Technical Note: Hyperspectral Lidar Time Series of Pine Canopy Physiological ParametersChlorophyll Content

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9 Abstract

We present an empirical application of hyperspectral lidar for monitoring the seasonal and spatial changes in pine chlorophyll content and upscaling the accurate leaf-level chlorophyll measurements into branch and tree level. The results show the capability of the new instrument for monitoring the changes in the shape and physiology of tree canopy: the spectral indices retrieved from the hyperspectral point cloud agree with laboratory measurements of the chlorophyll content. The approach opens new prospects for replacing destructive and labor-intensive manual sampling with remote observations of tree physiology.

17 **1** Introduction

The photosynthetic activity in tree canopy is an indicator of tree health. Vigorous trees with high foliar biomass and chlorophyll content have high carbon assimilation capacity. Stress in vegetation has been shown to induce changes in the photosynthetically-active pigments such as chlorophyll a and b. Therefore, the leaf chlorophyll content is an important indicator of the photosynthetic capacity as well as tree productivity and stress (Coops et al., 2003,Lausch et al. 2013).

The leaf properties and the distribution of chlorophyll and nutrients within a canopy vary as a function of time and space, and depending on the resource availability (Wang and Schjoerring, 2012, Peltoniemi et al., 2012). Seasonal changes in pPlant phenology and seasonal chlorophyll content cycle are correlated to the CO_2 flux. For monitoring these seasonal variations, methods are needed for accurate and nondestructive chlorophyll estimation, both at the leaf and canopy level (e.g., Gond et al., 1999). Chlorophyll estimation with spectral remote-sensing has been implemented increasingly in a number of studies (e.g., Coops et al., 2003, Lausch et al., 2013), but improved resolution and more accurate 3D position for the spectra are still being called for, to extend the accurate leaf-level measurement into canopy and stand level (cf. Gaulton et al., 2013). To investigate the spatial variation of the photosynthetic capacity and self-shading of photosynthetically active tissue, the canopy and branch structure must also be included in the measurement.

36 One way to provide simultaneous structural and spectral information is lidar combined with 37 hyperspectral passive sensing (e.g., Thomas et al., 2006, Asner et al., 2007, Jones et al., 38 2010), but new applications using multi or hyperspectral laser scanning have increased quite 39 recently. Hancock et al., (2012) demonstrated the potential of dual wavelength, large-40 footprint, spaceborne lidar to separate ground and canopy returns using the extra information 41 contained in a spectral ratio to complement the canopy height from laser scanning. Three-42 dimensional (3D) distributions of vegetation biochemical properties were measured with 43 spectral indices developed for the Salford Advanced Laser Canopy Analyser (SALCA), which is also a dual-wavelength lidar (Gaulton et al., 2013). A similar approach was used in the 44 Dual-Wavelength Echidna Lidar (DWEL) (Douglas et al., 2012). A multispectral canopy lidar 45 has also been introduced for simultaneous retrieval of vegetation structure and spectral indices 46 47 (Woodhouse et al., 2011). In this approach, a tunable laser operating at four wavelengths was used. The limitation of empirical vegetation indices estimating chlorophyll content is that they 48 49 are also affected by the canopy structural properties. In addition, they can be affected by the internal structure, size, surface and shape of leaves and can thus be species-specific, requiring 50 calibration when applied to specific species (Zhang et al., 2008). 51

In this technical note, an application of the recently developed hyperspectral lidar instrument (Hakala et al., 2012) is presented for monitoring the seasonal and spatial changes in pine chlorophyll content. As a non-destructive method, the capability of the instrument to upscale the accurate leaf-level chlorophyll content measurements into branch and tree level has been investigated and validated with chemical analysis of chlorophyll content. In this study, three spectral indices that showed good correlation with Scots Pine shoot chlorophyll concentration using the HSL instrument in Nevalainen et al. (2014) were used.

59 2 Materials and methods

Hyperspectral lidar (HSL) is a prototype laser scanning instrument (Hakala et al., 2012)
utilizing a supercontinuum laser. White laser pulses are sent-transmitted to a target and the

62 distances of reflected echoes are determined from time of flightare timed for distance. A spectrograph and an avalanche photodiode (APD) array connected to a high-speed digitizer 63 are used to determine the spectrum of each returning echo by measuring the intensity of the 64 65 echo at multiple wavelengths. Also the intensity of each transmitted laser pulse is measured 66 and used to normalize the echo intensity. Current prototype configuration uses a 16 element 67 APD array and an 8 channel digitizer, enabling us to measure at 8 different wavelength bands: 68 545, 641, 675, 711, 742, 778, 978, 1292 nm, full width at half maximum about 20 nm. Before 69 the target is measured Aa reference target with known reflectance (Spectralon) is measured at 70 distance intervals of about 30 cmfrom multiple distances and these data are used for 71 calibrating to calibrate the reflectance over the whole measurement range. Additionally the 72 Spectralon is placed in the scanned area during the actual measurement to validate the 73 calibration. The instrument and data processing presented in more detail in Hakala et al., 74 2012.

75 A Scots pine (Pinus sylvestris L.) was scanned five times during the 2013 growth season. The 76 tree was approximately 13 years old, 5.5 m high and it was growing in a small forest stand near the institute building. The HSL was mounted on a portable cart, and the tree was scanned 77 78 from two directions. The scans were co-registered using white spherical reference targets 79 placed on fixed locations on the target area. The distance between the scanner and the tree 80 was about 5 m. The tree was scanned with 0.1° horizontal and about 0.02° vertical resolution and the resulting point clouds contained 200 000- 470 000 echoes from the tree. The beam 81 82 diameter at the target was about 5 mm.

83 Needle samples were taken immediately after the scan for laboratory analysis. Six branches 84 were selected and the samples were taken from these branches according to needle cohorts 85 (current year needles, and 1-, 2, and 3-year old needles). Two needle pairs were taken from each cohort of each selected branch. Analysis of the chlorophyll contents followed the 86 87 protocol described in Wellburn (1994) for extraction with dimethyl-sulfoxide (DMSO). After 88 extraction, chlorophyll concentrations determined from solvents the were 89 spectrophotometrically using wave-lengths 480.0, 649.1 and 665.1 nm (resolution 0.1 - 0.590 nm).

91 Two of the six sampled branches were clearly identifiable from the HSL point cloud, having
92 enough point density and long enough growth of the branch. Parts of the Previous year cohorts
93 branches that carried previous year needles were selected for further analysis, since they had

needles present during all measurements. <u>Therefore the following analysis is performed for</u>
<u>two cohorts and five measurement dates.</u> The parts of the point cloud containing the selected
branch partscohorts were isolated in post processing. Three spectral indices were tested for
determining chlorophyll content of the needles. <u>Since it was not possible to tune all required</u>
wavelengths to optimal positions for every index, we used the nearest available band.

99 The Modified Chlorophyll Absorption Ratio Index using reflectance at 705 and 750 nm 100 (referred here as MCARI750) was first presented by Wu et al. (2008). Contrary to the original 101 MCARI (Daughtry et al. 2000), MCARI750 uses reflectance at 705 and 750 nm, which have 102 shown better sensitivity to high chlorophyll contents (Wu et al. 2008). MCARI has been 103 designed to measure the depth of the maximum chlorophyll absorption at 670 nm relative to 104 green reflectance peak at 550 nm and reflectance at 700 nm, at canopy scale (Daughtry et al., 105 2000).

106
$$MCARI750 = [(R_{750} - R_{705}) - 0.2 * (R_{750} - R_{550})] * (R_{750}/R_{705})$$
 (1)

107 The Modified Simple Ratio (MSR), developed by Chen (1996), strives to have low noise 108 effect and good linearity to vegetation biophysical parameters. MSR has been used to estimate 109 chlorophyll and Leaf Area Index (LAI) at canopy scale. Wu et al. (2008) also developed MSR 110 using reflectance at 705 and 750 nm, referred here as MSR2.

111
$$MSR2 = \frac{R_{750}/R_{705}-1}{\sqrt{R_{750}/R_{705}+1}}$$
 (2)

The Simple Ratio (SR) indices directly compare the reflectance and absorbance peaks of chlorophyll pigments, which make them sensitive to changes in chlorophyll content (Wu et al., 2008). Variety of wavelength combinations are used with simple ratio indices, but the one selected for this study is SR6 (Zarco-Tejada et al., 2001). It has been used to estimate chlorophyll at leaf level.

117
$$SR6 = \frac{R_{750}}{R_{710}}$$
 (3)

Additionally, normalised difference vegetation index (NDVI) (Rouse et al., 1973) was used to
separate needles from branches. NDVI is the most widely used vegetation index. It is based
on the contrast between high absorption at red and high reflectance at near-infrared (NIR).
NDVI has been developed for canopy scale and it has been used for both chlorophyll and LAI
estimation.

123
$$NDVI = \frac{R_{800} - R_{670}}{R_{800} + R_{670}}$$
 (4)

As the channels of the prototype HSL are limited to eight separate spectral bands, these indices had to be used with the closest available spectral band.

126 3 Results

The overall shape of the tree and changes in shape from May to November can be observed in Figure 1 where no spectral information is used. The changes in the shape and the spectra of tree parts are visible in the spectral point clouds. To demonstrate this, we plot the time series of the NDVI over the pine branch from May 15 to Nov 6, 2013 in Figure 2. The outbreak and growth of new shoots (May/Jun 2013) can be observed, as well as the year 2 parts drying outdefoliating (Sep/Oct 2013) and falling off completely (Nov 2013).

133 To validate the capability of the HSL to estimate the chlorophyll content using spectral 134 indices, we compared the HSL data with laboratory analysis over the growing season. We 135 present data for two branches cohorts, denoted M2_1 and M3_1 (one year old part of M2 and 136 M3), which were best visible in the HSL point clouds. The trends in the chlorophyll content 137 and the indices MCARI750, MSR2, and SR6 from HSL data are well reproduced for the individual branches (Figures 3-5). For all three indices, the sample branch M2_1 (year 1 part 138 of M2) was best correlated with the laboratory measurements with R^2 0.8-0.9. The R^2 for 139 MCARI750 and MSR2 for M3_1 was 0.7, whereas SR6 performed worse for M3_1 ($R^2 0.54$). 140 141 When the data from M2_1 and M3_1 were combined for regression, MCARI750 and 142 MSR2all indices correlated with the chlorophyll content measured in the laboratory, whereas there was distinct difference in SR6 value levels for M2 1 and M3 1 (Figure 6). The results 143 144 were worse for indices averaged over the entire tree point cloud (the right column in Figures 145 3-5), compared with the average of all year 1 needles measured in the laboratory. This is very 146 likely a result of the variation of the physiological conditions between the tree parts, which is 147 more pronounced when the sampling has been carried out over the entire tree (i.e., the point 148 cloud), rather than just a few needle samples (as in the laboratory experiment). All in all, the 149 analysis of branch parts shows that the spatial distribution of the HSL spectral indices 150 describes the chlorophyll content within the branch, although more measurements are needed 151 to better validate the results.

152 In figures 3-5, branch M2<u>1</u> and M3<u>1</u> laboratory measurements consist of two separate 153 needles only. More sampling should have been performed, however, the number of needles in

154 each branch part is limited and the tree had to be sampled several times during the year (this 155 emphasizes the need for non-destructive methods). The number of laser echoes from year 0 156 and 2 were highly varying; in the first spring lidar point clouds the year 0 growths were very 157 small providing very few echoes., and t The year 2 and older growths cohorts started dropping 158 needles before September measurement thus reducing the number of echoes during autumn compared to spring. If the laboratory measurements of all needles would have been 159 160 used Therefore we only used year 1 laboratory measurement of needles in plots 3-5 for whole tree (right column), since the weight of the year 0 and 2 laboratory measurements would have 161 been higher compared to the lidar point cloud (lidar point density variable and laboratory 162 163 sample number constant). Some lidar echoes still originate from the year 0 and 2 needles, 164 reducing the overall correlation between laboratory and lidar data for the whole tree.

The change in the shape of the tree point cloud is visible in Figure 1. The fact that tree shape can be retrieved from HSL point clouds has been shown before in numerous studies (see Kaasalainen et al., 2014 and Refs. therein). We have also shown in our previous study that the tree shape and its changes can be quantified from laser scanner point clouds using quantitative tree structure modelling (Kaasalainen et al., 2014). As the scope of this note was to show the added value of spectral data in the chlorophyll distribution monitoring, the changes in tree shape will be an object of our future study.



172

- 173 Figure 1. Co-registered point clouds from 2013-05-15 scan (grey) and 2013-11-06 scan (red).
- 174 Growth of the tree is visible and also some movement of the branches can be observed. The
- 175 <u>height of the tree is about 5.5 m.</u>



176

Figure 2. NDVI (see the colour bar for values) point clouds of a sample branch M2. The
growth of new needles (starting 05-27), already clearly visible new branch tips 06-19, fully
grown new needles 09-12 and dying and falloff of old needles (shown in bluish green , low
NDVI, colours in 09-12 and 10-03) are visible in the data measured at different times. The
measurement dates are shown in the plot titles.



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Figure 3. Top row: distribution of MCARI750 spectral index during separate HSL measurements, the central mark is the median, the edges of the box are the 25th and 75th percentiles, the whiskers extend to the most extreme data points not considered outliers. Middle row: Laboratory measurements chlorophyll a+b. Bottom row: Correlation of the spectral index and laboratory measurement. Subplot columns left to right: sample branch 3 year 1, sample branch 2 year 1, spectral index of whole tree and laboratory measurements of all year 1 samples.



Figure 4. Same as previous figure (top and bottom rows, laboratory data is the same as inprevious figure), this time using MSR2 spectral index.



194 Figure 5. Same as previous figure, this time using SR6 spectral index.

190



Figure 6. Correlation of spectral index and laboratory measurement for combined M2_1 and
M3 1 data. Left: MSR2, middle: MCARI750, right: SR6. Blue x: M3 1, red circle: M2 1.

198 4 Conclusions and discussion

We have shown that the hyperspectral lidar provides an empirical approach for efficient mapping the spatial distributions of tree physiological parameters that are correlated to reflectance of the foliage (such as chlorophyll a and b). Because the measurement is nondestructive, it can be repeated for the same target to produce time series of important tree functions, such as moisture condition, photosynthetic capacity, or physiological status.

We demonstrated that the seasonal changes in the shape and physiology of tree parts are visible in 3D; parameters affecting tree physiology can be quantified with spectral indices and linked to a specific location in the tree canopy using the HSL point cloud. We validated the method with reference measurements of chlorophyll a and b concentration in a laboratory. According to our results hyperspectral lidar can be used for the monitoring of the chlorophyll content, but similarly, the approach has potential in the monitoring of the water, carotenoid or lignin content, which all affect reflectance of the foliage (Austin and Ballare 2010).

211 The benefit of active measurement system, such as HSL, is that they measure backscattered 212 signal that has the potential to eliminate many of the multiple scattering and geometric 213 viewing effects caused by the canopy structure (Gaulton et al., 2013; Morsdorf et al., 2009). The major factors affecting the backscattered signal are the local incidence angle of the target 214 215 and the area of effective backscattering surface (Gaulton et al., 2013). These factors are also 216 present in this study as one 5mm footprint may include one or several needles with varying 217 incidence angles. However, the influence of these factors is similar with different wavelengths 218 measured at the same optical path. Thus by calculating spectral ratios (i.e. vegetation indices), 219 the influence of the incidence angle and target area can be reduced (Eitel et al., 2011; Gaulton 220 <u>et al., 2013).</u>

However, the influence of multiple scattering effects to the measured backscattered
 reflectance is not completely removed. Further study would be required to produce physically
 based model that would properly account for the multiple scattering of needles within single
 laser footprint and its effect to the measured backscattered reflectance. Some of the
 limitations of vegetation indices in chlorophyll estimation could be overcome by using
 inversion of radiative transfer models, such as LIBERTY (Leaf Incorporating Biochemistry

Exhibiting Reflectance and Transmittance Yields) (Dawson et al., 1998) which is specifically
 developed for needles, or PROSPECT model (Féret et al., 2011).

The tree was scanned from two directions only. Increasing the number of scans from different directions around the tree will improve the results by increasing the point coverage. This will require some instrument development to allow a more efficient field use. Increasing the point density is also an important object of instrument improvement. However, the prototype instrument was capable of showing the potential of 3D spectral measurements.

- A major factor causing error and uncertainty in this research was the use of nearest possible
 channel in vegetation index calculation instead of the band the index was designed to use.
 Especially close to the vegetation red-edge region even small shift in channel wavelength
 causes high change in reflectance. This affects the performance of the vegetation indices,
 especially with indices requiring channels at red edge. However, this was not considered as a
 major problem as the aim of this study was to test the ability of the HSL in chlorophyll
 estimation and not to optimize the performance of the indices.
- 241 Further work is needed to find the best spectral indices for different applications (e.g., 242 monitoring the effects of drought or limited amount of light on the physiology of different 243 tree parts), and then optimize the spectral channels to match with these indices. This will 244 improve the precision of the results. Increasing the number of spectral channels would also 245 improve the channel optimization and efficiency. Once the approach is well established and 246 calibrated, it has potential for replacing a number of laborious and destructive manual 247 experiments, and hence providing a new tool for remote observations of tree physiology. 248 Although the first results show the potential of the approach, further studies on the laser 249 interaction with the canopy are needed to establish the method physically.

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- 345 Comments by Mathias Disney
- 346 "I think this paper in general presents interesting results from a new hyperspectral lidar instrument.
- 347 This kind of measurement is likely to hold real promise for disentangling structure and spectral
- 348 properties of vegetation canopies. The work is generally clear and well-written. I have a few
- 349 comments on the limitations, but these are fairly minor. "
- 350 RESPONSE: Thank you for the comments.

351 "A limitation here is the very small number of needle samples taken for biochemical analysis - only 2 352 needles for M2 and M3 - what were the numbers for others? Chlorophyll content can vary quite a lot 353 between different cohorts of needles, so the resulting scatter plots are essentially extrapolations 354 from 2 needles only. I'm not sure this is useful. Fig 2 shows this variability (in part) - although of 355 course the fact is that the laser will return signals from multiple needles even for a single pulse. A 5 356 mm beam diameter is much larger than a single needle. What are the implications of this? There will 357 also be significant multiple scattering and shadowing at needle scale. Using spectral ratios may 358 average this effect out but it's still there. This means all results are a function of the spot size relative 359 to the needle size. This issue ought to be discussed and quantified if possible, or at least discussed. 360 Given the work is intended to look at small targets and the chemical analysis has been done on a very

361 small sample of these, I think this needs investigation."

RESPONSE: The denotation "M2, M3" was inconsistent in the article; all the analysis was for one year
 old cohort (M2_1, M3_1). The needle samples were taken from all cohorts of sample branches with
 needles during time of sampling. The cohorts were also isolated from the lidar point cloud, which
 was difficult to do reliably. Most of the cohorts were either too small to be reliably distinguishable
 from the point cloud or, after new growth, shadowed by other parts of the tree. Several branches
 were sampled, but only these two were clearly visible, and since the new grown cohort (year 0, eg.
 M2_0) was not present during all of the measurements we only used year 1 cohorts. Additionally,

- 369 only two needle pairs were taken from each cohort because we were worried that if we take too
- 370 many needles each time, the point cloud would be eventually affected by the reducing amount of
- 371 needles. The question about the spot size requires further research, but it mainly affects the intensity
- of the return signal and not the spectral content. And the effect of intensity is minimized by using
- 373 spectral ratios. At this stage, single echoes have significant uncertainty, and meaningful results can
- only be achieved by averaging.
- 375 "One other question here is why use spectral ratios at all? These are purely empirical and no
- rationale is given as to why one or other might be used. What kind of results are we to expect? There
- 377 are of course spectral models of needle reflectance properties which might be more appropriate to
- 378 use in analysis like this eg the LIBERTY needle model of Dawson et al."
- 379 RESPONSE: Spectral ratios are commonly used for estimating various parameters. They are simple,
- robust and easily implemented to our data. The purpose of this study is to show that useful
- information of the physiological state of the tree can be obtained by using this data. The advanced
- 382 modelling techniques (eg. LIBERTY) are certainly interesting, but out of the scope of this study and a
- 383 suitable topic for further papers.

- 384 "Minor points p15020 line19 phenology is periodic anyway by definition i.e. it's not seasonal
- 385 changes in phenology, it's just phenology. "
- RESPONSE: Rephrased to "Plant phenology and seasonal chlorophyll content cycle are correlated to
 the CO₂ flux."
- 388 "p15021 I5: worth mentioning work of Asner here has done a lot of this at large scale i.e. combining
 389 spectral and lidar. "
- 390 RESPONSE: Reference added.
- 391 "p15022 l15: why are these details approximate (scan resolution)?"
- 392 RESPONSE: The scan resolution is approximate due to mechanical configuration of the scanner; each
- 393 sweep is performed individually and the mirror is stopped after each sweep. At the beginning of the
- 394 sweep the mirror is accelerating and at the end of the sweep it's decelerating, while the pulse rate
- remains constant. Therefore the pulse density is higher at the beginning and end of the sweep.
- 396 "Fig 1 a scale would be useful, as would some indication of the accuracy of the co-registration. In397 addition, can the branches that are sampled be marked?"
- 398 RESPONSE: Marked the branches M2, M3 to the figure and added information about tree height to399 the caption.
- 400 "Fig 3 I'm not sure R2 values to 5 decimal places are useful. Also, can error bars be added to the401 scatter plots in fig 3-6?"
- 402 RESPONSE: Reduced to 2 decimal values, added error bars to figure 6. The error bars are also visible403 figures 3-5 top row.
- 404 ------
- 405 Comments by Anonymous referee #1
- 406 This is an interesting paper with significant novelty in testing a range of spectral indices derived from
- 407 multispectral laser scanning. The study is very small in scale and includes only very limited sampling,
- 408 but does provide an initial demonstration of the potential of this technology for plant physiological
- 409 measurements. In this context it does represent a significant and original contribution to the
- 410 literature. It is likely to be of significant interest to both the plant physiology and remote sensing
- 411 scientific communities. However, it could be improved by English language editing, clarification of the
- 412 methodology and a more thorough discussion of results as outlined below.
- 413 RESPONSE: Thank you for the comments.
- 414 Specific comments:
- 415 1)The title of the paper refers to 'physiological parameters' but the study only really considers the
- 416 single parameter of chlorophyll content. I think the title could be more specific and therefore more

- 417 fitting to the study. The lidar system would also be better described as multispectral as it measures
- 418 at only 8 discrete wavelengths.
- 419 RESPONSE: Changed the title from 'physiological parameters' to 'chlorophyll content'. The definition
- 420 of hyperspectral is generally vague. It is true that this particular prototype is more multispectral than
- 421 hyperspectral since it uses selected bands. However, we have 16 spectrally continuous channels
- 422 available and the reason we only use 8 is more financial and practical than technical. Therefore I
- 423 would define the instrument as prototype of hyperspectral.
- 424 2)Page 15022, lines 5-8: A single panel of 99% reflectance is used to normalise the lidar intensities.
- 425 This will account for range influences, but is a single reflectance panel sufficient? Is the detector
- 426 response linear? Is the laser output intensity constant? Given the focus of the paper is on the
- 427 intensity data the normalization method is of considerable relevance.
- 428 RESPONSE: The intensity of each transmitted pulse is not constant. This is taken into account by
- 429 measuring the intensity of each transmitted pulse using the same detector as for the echo
- 430 measurement. This is done by using a beam sampler and bypassing the other optics. This part of the
- 431 signal also triggers the measurement. We also have a 4 color Spectralon that we have used to check
- 432 the linearity, but these results are not published.
- 433 3)Page 15022, lines 18 24: Only a very small number of needles are sampled at each time period.
- 434 The majority of the results discussed rely on the Chlorophyll content of just 2 needles from 2
- 435 branches (i.e. 4 needles in total) at each time period. This limitation is acknowledged by the authors,
- 436 but does reduce conclusiveness of the study somewhat. Whilst little can be done retrospectively to
- 437 remedy this, the sample size should be made clear upfront in the methods not just later on in the
- 438 discussion (i.e. the number of needles per sample needs to be included here in all cases).
- 439 RESPONSE: Added information about sampling to methods. It was unfortunate that we were unable
- 440 to use most of the laboratory data. We measured 6 different branches with 3 cohorts each, but were
- 441 able to only use data of 2 cohorts. In future work the visibility of the sampled cohorts in lidar data
- 442 must be ensured.
- 443 4)Page 15023-15024: A range of indices are tested, benefitting from the multiple wavelengths of the
- 444 lidar. This is a novel and interesting aspect, representing an advance on previous attempts to retrieve
- 445 physiological parameters from single / dual-wavelength systems. However, a little more discussion of
- these indices would be useful in terms of the extent to which using different wavelengths (those of
- the lidar) to those for which they were designed might influence results and their sensitivity to
- 448 structural changes and multiple scattering. With this system, needles will be significantly smaller than
- the footprint so these factors as well as physiological parameters could have significant influence
- 450 (and structural changes might influence results based on a time series).
- 451 RESPONSE: We used slightly different wavelengths for the indices than what was stated in the
- 452 original articles describing the index. This will cause uncertainty and difficulties in comparing our
- 453 results to results published elsewhere. I added some discussion about this to results, and mentioned
- 454 this in methods before the indices are introduced. Also added to discussion that the use of spectral
- 455 indices reduce the effect of geometric effects (needles smaller than footprint). Also, since lidar
- 456 echoes from needles have high variance, multiple echoes are needed to get meaningful results.

- 457 5)Page 15024, line 14 (and fig. 2 caption): There is reference here to the branch parts 'drying out'. It
- is unclear where the physiological measurements to demonstrate the shoots are drying are and
- 459 which spectral index would show water loss (rather than other physiological / structural changes).
- 460 Only NDVI is plotted. Can it be demonstrated the NDVI changes are due to loss of moisture content?

461 RESPONSE: What was meant here was that the oldest needles defoliated and dropped off, which can

462 be observed as loss of chlorophyll and changes in NDVI. The drying out was a visual observation of

- the situation. This was normal for the growth of the tree, as these needles would be most shaded by
- 464 other parts and therefore less valuable than the new needles in outermost cohorts. I changed the
- 465 'drying out' to 'defoliate'.
- 466 6)Conclusions: I find the conclusions reached rather broad. The paper demonstrates, based on a 467 quite limited sample, that Chlorophyll content (not all 'physiological parameters') can be estimated
- 468 from a multispectral lidar system and that changes over time can be detected. It less clearly shows
- the extent to which spatial variation can be mapped as only a limited needle sample from a small
- 470 number of branches was taken. It would be useful to see a more thorough discussion of the findings
- 471 and the potential challenges of applying such systems (e.g the role of multiple scattering, how to
- 472 determine if a point is a needle rather than woody material, influence of structural change on
- 473 physiological parameter estimates). At least an acknowledgement of such issues should be included.
- 474 Re. the 'further work', what specifically would be needed that hasn't already been examined in the
- 475 hyperspectral remote sensing / leaf optical properties modelling literature? Are there reasons the
- 476 indices likely to work with lidar might be different to those for passive optical systems?
- 477 RESPONSE: Added several paragraphs to the discussion (paragraphs 3,4,6 in revised article) to478 address these questions.
- 479 7)Figure 3: While there is some relationship shown for mean values in Fig. 3 bottom row, it would be
- 480 useful to know if there was any statistically significant differences in laboratory and lidar
- 481 measurements for each branch (and the tree) between dates. The spectral changes look rather
- 482 limited and the indices quite variable (top row graphs) compared to the laboratory measurements.
- 483 RESPONSE: The variance of the lidar measurements is very high because of the nature of the
- 484 measurement. A single laser point may hit a needle/group of needles at any incidence angle relative
- 485 to the needle and also may hit any point at the length of the needle. Therefore only average of the
- 486 data is meaningful at this scale (cohort). The plots 3-5 top row show the 25 to 75 % percentiles (box)
- 487 that show significant differences between measurement dates and the trend of these follow
- 488 relatively well the laboratory measurements (as shown in the scatter plot).
- 489 Technical corrections:
- 490 There are a number of grammar errors in the paper. It would benefit from detailed language editing.
- 491 Page 15025, lines 16-19: This is unclear. Rephrase this. What is meant by 'the weight of the year 0
- 492 and 2 laboratory measurements'?
- 493 RESPONSE: Rephrased. What is meant here is that we took constant number of samples from the
 494 branches, but the point density varies when the needles are growing or defoliating. Therefore if we

- 495 average over whole tree and use all the laboratory measurements, the few needles that were left in
- 496 year 2 cohort after defoliation have higher weight in laboratory average than in the lidar point cloud,
- 497 since very few lidar points are acquired from cohort 2 compared to eg. cohort 0.
- 498 -----
- 499 List of relevant changes
- 500 Title changed to 'Technical Note: Hyperspectral Lidar Time Series of Pine Canopy Chlorophyll501 Content'
- 502 Introduction, paragraph 3, last 4 lines: Added information about the limitations of spectral indices.
- 503 Introduction, paragraph 4, last 3 lines: Added reference to our previous study (Nevalainen 2014), 504 where more information about the spectral indices used can be found.
- 505 Materials and methods, paragraph 1: Reformulated some sentences, and added more information506 about calibrations.
- 507 Materials and methods, paragraphs 3-4: Elaborated the fact that we only used two needle pairs from 508 two cohorts in final results and noted that we used nearest available bands for the indices.
- 509 Results: Notation for the branches was inconsistent and was corrected
- 510 Figure 1: added the positons of sample branches.
- 511 Figure 5: (SR6) was an issue with data processing; the values saturated causing the values for SR6 to
- 512 be too low. This also affected values of figure 6, right subplot. The values were also corrected to text,
- 513 but this didn't affect the conclusions from the data.
- 514 Figure 6: Added error bars for the index values.
- 515 Conclusions and discussion: Added paragraphs 3, 4, and 6 to address the issues raised by referees.
- 516
- 517