Models for estimating leaf pigments and relative water content in three vertical canopy levels of norway spruce based on laboratory spectroscopy

Lucie Cervena¹, Zuzana Lhotakova², Lucie Kupkova¹, Monika Kovarova² and Jana Albrechtova²

¹ Charles University in Prague, Faculty of Science, Department of Applied Geoinformatics and Cartography, Prague, Czech Republic; lucie.cervena@natur.cuni.cz, lucie.kupkova@gmail.com
² Charles University in Prague, Faculty of Science, Department of Experimental Plant Biology, Prague, Czech Republic; zuzana.lhotakova@natur.cuni.cz, monika.kovarova@natur.cuni.cz, jana.albrechtova@natur.cuni.cz

Abstract. Unique set of data was obtained during the field campaign in the Krušné hory Mts. (the western part of the Czech Republic) in August 2013. From fifty five representative 80-year-old trees of Norway Spruce (Picea abies L. Karst.) equally distributed on eleven study sites, branches were taken in three vertical canopy levels (sunlit productive upper and lower parts of a tree crown, shaded saturated part of a tree crown) and first three needle age classes were analysed. Spectral reflectance of these spruce foliage samples was measured in the range between 350 and 2,500 nm using an ASD FieldSpec 4 Wide-Res spectrometer in combination with the fibre optic contact probe. Some samples were also measured in the integrating sphere. Photosynthetic pigment (total chlorophylls, total carotenoids) contents and relative water content were determined in laboratory for all samples. The results of analysis of variance (ANOVA) show that the contents of pigments and relative water content are significantly different not only between the needle age classes (what is widely known) but also in the vertical canopy levels. There are only few studies dealing with vertical heterogeneity in Norway spruce canopy. Thus, the main goal of this study is to build and compare the statistically based prediction models for photosynthetic pigments and water content estimation for three vertical canopy levels of Norway Spruce. These results in classifications of biochemical and biophysical properties of Norway Spruce stands using hyperspectral remote sensing data.

Keywords. Norway Spruce, laboratory spectroscopy, Krusne hory Mts., RWC, Chlorophyll, Carotenoids.

1. Introduction

Remotely sensed leaf biochemical and biophysical properties can be used for large-scale spatial and temporal monitoring of vegetation physiological status or ecosystem functioning. For airborne hyperspectral data interpretation or modelling of reflectance at canopy level several foliar properties play minor role (e.g. internal leaf structure). However, leaf clumping, proportion of woody constituents or vertical gradients in biochemical and biophysical leaf traits may influence radiative transfer modelling at the canopy level.

Leaf biochemical composition affects foliar optical properties and can be retrieved from continuous spectral data [1], [2]. There are two modelling approaches to link content of biochemical compounds or biophysical parameters of vegetation to spectra – empirical (different regressions, e.g. [1]) and physical (radiative transfer models, e.g [3]). The main advantage of the empirical
models is their simplicity. The physical models are more precise but need more complex input information – e.g. PROSPECT needs at least four input parameters: leaf mesophyll structure parameter \( N \), chlorophyll concentration \( C_{ab} \), water depth \( C_w \), and the dry matter concentration \( C_m \) [4]. LIBERTY needs nine parameters (Average Cell Diameter, Intercellular Air Space, Baseline Absorption, Albino Absorption, Needle Thickness and concentrations of Chlorophyll, Water, Lignin/Cellulose and Nitrogen [5]. They usually require also a long computation time.

In mature tree canopies the vertical gradient of irradiance is the key factor, which determines leaf structure, biochemical composition and rate of physiological processes such as light harvesting and photosynthesis [6]. In terms of light-dependent leaf traits several studies were conducted with focus on leaf physiological functions or morphology (e.g. [7], [8]), but only few dealt with leaf optical properties along the vertical profile of the canopy (e.g. [9]). Wang and Li [9] showed that vertical variations in leaf biophysical and biochemical traits such as chlorophyll, water or dry mass content, significantly influence optical properties at the canopy level. Thus considering several vertical canopy layers consisting of leaves with different optical properties could be crucial for reflectance simulations and modelling at canopy level. Effect of vertical heterogeneity [9] and leaf clumping [10] on canopy optical properties and their modelling was studied as far for broadleaved trees and the information about conifer foliage is lacking.

Thus, the objectives of this study are: 1) to construct the prediction empirical models for photosynthetic pigments and water content based on laboratory spectroscopy for the three vertical canopy levels of Norway Spruce on the leaf level; 2) to compare the models for the individual canopy levels among them and with the model based on data from all levels.

2. Methods

2.1. Study area

The study area is located in the Western part of the Czech Republic in the Krušné hory Mts. Two localities situated about 50 km apart with different impact of the acidic deposition during 1970’s and 1980’s were selected. While trees in Přebuz (western part of the mountains) were healthy or just slightly damaged, trees in Kovářská (eastern part of the mountains) were heavily affected and still exhibited visible damage symptoms in 1990’s [11]. In 2013 the sampled trees on both localities already did not show any visible damage symptoms.

2.2. Field campaign

The field campaign was conducted within the period 22-25 August 201. On each locality six (Přebuz) resp. five (Kovářská) sites of even-aged forest stands older than 80 years were selected for further evaluation. From five representative trees per site branches were taken in three vertical canopy levels (sunlit productive upper and lower parts of a tree crown, shaded saturated part of a tree crown) and first three needle age classes were analysed (Figure 1). The content of photosynthetic pigments (total chlorophylls, total carotenoids) was determined using dimethylformamide extractions according to Porra [12], followed by spectrophotometric detection and calculations according to Welburn [13]. The pigment concentrations were expressed as weight of pigment per gram of needle dry mass (mg/g). The relative water content (RWC) was determined as the percentage of water in the fresh needles.

Simultaneously with the needle sampling the spectral reflectance of spruce foliage was measured in the range between 350 and 2,500 nm using an ASD FieldSpec 4 Wide-Res spectrometer in combination with the fibre optic contact probe. The radiance spectra were normalized against a 99% Spectralon white reference to produce relative reflectance spectra for each measurement. Shoots consisting of only one age class were arranged in the same direction to create a consistent layer to fill in the field of view (spot size 10 mm) of the contact probe. Shoots were placed on a

spectrally black surface to minimize the background spectral noise or radiation transmitted through the needles. The scan average on the spectroradiometer was set to 50 to improve the signal to noise ratio. Five independent spectra were taken on different parts of one sample and afterwards median spectrum was calculated (together 495 median spectra: 11 sites x 5 trees x 3 vertical canopy levels x 3 needle age classes). Due to the noise in the spectra in the 350 - 450 nm regions, this interval was excluded from further analyses.

Figure 1: Sampling scheme (adapted after Kopackova 2014 [14]).

2.3. Data processing

Firstly, hierarchical analysis of variance was performed. The aim of this method was to find out if there are significant differences in pigments (chlorophylls and carotenoids) contents and relative water contents between the studied localities, vertical canopy levels and needle age classes. Significant differences were proven between all the mentioned factors. Differences in pigments and relative water content between the localities were expected due to the different impact of pollution in these areas in the past, differences between the needle age classes are widely known [15], [16]. The differences in studied needle parameters between the vertical canopy levels reflected the light availability within the canopy and their relationships with needle optical properties will be further evaluated in this study. Obvious differences can be also seen in the averaged spectra, particularly in the NIR spectral region (Figure 2).
Secondly, selected spectral indices were calculated from the measured spectra: NDVI$_{705}$ [17], mNDVI$_{705}$ [18], MCARI [19], TCARI/OSAVI [20], TVI [21] and ANMB$_{650-725}$ [22] for correlations with total chlorophyll contents; CRI550, CRI700, RNIR*CRI550, RNIR*CRI700 [23], [24] for correlations with total carotenoids contents and water index (WI) [25] and normalize difference water index (NDWI) [26] for correlations with RWC. Also partial least square regressions (PLSR) using all the reflectance values and continuum removed (CR) values in range 450 – 2500 nm for estimation of RWC and pigments were performed. The regression models were built not only for all the data together but also for separate datasets containing data from only one canopy vertical level. The models were always trained on 4/5th of the dataset; data from one tree per site were used for the models validations. The models were evaluated using Coefficients of Determination (R$^2$) and Root Mean Square Errors (RMSE). All the calculations were performed in R software with pls package with help of Microsoft Excel 2007.

3. Results

Table 1 presents averages and standard deviations for the pigments and relative water contents in the different vertical levels of the tree crown. It can be seen that the averaged contents of pigments and water are slightly lowering with the higher position in the tree crown.
Results of all the computed statistical models are presented in Table 2. The best results were generally achieved by the PLSR method with coefficient of determination at least 0.61 (number of components used is presented in the brackets for each PLSR model in Table 2). By the simple regressions with the indices, the best results were obtained for models based on the data from productive lo-

Table 1. Descriptive statistics for the datasets for leaf biochemical and biophysical properties

<table>
<thead>
<tr>
<th>Part of the crown</th>
<th>shaded saturated</th>
<th>productive lower</th>
<th>productive upper</th>
<th>all data</th>
</tr>
</thead>
<tbody>
<tr>
<td>descriptive statistics</td>
<td>average</td>
<td>st.d.</td>
<td>average</td>
<td>st.d.</td>
</tr>
<tr>
<td>Total Chlorophyll (mg/g d.m.)</td>
<td>3.44</td>
<td>0.62</td>
<td>3.10</td>
<td>0.66</td>
</tr>
<tr>
<td>Total Carotenoids (mg/g d.m.)</td>
<td>0.44</td>
<td>0.07</td>
<td>0.41</td>
<td>0.08</td>
</tr>
<tr>
<td>Relative Water Content (%)</td>
<td>57.91</td>
<td>2.76</td>
<td>56.26</td>
<td>3.13</td>
</tr>
</tbody>
</table>

Table 2. Results of all the regression models between the spectra and biochemical and biophysical parameters (R^2 – coefficient of determination, number in the brackets – number of components considered in the PLSR model, RMSE – root mean square error; p-values for all the models were lower than 0.01.)

<table>
<thead>
<tr>
<th>Part of the crown</th>
<th>shaded saturated</th>
<th>productive lower</th>
<th>productive upper</th>
<th>all data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Chlorophyll (mg/g)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>statistical model</td>
<td>R^2</td>
<td>RMSE</td>
<td>R^2</td>
<td>RMSE</td>
</tr>
<tr>
<td>MCARI</td>
<td>0.6197</td>
<td>0.46</td>
<td>0.4956</td>
<td>0.41</td>
</tr>
<tr>
<td>TCARI</td>
<td>0.7107</td>
<td>0.38</td>
<td>0.5792</td>
<td>0.33</td>
</tr>
<tr>
<td>TCARI/OSAVI</td>
<td>0.7228</td>
<td>0.38</td>
<td>0.5588</td>
<td>0.31</td>
</tr>
<tr>
<td>TVI</td>
<td>0.5553</td>
<td>0.48</td>
<td>0.5516</td>
<td>0.46</td>
</tr>
<tr>
<td>PLSR (450-2,500 nm)</td>
<td>(6)</td>
<td>0.7763</td>
<td>0.33</td>
<td>(6)</td>
</tr>
<tr>
<td>PLSR (450-2,500) - CR</td>
<td>(5)</td>
<td>0.7758</td>
<td>0.33</td>
<td>(4)</td>
</tr>
<tr>
<td>Total Carotenoids (mg/g)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>statistical model</td>
<td>R^2</td>
<td>RMSE</td>
<td>R^2</td>
<td>RMSE</td>
</tr>
<tr>
<td>CRI550</td>
<td>0.1211</td>
<td>0.07</td>
<td>0.1538</td>
<td>0.07</td>
</tr>
</tbody>
</table>
**4. Conclusions**

The best prediction models for estimating the contents of leaf pigments and relative water content were achieved by the PLSR method (as in e.g. [27]). The simple linear regressions with the indices did not work very well, although they are often used [17] – [26]. The $R^2$ values for simple regressions with chlorophyll content reached in this study for shaded canopy part were comparable with values presented by Croft et al. 2014 [28] ($R^2 = 0.61$ - 0.59) on needles of *Picea mariana* using wide range of different indices. Poor CRI550 and CRI700 performance for prediction of carotenoids content is rather surprising as these indices have already been successfully applied on conifers (*Pinus sylvestris*, $R^2 = 0.72$ and 0.73 respectively) [29]. The best models using the indices were achieved for the datasets coming from the shaded saturated part of the tree crown. In contrary; the worst results were achieved for the sunlit productive upper part of the tree crown. This could be caused by the more complex shoot architecture occupying larger spatial fraction in this part of the crown. The needles from this part of the tree crown were often very short and/or very tough, perpendicular to the shoot, which may affect resulting spectra by additional scattering. Whether this is the main reason for less accurate models for the sunlit upper part of the Norway spruce crown could be tested in the future by another method of measurements – e.g. acquiring the spectra in the integrating sphere.

Croft et al. 2014 [28] also discussed that generally poorer performance of chlorophyll indices on conifers in comparison to broadleaved species is based on higher complexity of a coniferous shoots and in technical difficulties of reflectance measurements on needles even if an integration sphere is used. The highest $R^2$ values for regressions of indices with water content (0.57 and above) are similar with results achieved by Stimson et al. 2005 [30] on *Pinus edulis*. However, the different influence of the vertical canopy level on the performance of pigment- and water-related vegetation indices should be taken into account if dealing with multiple layer canopy data.

The PLSR showed the best model performance for all studied biochemical and biophysical needle parameters and also the variability in RMSE between three vertical canopy levels was lower than RMSE achieved with vegetation indices. We conclude that simple regressions using vegetation indices for prediction of needle biochemical and biophysical properties are more prone to be species- or structure-dependent and may cause bias if dealing with spectral data from multiple vertical levels within a coniferous canopy. We suggest that the light-driven vertical heterogeneity in biochemical and biophysical foliage properties should be taken in account when dealing with using field ground truth data for interpretation of airborne spectral data.

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References


