine (*Pinus sylvestris* L.) were used to facilitate a direct comparison between these parameters. A series of experiments aimed at identifying and addressing the limitations of BI suggest that while some potential limitations exist, these can be minimised by adhering to appropriate BI generation protocols. The comparison of BI data produced using different resin-extraction methods (acetone vs. ethanol) and measurement systems (CooRecorder vs. WinDendro) indicates that comparable results can be achieved. Using samples from the same trees, a comparison of both BI and MXD with instrumental climate data revealed that overall, BI performs as well as, if not better than, MXD in reconstructing past summer temperatures (BI r^2 = 0.38–0.46; MXD r^2 = 0.34–0.35). Although reconstructions developed using BI and MXD data appeared equally robust, BI chronologies were more sensitive to the choice of detrending method due to differences in the relative trends of non-detrended raw BI and MXD data. This observation suggests that the heartwood–sapwood colour difference is not entirely removed using either acetone or ethanol chemical treatment, which may ultimately pose a potential limitation for extracting centennial and longer timescale information when using BI data from tree species that exhibit a distinct heartwood–sapwood colour difference. Additional research is required in order to develop new methods to overcome this potential limitation. However, the ease with which BI data can be produced should help justify and recognise the role of this parameter as a potential alternative to MXD, particularly when MXD generation may be impractical or unfeasible for financial or other reasons.

Introduction

Within the context of late-Holocene palaeoclimatic research, tree rings are among the most important proxy archives, not least for their ability to preserve annually resolved climate information (Jones et al., 2009). It is possible to utilise the measurements of multiple tree-ring parameters, each with their own specific strengths and limitations. As well as more conventional parameters such as ring-width (RW), maximum latewood density (MXD) and stable isotopes, minimum blue reflectance or blue intensity (BI) is emerging as a potentially important new parameter relevant to dendroclimatological research. Traditionally, with respect to their ability to capture past climate, RW has been seen as superior to maximum latewood density (MXD) in the low frequency domain, while MXD was considered to better represent inter-annual variability (D'Arrigo et al., 2014). More recently, this view has been challenged (Luckman and Wilson, 2005; Büntgen et al., 2006; Esper et al., 2012). However, the costs associated with

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MXD data generation are relatively high both from a financial viewpoint (including equipment, software and maintenance costs) and in terms of the time and effort required compared to the measurement of RW. Therefore, only a few tree-ring laboratories around the world have the ability to measure MXD from conifer tree-ring samples. This has undoubtedly been the primary reason for the limited development and utilisation of MXD chronologies compared to RW. The prospect of a more affordable alternative to MXD would have positive consequences for dendroclimatological research and the field of dendrochronology more generally.

The relatively strong absorption of ultraviolet (UV) light by lignin (Fukazawa, 1992) has been utilised in several previous studies. Lange (1954) was one of the first studies to utilise this property and measured the cell wall lignin content of spruce with UV photomicrographs. More recently, Gindl et al. (2000) explored the sensitivity of latewood lignin content in Norway spruce to temperature. By using UV microscopy they identified a significant positive relationship between the lignin content of terminal latewood tracheids and late summer/early autumn temperatures.

Early attempts to utilise digital images of tree rings to identify and analyse anatomical features and to recognise the potential for using reflected *visible* light as a alternative to X-ray densitometry, were undertaken by Yanosky and Robinove (1986) and Yanosky et al. (1987). These studies were also among the first to recognise the potential of using reflectance as a substitute for MXD. A different approach attempted to analyse light passed through thin wood microsections (Park and Telewski, 1993), although this method required thin-sectioning of the samples with a microtome.

Sheppard et al. (1996) utilised reflected light from the surface of latewood for dendroclimatic purposes. Their work reconstructed April–May temperatures back to the beginning of the 19th century using MXD and reflected light measurements with density and brightness explaining 30.8% and 29% of variance, respectively, during the 1899–1976 period. The work was the first and so far only study to derive a temperature reconstruction using only reflected light data, although a BI chronology was included in a 1200 year multiparameter composite reconstruction from northern Fennoscandia by McCarroll et al. (2013).

Using scanned images, McCarroll et al. (2002) examined the agreement of maximum, mean and minimum reflectance spectra of visible light (blue, green and red wavelengths), infrared and UV with MXD from the latewood of Scots pine samples from Finland. They observed that the strongest correlation with MXD was with minimum blue reflectance ($r = -0.976$ with raw non-detrended data for the period 1960–1995). This result was attributed to the high sensitivity of absorbed blue light to lignin.

Campbell et al. (2007) further emphasised the relevance of BI as a palaeoclimatic proxy parameter by comparing MXD and BI data of Scots pine (measured with WinDENDRO^{TM} (hereafter WinDendro)) from northern Finland to several climate variables including temperature. The study reported a similar response of both parameters to climate and a stronger correlation between BI and mean June–August temperature from two weather stations compared to MXD ($r = -0.80$ and -0.65 for BI; $r = 0.59$ and 0.58 for MXD).

Additionally, Sheppard and Wiedenhoeft (2007) discussed the potential of measuring latewood reflectance of visible light using samples with a noticeable heartwood–sapwood (HW–SW) colour difference using a peroxide and ethanol treatment. They were then able to model and reconstruct summer (May–September) precipitation using latewood width and reflected light with an r^2 of 0.25. An attempt to provide guidelines and basic protocols for sample preparation of 'optimal' samples (i.e. that did not contain tight-ringed segments or distinctly coloured HW–SW sections), was presented in the study by Babst et al. (2009). Their results suggest that BI data quality may decline if samples are sanded with less than 400 grit sandpaper, scanned using a resolution lower than 1200 dpi and if the colour saturation level of the digital image is reduced below 100%.

Although some studies (e.g. Yanosky and Robinove, 1986; Sheppard et al., 1996; Sheppard and Wiedenhoeft, 2007) approached the issue of image capture by using a camera system coupled with a microscope, most were restricted to using a single magnification setting to avoid brightness variations due to changes in magnification, although Sheppard and Singavarapu (2006) were recently able to compensate for this effect. Other researchers (e.g. McCarroll et al., 2002; Campbell et al., 2007, 2011; Babst et al., 2009; Tene et al., 2011; Wilson et al., 2012) adopted a different approach using a flatbed scanner to capture sample images which entirely bypasses the magnification issue.

Despite BI showing great promise as a cheaper alternative to MXD, only very limited attempts have been made to reconstruct past summer temperatures using this parameter. However, even without the need for an X-ray densitometer, publicly available software still represents a significant investment for laboratories wishing to commence research using this parameter. The most widely applied method for generating BI data from tree cores currently uses the WinDendro software package (WinDendro, 2013). Although this approach produces reliable results for dendroclimatic purposes (Campbell et al., 2007, 2011), financially, it may be beyond the reach of many researchers who would consider undertaking research with the BI parameter or who wish to use it for teaching and training purposes. If a cheaper software package were to be available then this would not only help broaden access to this powerful new proxy, but also provide a potential teaching resource for student projects and field courses.

Over recent years, a low cost alternative for measuring BI has been incorporated into the CooRecorder/CDendro software package (Larsson, 2013). This software is more accessible to a wider range of individuals and organisations seeking to undertake BI analysis. Building on preliminary work by Wilson et al. (2012), we present here the methodological steps for sample preparation and BI data generation using the CooRecorder/CDendro software package.

The paper is divided into three sections; the first describes sample preparation and BI data generation using CooRecorder, the second presents data produced using alternative measurement software and sample preparation techniques including both WinDendro and the MXD parameter. Finally, the ability of each method to capture climatic information is explored through comparison of the BI and MXD parameters with monthly temperature data to assess their quality as summer temperature proxies through the development of well-replicated living tree chronologies of Scots pine (*P. sylvestris* L.) from the Scottish Highlands.

Through this process, we also identify and explore several potential biases which, if not considered and addressed, could limit the ability of the data to represent climatic information robustly. In a companion paper, the techniques described here are employed to develop a regional climate reconstruction using samples of Engelmann Spruce from British Columbia (Wilson et al., in review).

Methodology

Measurement of RW, MXD and BI was carried out on samples of Scots pine from selected sites around the Scottish Highlands (Fig. 1) for the purpose of assessing the BI parameter. Although BI has been measured from a total of 17 sites, in order to focus on methodological aspects, we focus on data from only four pine woodlands (including Ballochbuie and Ryvoan for which MXD chronologies were also developed, Loch an Eilein, where three cores per tree

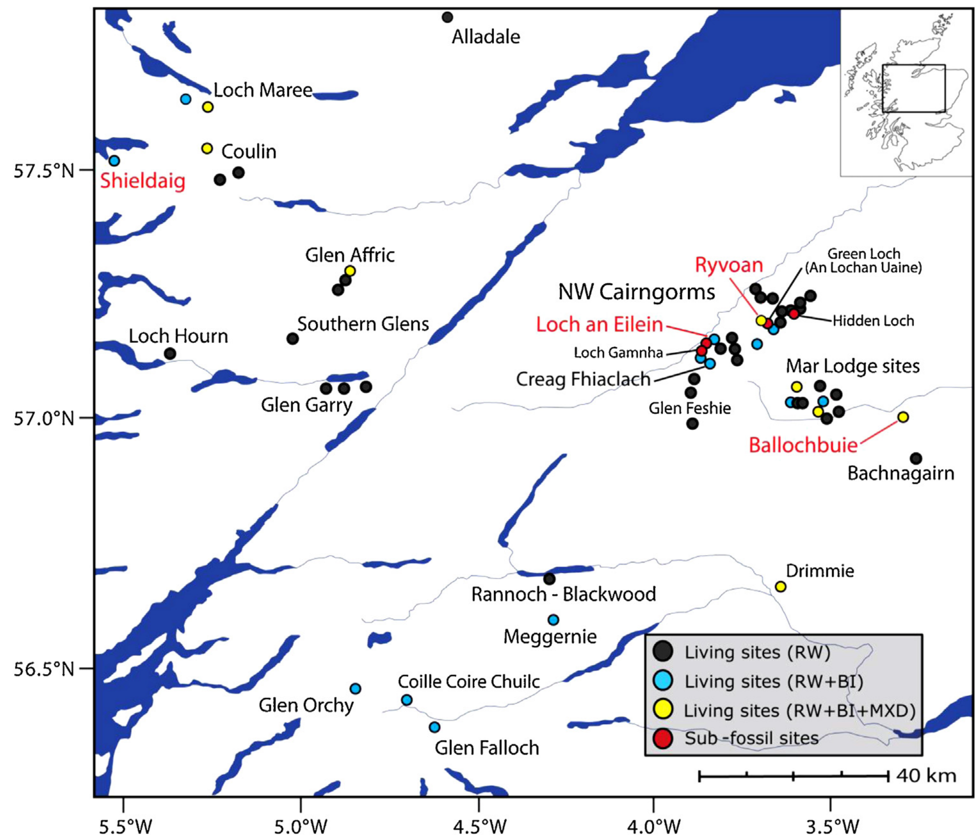

Fig. 1. Scottish Pine Project network. Site names highlighted in red are used in this study. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of the article.)

were collected in order to perform signal strength tests, and Shieldaig, which was selected for experimentation due to the relative shortness of samples from this site) using a variety of experiments to assess the BI parameter. We firstly detail the current methods employed by the St Andrews Tree-Ring laboratory developed over several years of experimentation. We then also discuss methodological aspects of the approach which likely still need refinement.

St Andrews preparation and measurement method

Here we provide the methodological steps required to generate BI data employing a resin extraction technique using acetone and the CooRecorder measurement system (hereafter referred to as the 'St Andrews' method). 5 mm tree core samples were taken using a Swedish increment corer, stored in slitted plastic straws and allowed to air dry. If left moist for an extended period of time, the growth of fungi and mould could result in discolouration of the sample with the potential risk of degrading the quality of extractable BI data as it may change the reflectance properties of the sample.

Prior to acetone treatment, the sample code of each core was noted and the approximate length of each sample measured using a ruler to permit identification of each core following acetone treatment. Contrary to MXD preparation methods, whole cores were used as it was not necessary to cut samples into thin laths. Samples were attached to long, thin stainless steel rods (~5–6 cores per rod) by placing them circumferentially around the rod and tying them with string every ~5 cm along the length of the cores. In this way, deformation and warping of the cores as they expanded in acetone was largely prevented. The metal rods were then placed inside tall 1L glass cylinders permitting them to be entirely submerged in acetone (≥99.5%, Fisher Scientific UK) at room temperature. The containers were then sealed with rubber plugs to prevent evaporation of acetone. Metal wire or string was attached to the rods and placed partially outside the containers, ensuring easy removal after acetone treatment.

Early experiments by Frith (2009) using 5 mm Scots pine cores found that a 72 h acetone treatment period was sufficient to remove extractives, although that result was achieved using only partial immersion. However, a longer period of 1 week was initially chosen here to ensure that all extractives were removed from fully immersed 5 mm samples. Experiments detailed later in this paper determine that a shorter period of acetone immersion is adequate for resin extraction and that prolonged immersion can cause weakening and gradual disintegration of the wood structure.

As core samples were used, as is the norm for measuring ring-width, it was important to maintain correct orientation of the core during mounting (i.e. vertical alignment of trachea cells). After mounting, samples were sanded smooth using a fixed rotating disc sander up to 1200 grit, which resulted in a smoother surface than the 400 grit considered adequate for BI analysis by Babst et al. (2009). While sanding, samples were periodically checked to ensure 'flatness' of the sanded surface, particularly while using lower grit sandpaper, as an uneven surface would result in sections of scanned images being out of focus.

A Canon CanoScan 9000F flatbed scanner in conjunction with Silverfast (version 8.0.1.24) was used to scan cores for measurement and image analysis. Although a scanning resolution of 1200 dpi was suggested as adequate by Babst et al. (2009), a resolution of 2400 dpi facilitated working with cores containing thinner rings. Images were saved in the JPEG format, an image format compatible with CooRecorder. An even higher resolution

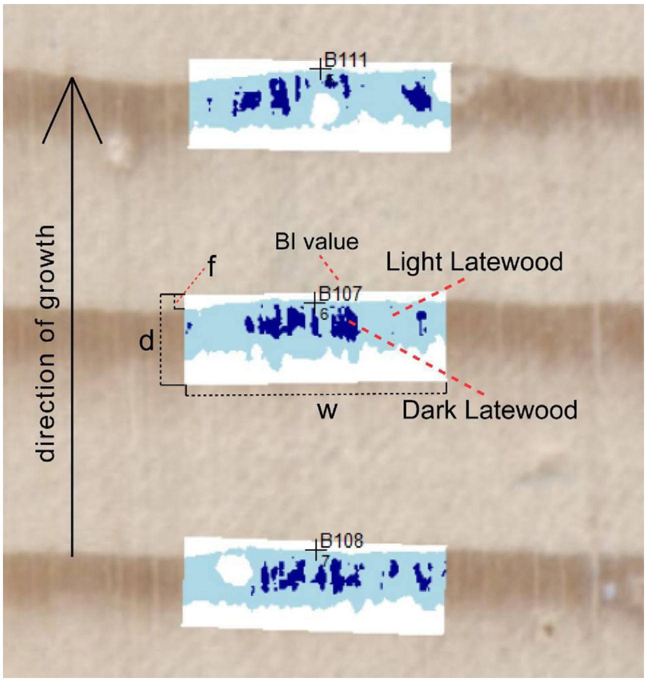

Fig. 2. CooRecorder BI generation window with adjustable parameters *w*: width, *f*: offset, *d*: depth, and % dark latewood (parameters *w*, *f* and *d* are measured in number of pixels).

(3200–4800 dpi) may be beneficial for scanning cores containing particularly thin-ringed sections (i.e. <0.05 mm). However, as Babst et al. (2009) caution, the scanning resolution should not exceed the hardware resolution of the scanning device to avoid affecting BI by the creation of artefacts from the interpolation algorithm. Furthermore, doubling the scanning resolution increases scanning time and results in a quadruple increase in storage requirements, which may be undesirable. Scanning a large area at a very high resolution may also lead to memory or software limitations during the scanning process, in which case either a lower resolution should be used or the sample can be scanned in sections, producing multiple images that can then be spliced together.

Calibration of the scanner was necessary in order to ensure that any data generated are comparable between different scanners and research facilities, and to prevent drift of BI measurements as output from the scanner light may degrade over time (Campbell et al., 2011). An IT8 Calibration Target (IT8.7/2) printed on Kodak Professional Endura paper was used to calibrate the scanner by using the SilverFast Auto IT8 Calibration procedure integrated in the SilverFast scanning software. This procedure was repeated for each scanning session. In order to minimise any influence of ambient light which may change with time and place, isolation of the scanning surface from its surroundings was carried out by placing a box, fitted to the shape of the scanner, over the scanning area. Calibration and all subsequent scanning were performed with this apparatus.

Samples were cross-dated using standard dendrochronological methods (Stokes and Smiley, 1968). RW and BI measurements were made using the programme CooRecorder which uses point placement to calculate RW by calculating the distance between successive (*X*,*Y*) coordinates. The same points are then used to generate BI data. The programme places a window around each point according to adjustable parameters that include width, offset, depth and % of latewood recognised as 'dark' (Fig. 2). The mean BI value of the 'dark' latewood is then calculated and used as the BI value for a particular ring. For more information on the basic functionality of CooRecorder and a detailed step-by-step guide for the generation of BI data refer to the Cybis website (Cybis, 2013). Individual BI and RW data series were then transformed using CDendro to the standard Tucson decadal format. Cross-dating of the time-series (using both RW and BI) was verified using COFECHA (Grissino-Mayer, 2001) and CDendro.

BI is inversely correlated with MXD (i.e. dense latewood will express low reflectance). Therefore, in order to facilitate a direct comparison between BI and MXD whilst also allowing the same detrending functions to be used for each parameter, all raw BI series were inverted prior to detrending according to Eq. (1):

$$x_{i(adj)} = 2.56 - \frac{x_i}{100} \quad (1)$$

where x_i is the raw BI value in year *i*. The constant 2.56 is used in order to prevent $x_{i(adj)}$ from becoming ≤ 0, considering all possible values of x_i $\{0...255\}$. This inversion step is implemented as an option for the output of BI data in CooRecorder.

Detrending the age related growth trend was undertaken using the programme ARSTAN (Cook and Holmes, 1986). Although using single series data adaptive approaches, it is common to use a linear (or negative exponential) curve to detrend MXD data (e.g. Davi et al., 2003; Wilson and Luckman, 2003), it has been assumed that BI data should also be detrended in a similar way (e.g. McCarroll et al., 2002; Campbell et al., 2007, 2011). However, in many BI series (following inversion according to Eq. (1)) an initial increase in BI is apparent in the juvenile period, followed by a steady decrease thereafter. For this reason, fitting a Hugershoff curve (Bräker, 1981; Cook et al., 1990a) may be more appropriate due to its extra flexibility to fit the early period trends. Therefore all BI data herein were detrended using this approach. This method was used extensively by Briffa et al. (1992a) for their MXD based western Northern American spatial temperature reconstruction. For this reason, all MXD data were also detrended using a Hugershoff curve in order to facilitate direct comparison between the two parameters while ensuring that any differences were not subject to the choice of alternative detrending methods.

Due to the homoscedastic nature of the data, it was possible to detrend the series by calculating BI and MXD indices as residuals of the raw data from the fitted curve (Schweingruber, 1988; Cook et al., 1990a) and chronologies were calculated using a robust (bi-weight) mean to limit the influence of outliers (Cook et al., 1990b). For all series, the BI measurement of the most recent ring was excluded from analysis as it consistently exhibits an anomalously low BI signature due to an edge effect likely related to the latewood-bark boundary.

The above description details the basic methodology used to measure and process the BI data. However, in order to assess potential problems or biases associated with BI generation, a series of experiments were performed which are detailed below.

BI measurement experiments

Window parameter settings

To evaluate the influence of changing the window parameters in CooRecorder (Fig. 2) on the overall BI chronology from Ballochbuie, the chronology was generated multiple times with different parameter settings. As well as assessing the influence of modifying all parameters, one reason in particular for undertaking this experiment was to determine whether the size of the window affects the quality of the data, as a larger window should theoretically produce a more representative mean BI estimate for a ring than a smaller one.

Acetone immersion experiments

As with any method intended to remove extractives using a chemical solvent, it should be possible to determine an immersion time which produces optimal results with tree cores. Although preliminary experiments were undertaken by Frith (2009), these results were never published and as far as we are aware no other group has published such experiments for acetone. We therefore show experimental results using samples from the Shieldaig site (Fig. 1) used to assess the improvement in data quality and to identify the duration of a sufficient time period for acetone extraction by comparing BI results from the samples left in acetone for 0–144 h in 48 h increments.

Ambient and reflected light

As scanners are essentially designed to capture an image of a flat object such as a picture, as soon as a raised sample (or disc) is scanned, there is potential for ambient light from the surrounding room to influence the scanned image. To minimise this effect, a box was constructed to surround the whole scanner (without lid) to stop any ambient light contamination. Initially, as all scanners have a white inner surface on the lid, we used a box with a white, smooth inner surface. However, when scanning the same sample using different orientations, we noted that the BI results differed slightly which initially could not be explained. We hypothesised that light from the light source is reflected within the box, inducing a potential bias. To test whether the use of boxes with different inner surface properties (colour and texture) affected BI results, an image of the entire empty (no sample) scanning area was obtained with the scanner covered by a box with the smooth, white inner surface. BI measurements were then carried out along several tracks, representing a range of potential sample placement and orientation choices on the scanning surface. This process was then repeated using a box with a black, non-reflective inner surface.

Parameter comparison

EPS and replication

Data from cores collected from the Ballochbuie and Loch an Eilein sites were examined to identify whether any differences in expressed signal strength (Briffa and Jones, 1990) existed between BI and MXD and to explore any potential improvements in the common signal that could be gained by averaging data from multiple cores per tree.

BI vs. MXD

To facilitate a comparison of BI and MXD, a set of 21 tree core pairs ('A' + 'B' cores from the same tree) were sampled from Ballochbuie and 17 pairs at Ryvoan (Fig. 1). To generate BI data, the first subset of 5 mm diameter cores ('A' cores) was processed using the St Andrews method described above. MXD was measured from the second ('B' core) subset of 10 mm diameter cores from both sites at the Bolin Centre for Climate Research at Stockholm University (hereafter referred to as 'Stockholm' samples). The MXD data were produced using an ITRAX multiscanner from Cox Analytical Systems (Cox, 2013). The samples were prepared according to standard dendrochronological techniques (Schweingruber et al., 1978). Thin laths were cut from the samples using a twin-bladed circular saw (1.20 mm thickness) and treated with ethanol in a Soxhlet apparatus for 24 h in order to extract resins and other soluble compounds. The laths were X-rayed (with 12% water content (air dry)) in the ITRAX equipped with a chrome tube tuned to 30 kV and 50 mA, with the steptime at 75 mS. The multiscanner produces a 16 bit, inverted grey scale digital image at 1270 dpi resolution. Calibration of the grey scale was done with a calibration wedge of cellulose acetate with a density of 1.274 g/cm^3 from Walesh Electronic (Schweingruber, 1988). The radiographic images were evaluated using WinDendro, to provide ring width and density data (Guay et al., 1992).

In addition to comparing the BI and MXD to each other, the relationship between the two parameters and climate was assessed using the Scottish dataset of mean monthly surface temperature (hereafter referred to as 'MST') developed by Jones and Lister (2004). As this dataset ends in 2004, 0.5° gridded mean monthly temperature CRU TS3.10 data (hereafter 'CRU' – Harris et al., 2014) were used to extend the MST dataset to 2009 by scaling the post-2004 period CRU temperature data from the region covered by MST over the common period of overlap of the two datasets. For the purposes of this study, the earliest date of the MST dataset used is 1866 due to potential homogeneity issues prior to this time discussed by Jones and Lister (2004). The combined MST and CRU temperature data are hereafter referred to as 'Extended-MST' (EMST).

Ethanol vs. acetone

To evaluate whether BI results are influenced by the choice of chemical treatment, off-cuts from a subset of nine of the ethanol treated 'Stockholm' samples were scanned and measured for BI with CooRecorder. Separate chronologies were developed using these nine ethanol-treated 'B' cores and the nine acetone-treated 'A' cores and then compared using correlation response function analysis (CRFA) to the EMST dataset.

CooRecorder vs. WinDendro

To assess the performance of two alternative measurement systems, an independent group of nine samples from Ballochbuie were prepared at Swansea University. Samples were cut into thin laths of 2 mm thickness using a twin-bladed saw and extractives removed using ethanol (≥99.5%, Fisher Scientific UK) in a Soxhlet apparatus for 48 h followed by repeated washing in boiling deionised water and air drying, prior to sanding and scanning. These 10 mm diameter samples were measured with WinDendro and subsequently using CooRecorder at St Andrews. Both resultant BI chronologies produced from these measurement systems were compared with each other and to the EMST data with CRFA.

Results and discussion

BI measurement experiments

Window parameter settings

In order to assess the sensitivity of BI data to the selection of BI window parameters in CooRecorder (Fig. 2), the Ballochbuie chronology was generated multiple times by systematically modifying each of the adjustable parameters in turn, producing a range of 'raw' BI values for each year (Fig. 3). The parameter selection together with the inter-series correlation (RBAR) of the detrended version of each chronology is provided in Table 1. The results indicate that the raw BI data remain relatively constrained regardless of the choice of BI window parameters, although a wider range becomes apparent as replication decreases in the early portion of the chronology.

The results from this test suggest using a large (yet not excessively large) window and lower % dark latewood for optimum results. Although optimal settings may be species, site and sample specific, for all following analyses (including Wilson et al., in review) we utilise 160–5–50–15 as the optimal window parameters yielding the strongest common signal (Table 1).

While the variability between the different versions remains fairly constrained regardless of parameter choice (Fig. 3), the size and placement of the window which measures blue reflectance in CooRecorder can have a significant influence on the quality

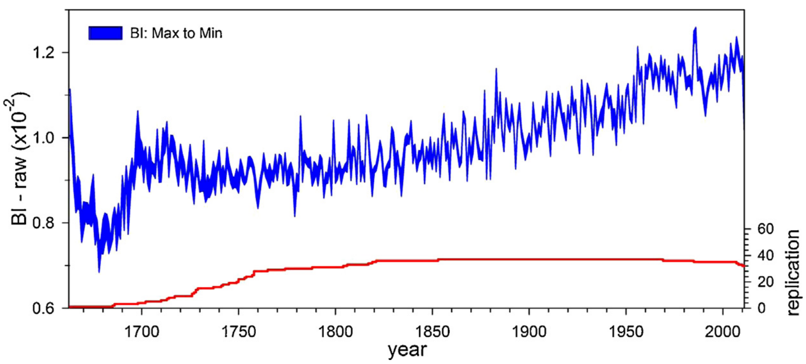

Fig. 3. Range of Ballochbuie BI chronology variants using various CooRecorder frame parameter choices.

(in terms of inter-series agreement – RBAR) of data produced. Choosing a larger window size allows the derivation of the most representative BI value for the latewood of each ring by integrating information from as large an area as possible, minimising the influence of anomalous features (e.g. resin ducts). However, the window width should not be so large as to exceed the sample boundary or integrate data from previous or subsequent rings (for example when ring boundaries are substantially curved such as near the pith). This is also related to the orientation of the window since the placement of the succeeding point in CooRecorder determines the orientation of a given window (i.e. the wider the window, the greater the chance that they become rotated – possibly leading to integration of areas from other rings than the one for which a value is being derived).

Likewise, although increasing the depth parameter will also benefit BI results, beyond a certain point, only earlywood will be added as the parameter increases and so will therefore not affect the BI estimate for that ring. Similarly, the depth of a window will be limited if a window's edge touches the boundary of the previous year's window, thus preventing overlap. A small offset parameter (parameter *f* in Fig. 2) set to ∼5 pixels can in some cases be useful for including latewood sections at ring boundaries that have a non-uniform shape. Ultimately however, although the "optimal" parameter selection could theoretically vary from sample to sample, the BI measurements on the whole appear generally insensitive to variations in the window parameters (Fig. 3). A simple rule of thumb is that a larger box provides a more robust mean estimate of BI.

Table 1
Mean inter-series correlation (RBAR) of Hugershoff detrended Ballochbuie chronologies developed with a range of BI window parameters. (The optimal parameter option for the particular image resolution utilised in this study is highlighted.).

Width–offset–depth–%blue	RBAR
5–5–10–15	0.155
10–5–10–15	0.165
20–5–10–15	0.176
40–5–10–15	0.188
60–5–10–15	0.190
80–5–10–15	0.191
160–5–10–15	0.196
40–0–10–15	0.185
40–5–15–15	0.200
40–5–30–15	0.212
40–5–50–15	0.221
40–5–10–5	0.188
40–5–10–25	0.186
40–5–10–50	0.183
40–5–10–75	0.176
160–5–50–15	**0.224**
240–5–50–15	0.211

In general, it is important to avoid anomalously dark areas or artefacts (e.g. cracks) which may radically depress the measured BI value. If not avoided, such anomalous values may primarily affect the correlation of a sample with a master chronology. Likewise, if missing rings in individual series are represented by 'zero values', this may adversely affect correlations because while low RW or MXD values represent a poor growing season, low BI values correspond to very productive years (or high RW and MXD). Thus, setting a BI value to zero to represent no growth would run contrary to a high BI value that would be expected for such a 'ring'. Unlike with RW for example, values of BI should never approach zero (due to the inverted nature of the parameter relative to RW). Although some software such as CDendro exclude missing rings from affecting analysis (e.g. correlation calculation), others may not. This ceases to be an issue once the data are inverted before any other data manipulation or analysis takes place. We therefore advise that the raw BI data should be inverted according to Eq. (1) to allow for the retention of missing ring information, but also to allow similar detrending methods to be employed as used for MXD.

Acetone immersion experiments

In order to evaluate the effect of varying periods of time used to treat the Scots pine samples with acetone, a chronology was developed with samples treated from 0 to 144 h in 48 h increments (Fig. 4). As the HW–SW transition occurs around the same point in the majority of core samples, it is possible to discern relative differences in the two sections of the chronology. BI changes are most noticeable in the heartwood portion where reflectance increases following treatment, with relatively little change occurring in the sapwood. That the greatest change is observed in the heartwood is not surprising as the darker heartwood contains a greater amount of extractives (Ekeberg et al., 2006). A step trend in BI is apparent around the HW–SW transition, which becomes reduced but does not disappear following acetone treatment. Treatment time tests suggest that the majority of extractable material is removed from the heartwood within the first 48 h of acetone treatment. This result indicates that 48 h, shorter than 72 h suggested by Frith (2009), is a sufficiently long treatment time (to remove extractives to the extent that it is possible with acetone) and so longer treatment times are unnecessary when samples are fully immersed.

Ambient and reflected light

BI measurements of several measurement tracks using a box with a white, smooth inner surface are presented in Fig. 5a. The results reveal BI data trends, that should not be present, varying in shape and magnitude for different sample orientations. This process was repeated using a box with a black, non-reflective inner surface (Fig. 5b) where all trends in essence disappeared. The results of scanning with a 'black' or 'white' background clearly

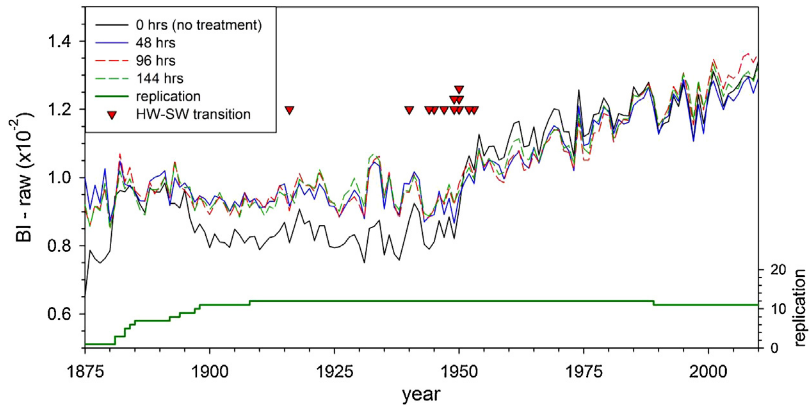

Fig. 4. Change in BI properties with varying acetone treatment time using samples from Shieldaig (measured along same tracks following each treatment).

indicate the potential of introducing a bias merely by the scanning process alone. Although ideally, scanning different sections in different ways would potentially minimise some of this potential low frequency bias, this would only be true if samples of the same length were scanned vertically in 50% of cases and upside down with the other 50%. However, this probably would not happen as samples would typically be scanned the same way in the majority of cases. If these results become systematically biased in this way, as illustrated by Fig. 5, it would translate to a warmer (colder) early (late) period.

Fortunately, the problem can be avoided entirely by using a black, non-reflective scanning background. Although placing a box over the scanner represents one way of doing this, it should be possible to achieve the same results through other means such as covering the scanner area with a black velvet fabric. It is likely that earlier BI chronologies produced for Scotland (Wilson et al., 2012) have been affected to some degree by this phenomenon and the extent of this should be assessed in future work and corrected for if possible.

Parameter comparison

EPS and replication

The assessment of averaging multiple cores per tree and its effect on the strength of common chronology signal with changing replication is presented in Fig. 6b. The Expressed Population

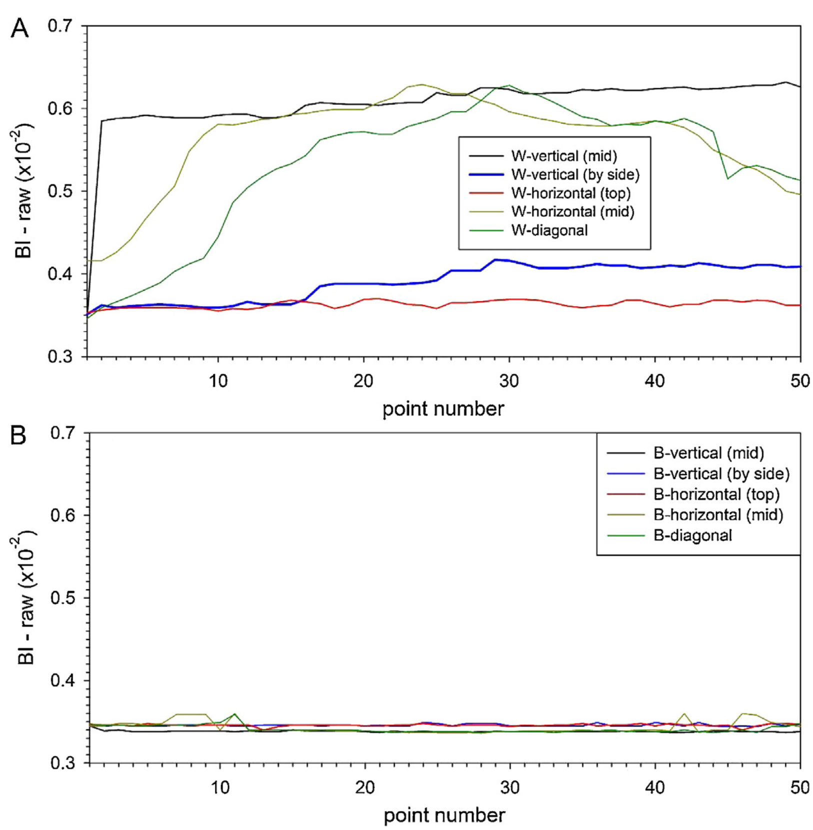

Fig. 5. BI measurements of empty scanned image using (A) a smooth, white background and (B) a non-reflective, black background along various tracks to assess the influence of reflected light (highlighted blue line represents the most frequently used placement). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of the article.)

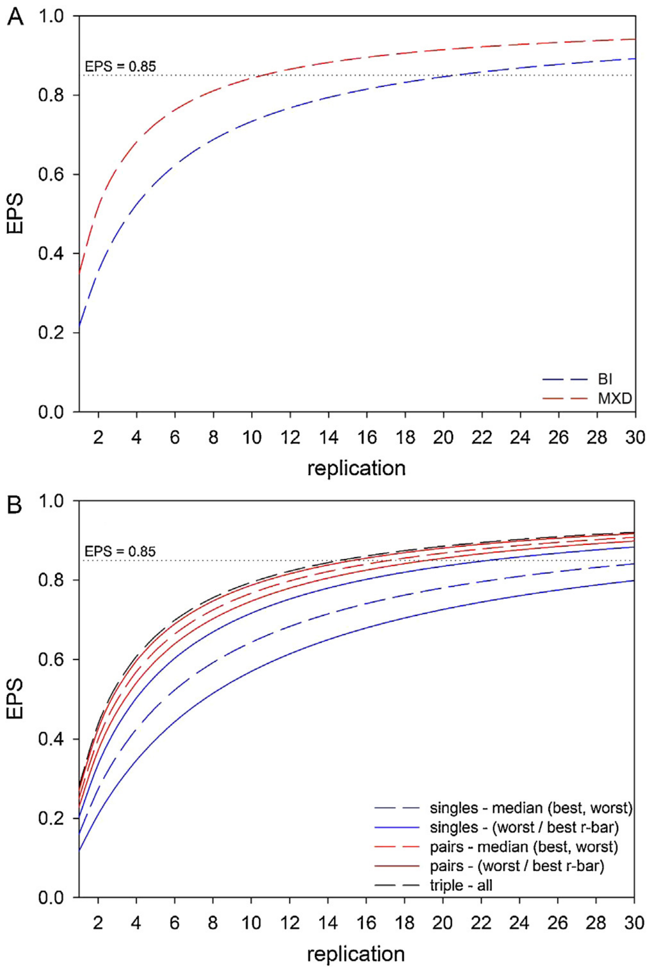

Fig. 6. (A) EPS estimates based on 21 detrended series using BI and MXD data from Ballochbuie with the 1901–2010 common analysis interval and (B) EPS estimates based on detrended BI data from 17 trees (three cores per tree) from Loch an Eilein with the 1901–2010 common interval for analysis. 'Worst/best' represents the combination of 17 series producing the worst and best RBAR (and EPS). One series per tree is used for the 'individual' results (i.e. A or B or C). Averages of two cores from each tree (i.e. A + B or B + C or A + C) are used for 'pairs', and 'triple-all' represents the average of all three samples (i.e. A + B + C). Average (worst/best) simply represents an average of the worst and best results of the 'individual' and 'pair' results, indicating a typical (most probable) scenario (between-tree RBAR is used in both (A) and (B) as effectively only one series per tree is used for each analysis).

Signal (EPS – Briffa and Jones, 1990) increases for any given level of replication when BI data from an increasing number of within-tree cores are averaged. However, greatest gains occur when using an average of two cores instead of one, with improvement diminishing as the number of averaged cores increases. The EPS and sample replication results provide evidence in favour of using averaged data of several same-tree samples as opposed to individual cores when using BI data. The possibility of obtaining stronger common signal by using the average of more than one same-tree sample can be useful when the number of tree samples is limited or the common signal is weak. This may be particularly helpful in the context of attempting to date undated samples (e.g. subfossil or historical wood).

A similar analysis of EPS using Ballochbuie BI and MXD data with only single cores per tree is shown in Fig. 6a. These results demonstrate that BI data contain a weaker common signal than MXD when replication is equal for both. This observation was also highlighted by Wilson et al. (in review). While at the individual tree level the signal from BI can be improved by averaging 2 or 3 radii, MXD ultimately still has a stronger common signal. However, as the costs and effort associated with generating additional BI data are negligible compared to MXD, this does not present a problem as in most cases more data can easily be obtained by sampling more trees.

BI vs. MXD

Correlation response function analysis (CRFA) of monthly EMST data against detrended chronologies from Ballochbuie (Fig. 7) and Ryvoan (Fig. 8) over the 1866–2009 period reveal the highest seasonal response to mean temperature for the July–August (JA) season when considering BI and MXD results for both sites (although MXD at Ballochbuie responds marginally more strongly to the broader February–August season). The Ballochbuie results of the two parameters are nearly identical, while the Ryvoan data indicate a modestly better representation of JA temperatures using BI. Using the period when EPS > ~0.85, the correlation between the two parameters is $r = 0.90$ ($p < 0.001$) for Ballochbuie for the 1768–2010 period and $r = 0.85$ ($p < 0.001$) for Ryvoan for the 1811–2010 period. Furthermore, despite a few minor departures, very few qualitative differences are apparent between the MXD and BI chronologies of the two sites (Figs. 7b and 8b). The CRFA results and the chronology comparisons suggest that despite all of the potential biases highlighted above, given that replication is sufficiently high, BI data from acetone treated samples can represent temperature similarly to MXD. The similarity and high level of agreement between the two parameters is also highlighted by their high between parameter correlations. Similar results were also reported in Wilson et al. (in review).

Ethanol vs. acetone

In order to test whether different resin extraction methods may lead to differences in results, the response to monthly temperature data was also assessed using a subset of nine sample pairs from Ballochbuie that either underwent acetone or ethanol treatment and were then measured with CooRecorder (Fig. 9). MXD data from the same samples as 'BI Stockholm' were also included for comparison. While the 'Stockholm' BI chronology responds marginally better to the July–September season than to July–August, 'St Andrews' BI has a stronger response with JA temperature. MXD also mainly responds to JA temperatures, although there is also a strong response to a broader February–August season. On the whole, with this particular sample subset, 'Stockholm' BI appears to show the best response followed by MXD and then 'St Andrews' BI. These results appear to provide an interesting indication of the relative strength of each method. However, they should be interpreted with caution as replication is not especially high (~20 series required for EPS > 0.85). Additionally, although the ethanol treated 'BI Stockholm' and MXD data were measured from the same samples ('B' cores), the acetone treated 'BI St Andrews' data were measured from 'A' cores (i.e. not the exact same samples). This caveat is further emphasised by the fact that the 'BI St Andrews' correlations in Fig. 9 are lower compared to MXD data, although those results represent a subset of the Fig. 7 data which show 'BI St Andrews' and MXD correlations that are more similar. Nevertheless, the results suggest that BI data from hot ethanol-treated samples may produce marginally better results than cold acetone-treated samples. It is not, however, possible to draw a conclusive statement whether one method is better than the other based solely on these results. What these results suggest is the stronger common signal of the MXD data when replication is low as already discussed in relation to Fig. 6a.

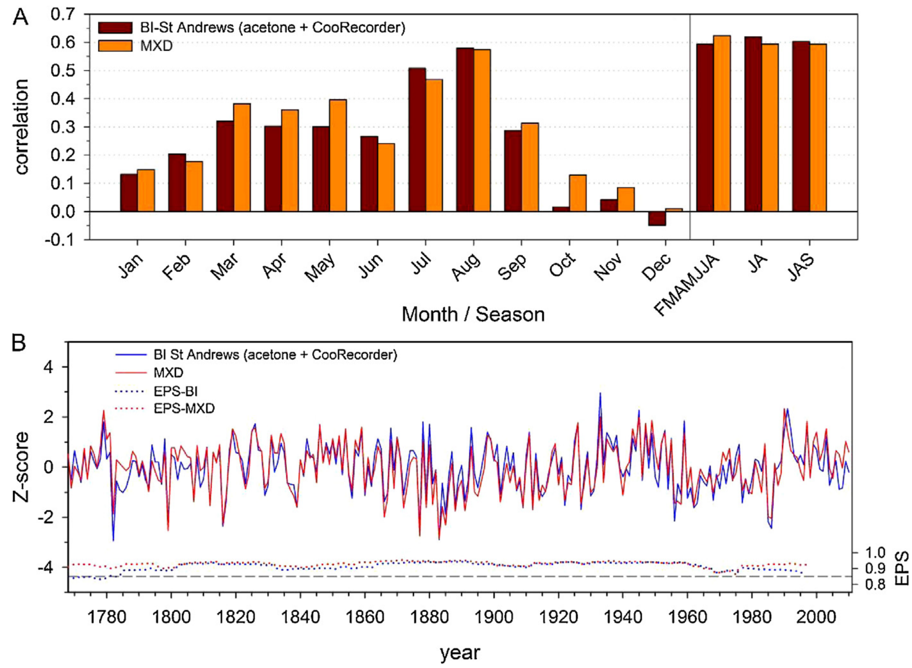

Fig. 7. (A) Correlation response function analysis of BI and MXD chronologies composed of 21 series (same tree samples) from Ballochbuie with monthly and seasonal EMST data over the 1866–2009 period and (B) time-series of the chronologies with EPS calculated using 30-yr running RBAR with 29-yr overlap.

CooRecorder vs. WinDendro

In order to evaluate any potential difference between measuring systems, a set of 'Swansea' ethanol treated BI samples were measured using CooRecorder and also with WinDendro (Fig. 10a and b). The direct comparison of BI data treated with ethanol but measured separately with CooRecorder and with WinDendro demonstrates that although not identical, the two methods broadly produce chronologies that are very similar. Overall, the monthly and seasonal responses are very similar for both datasets, although the CooRecorder results display higher correlations with instrumental temperature data for all months prior to October compared to the WinDendro results, which is also reflected in the seasonalised parameters.

Although in this case the data were generated from the same samples, these results should however be viewed with some caution, taking into account the limited sample replication. It is likely that results would be more comparable with higher replication. One additional explanation for any differences in BI data produced by each measuring system is that CooRecorder integrates information from a larger area of latewood (i.e. using a window) to produce

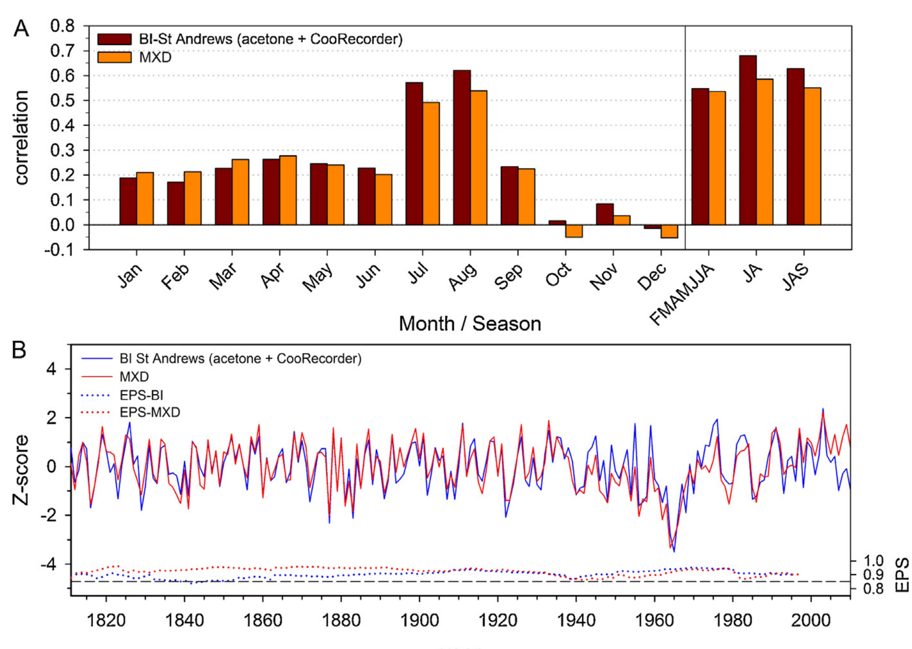

Fig. 8. (A) Correlation response function analysis of BI and MXD chronologies composed of 17 series (same tree samples) from Ryvoan with monthly and seasonal EMST data over the 1866–2009 period and (B) time-series of the chronologies with EPS calculated using 30-yr running RBAR with 29-yr overlap.

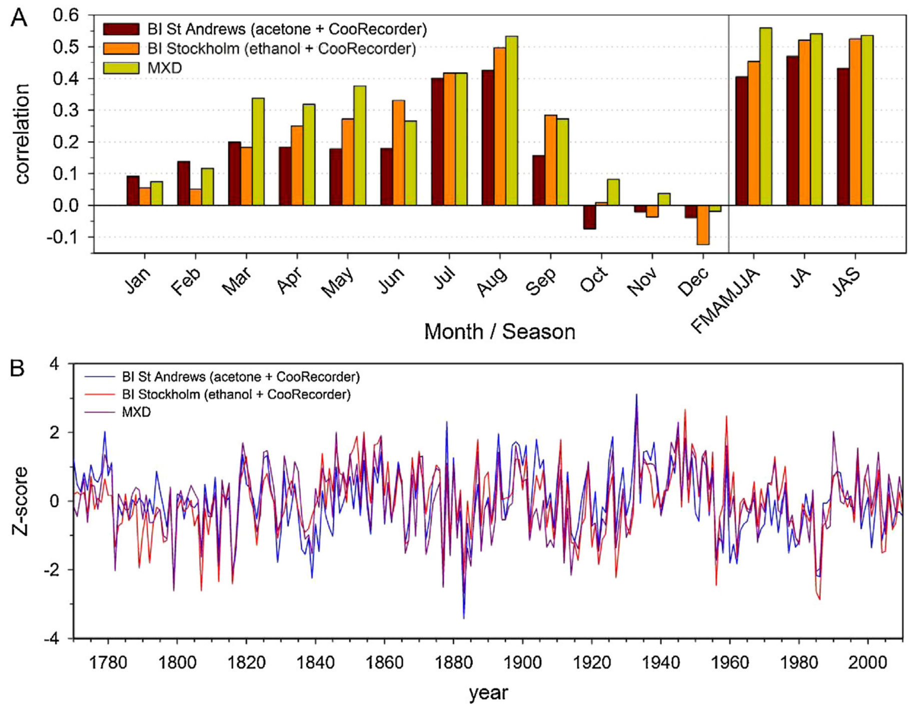

Fig. 9. (A) Correlation response function analysis of BI and MXD chronologies composed of nine series from Ballochbuie with monthly and seasonal EMST data over the 1866–2009 period and (B) time-series of the chronologies for the period when replication is more than seven series (MXD and BI Stockholm were measured on same samples, while BI St Andrews was measured from same tree samples as the other two chronologies).

an average minimum value of blue reflectance while WinDendro generates a BI value from a single point of lowest reflectance from a more restricted area (i.e. along a track). We should stress that at this time, as the two systems measure BI using different approaches, the merging of BI data from the two systems should be approached with caution and it is recommended that they be checked and adjusted for homogeneity prior to combination (analogous to different density measurement systems). An assessment of the relative merits of each system will require a more systematic and detailed examination in future.

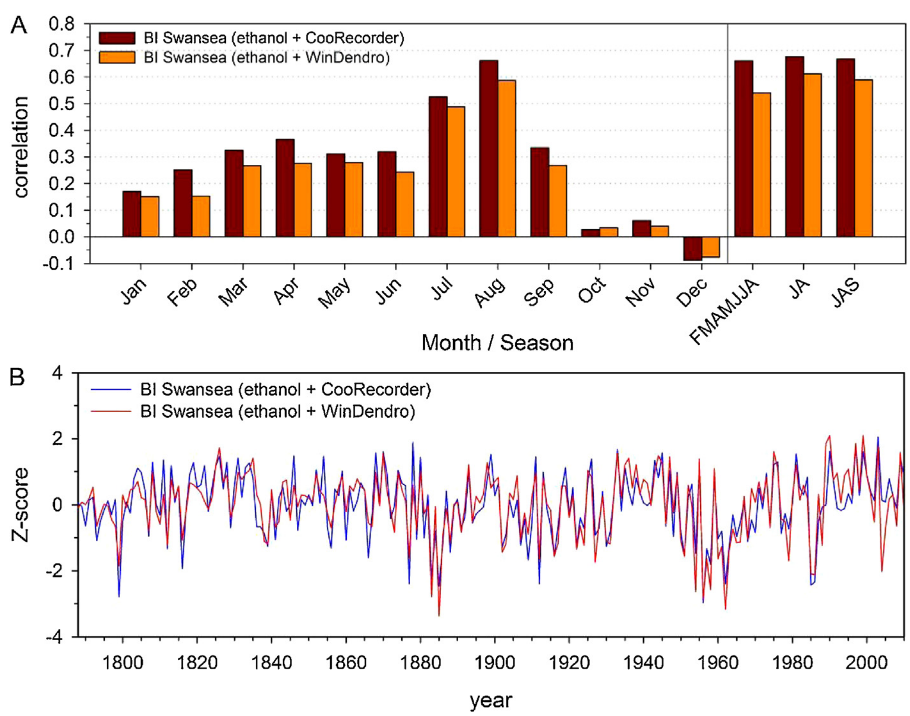

Fig. 10. BI chronologies from Ballochbuie consisting of nine series generated using ethanol Soxhlet extraction and measured either with CooRecorder or WinDendro (same samples used). (A) Correlation response function analysis with monthly and seasonal EMST data over the 1866–2009 period; (B) full length graph of chronologies for the period when replication is more than seven series.

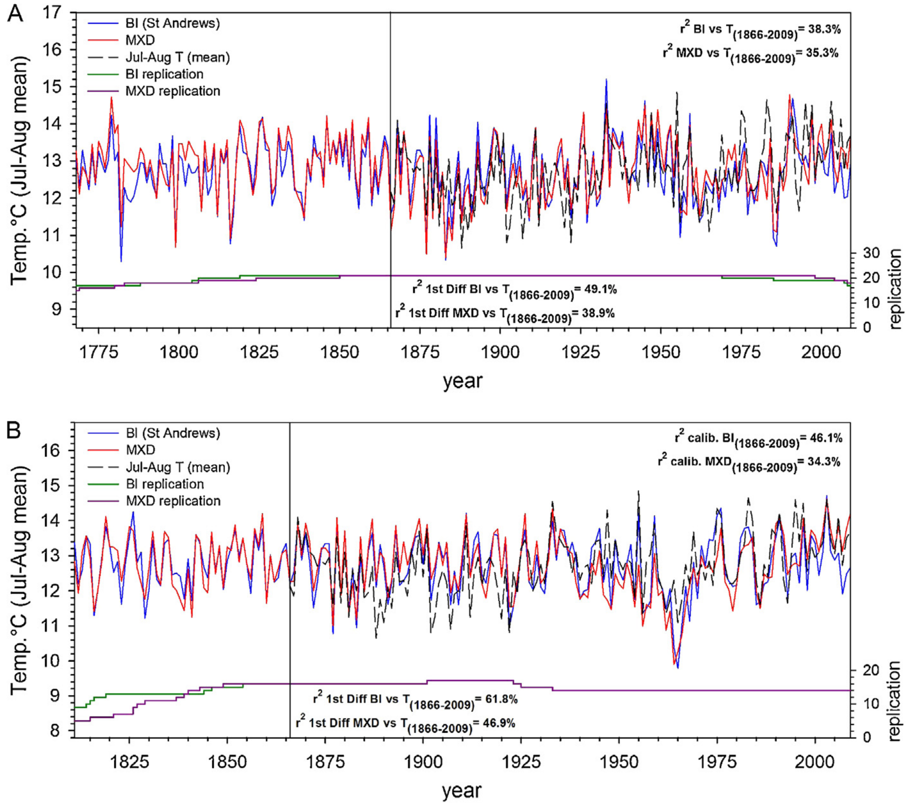

Fig. 11. BI and MXD scaled (Esper et al., 2005) reconstruction of mean July–August temperatures using the 1866–2009 calibration period based on (A) Ballochbuie (21 series) and (B) Ryvoan (17 series) MXD and BI chronologies (samples from same trees).

Temperature reconstruction

Fig. 11a and b presents reconstructions of JA mean temperature performed using the full 21 series Ballochbuie and 17 series Ryvoan chronologies of BI and MXD introduced in Figs. 9 and 10. A comparison of the regular and first-differenced r^2 results between each of the two parameters and JA temperatures indicates a higher percentage of variance explained at higher frequencies for both parameters. Although BI calibrates somewhat better with Ballochbuie and noticeably better with Ryvoan than MXD (despite a clear disturbance event in the early 1960s in the Ryvoan data), BI and MXD produce very similar results. These results are in agreement with results from previous studies (e.g. McCarroll et al., 2002; Campbell et al., 2007). In fact, they suggest that BI is not only comparable to MXD, but that in some ways and under certain circumstances the parameter is able to modestly surpass the results obtainable with MXD, as is the case here and as was reported in Campbell et al. (2007).

The first-difference results indicate that although BI data explain more of the JA temperature variance than MXD, proportionally more of the high-frequency variance is explained by BI, suggesting perhaps that at lower frequencies it does not perform as well. One likely contributor to this effect is the previously noted HW–SW bias that could still exist even after acetone or ethanol resin extraction has been performed. Based on a visual assessment of samples treated with ethanol using a Soxhlet apparatus, this feature does not appear to be entirely removed using this treatment method either. This perhaps suggests that there may be limits regarding the extent to which both acetone and ethanol treatments are able to reduce wood discolouration and it is therefore clear that this issue requires further attention.

To explore these ideas further, Fig. 12a and b highlights differences in the trends of non-detrended BI and MXD chronologies using data from Ballochbuie and Ryvoan. The raw non-detrended BI data from both sites clearly show a divergence in trends in the 20th century related to the HW–SW colour change. In addition to earlier results (Fig. 4) suggesting that a step-trend remains after immersion in acetone, these results further indicate that some extractives are not removed, causing a discolouration difference between the HW and SW. This is also observed visually in many samples. Note that this is also relevant for samples processed using the ethanol Soxhlet approach and regardless of which measuring system is used. Figs. 7 and 8 suggested that the Hugershoff detrended MXD and BI chronologies for both locations were very similar. However, Fig. 12c and d, which show both chronologies after using a more conservative linear regression function for detrending, clearly highlight a potential bias in the recent period of the BI data where the recent warming signal, clearly picked up in the MXD data, is not captured in the BI chronologies. This issue was also highlighted in Björklund et al. (2014).

In order to optimise the utility of BI from samples containing a distinct HW–SW colour change and thus yield useful information in the low-frequency domain in addition to mid-to-high frequencies, an objective method must be developed to minimise the non-climatic colour trend. There are several potential solutions to this type of problem including (1) utilisation of a combination of hydrogen peroxide and ethanol treatments explored by Sheppard and Wiedenhoeft (2007), (2) statistical adjustment of the raw data from the HW and SW sections relative to each other to eliminate an artificial step or gradient in the data, (3) digital post-processing of the scanned image to adjust the reflectance levels of the HW relative to the SW, (4) using a high pass/low pass filter approach to retain

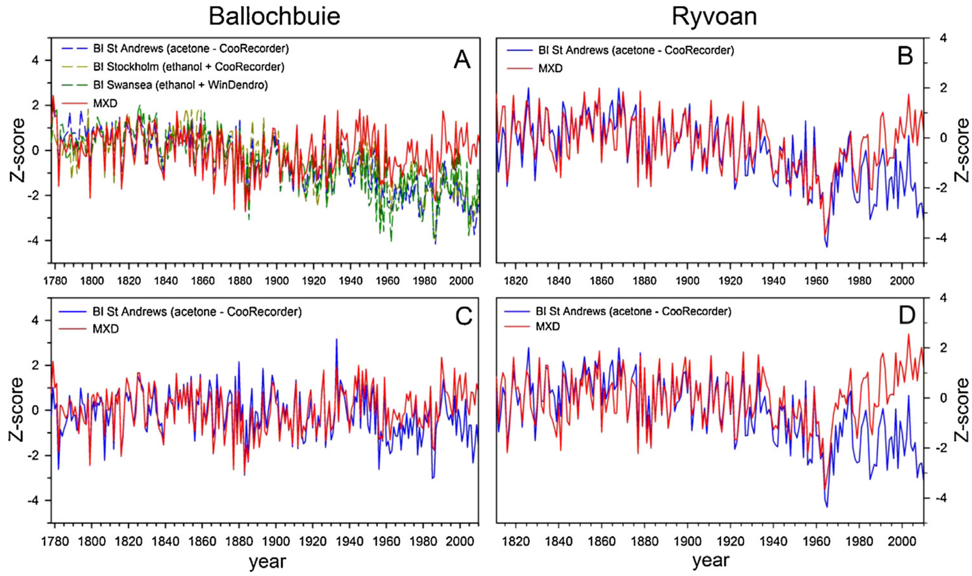

Fig. 12. Raw (non-detrended) MXD and BI chronologies converted to *z*-scores for (A) Ballochbuie (21 series – same as in Figs. 7 and 11a), ethanol-treated 'Stockholm' samples (nine series – same as in Fig. 9) and ethanol-treated samples measured with WinDendro (nine series – same as in Fig. 10) covering the 1778–2010 period, and (B) Ryvoan (17 series – same as in Figs. 8 and 11b) covering the period 1811–2010 (raw BI data were inverted for comparison); (C) and (D) include the full 21 series and 17 series BI and MXD chronologies in (A) and (B), respectively, detrended using linear regression (Ballochbuie chronologies were normalised to the 1778–1950 period and Ryvoan chronologies to the 1811–1950 period to emphasise any differences in trend post-1950).

the high frequency variance from BI and low frequency variance from RW, and then combining the two band-pass filtered parameters into a single time-series optimised at each frequency (Osborn and Briffa, 2000), or (5) using the reflectance properties of earlywood to adjust those of the latewood for each ring as proposed by Sheppard et al. (1996). This approach has recently been further developed in the BI context by Björklund et al. (2014) who presented the adjusted BI parameter (termed ΔBI) in order to adjust for local discolouration and removal of any undesired trends unrelated to lignin content.

While the chronologies presented in Fig. 11 undeniably produce results that are at least as good as MXD data in their detrended form based on their correlations with instrumental temperature data, the utilisation of the more flexible Hugershoff detrending here serves to illustrate that it is possible to overlook limitations of the raw, non-detrended data and which the detrending process can mask. Although Hugershoff-detrended chronologies appear to behave well, failure to adequately address anomalous trends in the raw BI data could undoubtedly affect low frequency trends and result in the production of potentially biased chronologies and reconstructions, particularly if they are intended to retain low frequency variance using methods such as Regional Curve Standardisation (RCS – Briffa et al., 1992b; Briffa and Melvin, 2011) or signal free detrending (Melvin and Briffa, 2008).

Conclusion

BI is a promising new parameter for the reconstruction of past climate from conifer tree-ring samples. Although a variety of factors can potentially affect the quality of BI data to various degrees, in many cases their effects can be identified and minimised. The direct comparison of BI and MXD for the two sites in this study confirms the ability of BI data to represent summer temperatures in a similar way to MXD – at least at mid-to-high frequencies. However, differences in methodological treatment of BI data generation and their consequences for data quality should be explored further.

The ability of BI to capture longer time-scale information (i.e. using RCS or signal free methods that better preserve mid-to-low-frequency variance) need also be explored further (see Wilson et al., in review). Although BI undoubtedly performs at least as well as MXD in the mid-to-high frequency domains, there is evidence to suggest that low frequency trends may not be fully represented. This effect is most likely primarily caused by the HW–SW colour difference and the inability of the chemical treatments as described herein to fully remove this discolouration. Such factors warrant further attention and require more detailed evaluation in order to ensure that BI data are able to adequately capture climatic variability at all frequencies. However, although this presents a challenge for coniferous species with distinct HW–SW discolouration, it should in no way affect those species that do not exhibit such HW–SW differences, although other sources of discolouration may still pose a problem.

Discolouration of the wood surface is among the most serious bias that may affect BI data. This issue may have several potential causes including fungal growth on tree cores that are not allowed to dry adequately, use of inferior quality sandpaper in the sanding process, discolouration of subfossil material due to decay, and possibly the treatment of historical material with dyes, varnishes, etc. However, the quick, simple and affordable generation of BI data using CooRecorder presents a unique opportunity to assist with dating of historical or subfossil wood material (Wilson et al., 2012). In particular, the strong common signal which, at mid-to-high frequencies at least, is comparable to that of MXD, reduces the level of uncertainty inherent in attempting to crossdate material of unknown temporal origin.

In this paper, we have introduced the methodological steps required to generate BI data using CooRecorder. The use of this software allows the affordable generation of climate-sensitive data that appear to be comparable to MXD. In this way, BI data can be generated for more sites and at a lower cost than would ever be possible for MXD. In addition to refining sample preparation procedures and exploring signal stability through time, a challenge for further work will be to overcome potential lower frequency biases in those species (e.g. Scots pine) that exhibit a distinct colour change between the heartwood and sapwood. Standard resin extraction methods do not remove this potential bias entirely and therefore other techniques need to be explored. Although a range of potential solutions exist to address this issue, the most feasible of these approaches, which will be explored in further research, includes generation of the ΔBI parameter (Björklund et al., 2014) in order to adjust for local discolouration or by utilising only the high-to-mid frequencies from the BI data with low frequencies from RW in a single chronology to produce a combination of the two high/low-pass filtered time-series (Osborn and Briffa, 2000).

Although some methodological issues still remain to be resolved, it is certainly not the case that there is little justification for the use of BI over MXD as suggested by Tene et al. (2011). On the contrary, the ease with which BI data can currently be generated and relatively low associated costs alone should justify the attractiveness of this parameter for dendroclimatological research.

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