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OPTICAL INDICATORS OF PLANT PHYSIOLOGICAL ACTIVITY

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Abstract

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Retrieving information on the plant physiological status from spectral reflectance is a challenging task in many ways. The easiest way to get the information is through normalized vegetation indices, that are based on a normalization of reflectance in specific wavebands. Beside the most common spectral indices such as Photochemical Reflectance Index PRI or Normalized Difference Vegetation Index NDVI, several new indices has been proposed during the past decade as potential physiological indicators. In this paper, the performance of several of them for determining the physiological status of the foliage is evaluated on an experimental Norway spruce needles (*Picea abies* (L.) Karst) plot. Four needle classess of 27 years old spruce have been sampled throughout cloudy and sunny day. Needle classes were represented by the needles of the branchlets of the 4th, 7th, 9th and 12th whorls. Study was conducted on one-year old needles and sampling ran over several times a day to separate influence of dynamic processess on parameters. Results show that the ratio of reflectance in the green and red region (presented as ratio of reflectance at 560 nm and 694 nm) outperforms the others examinated vegetation indices, and suggest that it would be best suited for the characterization of the leaf status. Being always the highest in the uppermost part of the crown and the lowest in shaded part of the crown, parameter is strictly stratificated throughout the crown. Furthermore, parameter values correspond with intensity of physiological processess ongoing in needles during midday. The values of the other indicators seem to be affected by leaf pigment content and morphology too much. Neither PRI nor NDVI were able to distinguish differences in needle properties sufficiently.

Picea abies, reflectance, vegetation indices, photosynthesis, ecophysiology

Global and spatial estimates of plant photosynthesis are required for a comprehensive understanding of the terrestrial carbon cycle and the determination of CO_2 uptake by plants (Baldocchi, 2003). Key technique for evaluating CO_2 exchange between the canopy surface and its surrounding air column is eddy covariance measurements. Eddy covariance measurements allow to estimate carbon flux at local scales. To observe CO_2 exchange in global scale, appropriate techniques are required to approximate the plot measurements of eddy-covariance towers up to the larger areas.

There are three approaches that have been employed to identify and quantify carbon sources and sinks at regional and global scales: atmospheric transport modeling, ecosystem carbon exchange modeling (Battle *et al.*, 2000), and remote sensing driven statistical models (Rahman *et al.*, 2001; 2004). The first two of these approaches are complex and require a large number of parameters that are often scarce or not available at the required spatial and temporal scale (Randerson *et al.*, 2002). The third approach, on the other hand, is simple as it derives its parameters directly from remote sensing and calibrates them with ground eddy covariance measurements.

Basic model used for description of carbon uptake by plants is light use efficiency (ε). That express amount of carbon taken per unit of incoming or absorbed photosynthetically active radiation. One of the most common methods used for remote sensing of light use efficiency is the photochemical reflectance index (PRI) (Gamon *et al.*, 1992), a narrow waveband, normalized difference index that relates ε to a xanthophyll cycle-induced absorption feature at 531 nm. PRI is conventionally calculated as (R531 – R570) / (R531 + R570) (Gamon *et al.*, 1997; Peñuelas *et al.*, 1995), where R is the leaf reflectance at a given wavelength. A change in PRI is linked to the action of a photoprotective mechanism preventing the assimilation apparatus of plants from photooxidative damage, by dissipating excessive radiation energy as heat (Demmig-Adams and Adams, 1996). Seasonal variations in stands light use efficiency readable through PRI have also been connected to changes in photosynthetic pigment composition of plant leaves. To changes in chlorophyll/carotenoid ratio affecting the conversional factor of light energy into biomass (Stylinski *et al.*, 2002) and with long term stress effects on total chlorophyll concentration (Filella *et al.*, 2004). A degradation of foliar chlorophyll generally reduces foliar PRI as a result of the relative increase in reflectance at 570 nm (Moran *et al.*, 2000; Nakaji *et al.*, 2006; Sims and Gamon, 2002).

Numerous studies have shown that the normalized difference vegetation index (NDVI), a red and near infrared reflectance ratio, has a very close relationship to the fraction of photosynthetically active radiation (PAR) absorbed by green vegetation (fAPAR) (Myneni and Williams, 1994). NDVI value is affected primarily by the amount of the biomass observed (Fuentes *et al.*, 2006). However, NDVI may also be used as a chlorophyll index (Lichtenthaler *et al.*, 1996). Numerous works pointed out insufficient sensitivity to quick changes in the assimilation activity of forest stands (Stylinski *et al.*, 2002; Xiao *et al.*, 2004). Greenness indices are not sensitive enough, especially in evergreen stands that remain green even during drought periods when the assimilation activity is changing (Gamon *et al.*, 1995). On the other hand, in deciduous plants, the reduction of NDVI during autumn senescence leads to a distortion in ε prediction (Viña and Gitelson, 2005). Furthermore, greenness indices are getting saturated at stands with a higher leaf area index. At stands with a lower LAI, higher absorption by non-photosynthetically active woody tissue of trees leads to an overvaluation of NDVI (Asner, 1998).

A lower performance of both PRI and NDVI is also attributed to state of the atmosphere, particularly the presence of aerosols. An increasing influence of the atmosphere state on reflectance signal towards higher wavebands has been reported. Thereby, NDVI is influenced by the presence of atmospheric particles more. The vegetation index EVI, principally being a NDVI derived algorithm, has been designed to proofread the influence of atmospheric conditions directly from the measured spectra (Xiao *et al.*, 2003). An improvement in EVI is reached by incorporating the blue reflectance into the equation. More advanced variations of EVI are taking into account additional more sophisticated correctional factors describing the state of the atmosphere.

There are still numerous issues that will have to be resolved before using vegetation indices as accurate pointers of assimilation activity. For example, PRI has been used as a full-fledged physiological index for the first time after adjusting the viewing geometry to separate the extraneous effects from the physiological signal contained in stand level PRI by Hilker *et al.* (2010). The adjustment of viewing geometry is, however, based on a prior knowledge of the Bidirectional Reflectance Distribution Function (BRDF) of the forest stand examined for each particular LUE level. The basic requirement for realisation is a multi-angular observational setup (Hilker *et al.*, 2009).

However, relations have been derived from the knowledge of interaction between the leaf tissue and incoming photons at the elementary leaf level. Over the past two decades other potential vegetation indices for photosynthesis estimates emerged as well. Therefore, based on finding commonalities in simple, normalized vegetation indices within particular types of Norway spruce needles and comparing them with similar alternatives sensitive to variations in stands LUE according to the literature, this work was conducted to exploit background knowledge behind the sensitivity to differences in photosynthesis performance. For this purpose a database of 96 reflectance spectra was examined. Norway spruce (*Picea abies* (L.) Karst) was selected for this study as the most widespread tree in Czech Republic and as one of the most economically important coniferous species in Europe.

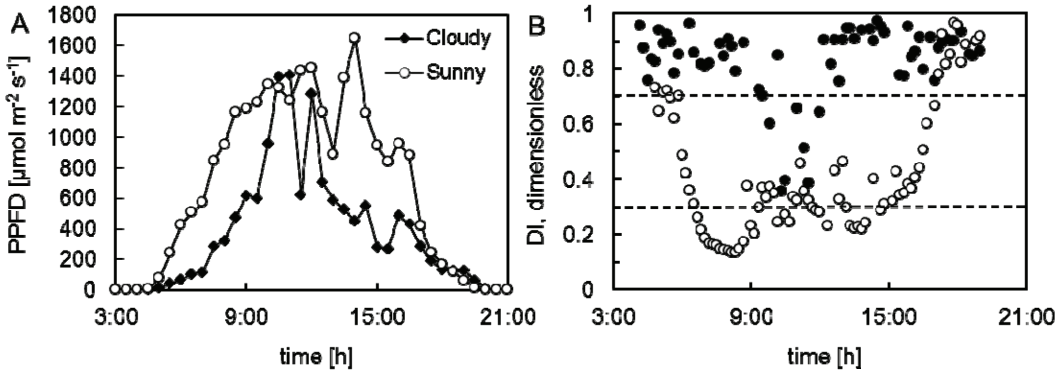
MATERIALS AND METHODS

Site description

The experiment was performed in the forest stand at the experimental research site Bílý Kříž (Beskydy Mountains, 49°33'N, 18°32'E, NE, of the Czech Republic, 908 m a.s.l.). The area is characterized as cool (annual mean temperature 5.5 °C) and humid (annual mean relative air humidity 80%) with high precipitation (annual amount 1 000–1 400 mm). The geological bedrock is formed by Mesozoic Godula sandstone (flysch type), with ferric podzols. The forest stand was planted in 1981 with 4-year old seedlings of *Picea abies* (L.) Karst (99%) and *Abies alba* Mill. (1%) on the slope (11–16°) with SSW orientation over an area of 6.2 ha. In July 2008, the stand density was 1430 trees per ha and a hemi-surface leaf area index 9.5 m^2 m^{-2} (± 0.27; standard error of LAI estimation). The mean tree height was 13.4 m (± 0.1m) and the stem diameter at 1.3 m was 15.8 ± 0.2 cm.

Basic description of the experiment

The experiment took place on July 16th and 30th during the vegetation season of 2008. These two days were characterized by distinct atmospheric conditions (Fig. 1). The differences in sky conditions during these two days tended to change other



1: *General changes in (A) photosynthetic photon flux density (PPFD) and (B) diffusion index (diffuse/total irradiation ratio) at study site during the cloudy and sunny day the measurements proceeded on*

microclimatic parameters as well. The average microclimatic conditions over the three days preceding the measurements were similar to those during the measurement.

One-year old needles of the 4th, 7th, 9th and 12th whorls with S/SW orientation within one spruce crown were examined. The measurements took place four times a day, at 5:00 am, 8:00 am, at noon and at 17:00 pm. The days on which the measurements were carried out were neither typically sunny nor cloudy. Gas exchange measurements and sample collections were performed during cloudy periods characterized by DI (diffuse/total irradiation ratio) > 0.7 on cloudy day and during sunny periods characterized by DI < 0.3 on sunny day (Fig. 1B).

After the gas-exchange measurement, the twigs were removed from the tree and the needles ran through another analyses. Part of them immediately underwent optical property measurements to prevent a degradation of the examined leaf tissue that might occur in case of storage. The needles that were assigned for the pigment analysis were removed from the branchlet, weighted, their projective area was scanned, and subsequently they were illuminated for ten minutes at the irradiation recorded during the photosynthesis measurement to readapt them to those light conditions they were exposed to while on the tree. Light adapted needles were stored in liquid nitrogen until the pigment analysis took place.

Gas-exchange measurements

An open, portable gasometrical system with the infra-red gas analyser Li-6400 (Li-Cor, Nebrasca, USA) was used for in-situ measurement of the relationships between the incident photosynthetic photon flux density (PPFD) and CO_2 assimilation rate. The actual assimilation rate (A_N) and stomatal conductivity (Gs) were recorded under conditions of natural irradiance and water pressure deficit (VPD). CO_2 concentration inside the assimilation chamber remained constant: ambient 385 ± 5 μmol (CO_2) mol^{-1}. During the gas-exchange measurements shoots were positioned as if under natural conditions in the field. The actual irradiation to which the needles were exposed during each measurement was recorded via a sensor placed inside the assimilation chamber. This light sensor usually faced the sun.

Pigment analysis

The contents of total chlorophylls (Chl $a + b$), total carotenoids (Car $x + c$), and Chl a/b and Chl $a + b$/Car $x + c$ ratios were estimated spectrophotometrically (UV/VIS 550, Unicam, England) from the supernatant obtained after the centrifugation (for 3 min at 480 g) of pigment extracts in 80% acetone with a small amount of $MgCO_3$. The needle pigment content was determined according to Lichtenthaler's equations (Lichtenthaler, 1987). Chl $a + b$ and Car $x + c$ contents were expressed per needle projection area that was estimated using a Mustek BearPaw 2448TA Pro scanner and Cernota software (Kalina and Slovák, 2004).

The content of xanthophyll cycle pigments, i.e. A, antheraxanthin; V, violaxanthin; Z, zeaxanthin, was estimated by means of gradient reversed-phase HPLC (Kurasova *et al.*, 2003). The conversion factors for the contents of the individual carotenoids (the pool of xanthophyll cycle pigments, i.e. V + A + Z) were applied according to Färber and Jahns (1998). The conversion state of the xanthophyll cycle pigments (de-epoxidation state; DEPS) was calculated as DEPS = [A + Z]/[V + A + Z] (Gilmore and Björkman, 1994).

Optical property measurements

The original Daughtry method (Daughtry *et al.*, 1989) adapted by Malenovský *et al.* (2006) was used to measure optical properties of the Norway spruce needles. We used the ASD FieldSpec-3 spectroradiometer (ASD Inc., Colorado, USA) coupled with an integrating sphere LI-1800-12 (Li-Cor Inc., Lincoln, NE, USA) to measure spruce needle hemispherical reflectance between 400–1 650 nm with a wavelength interval of 1 nm. The detailed description of the method can be found in Malenovský *et al.* (2006).

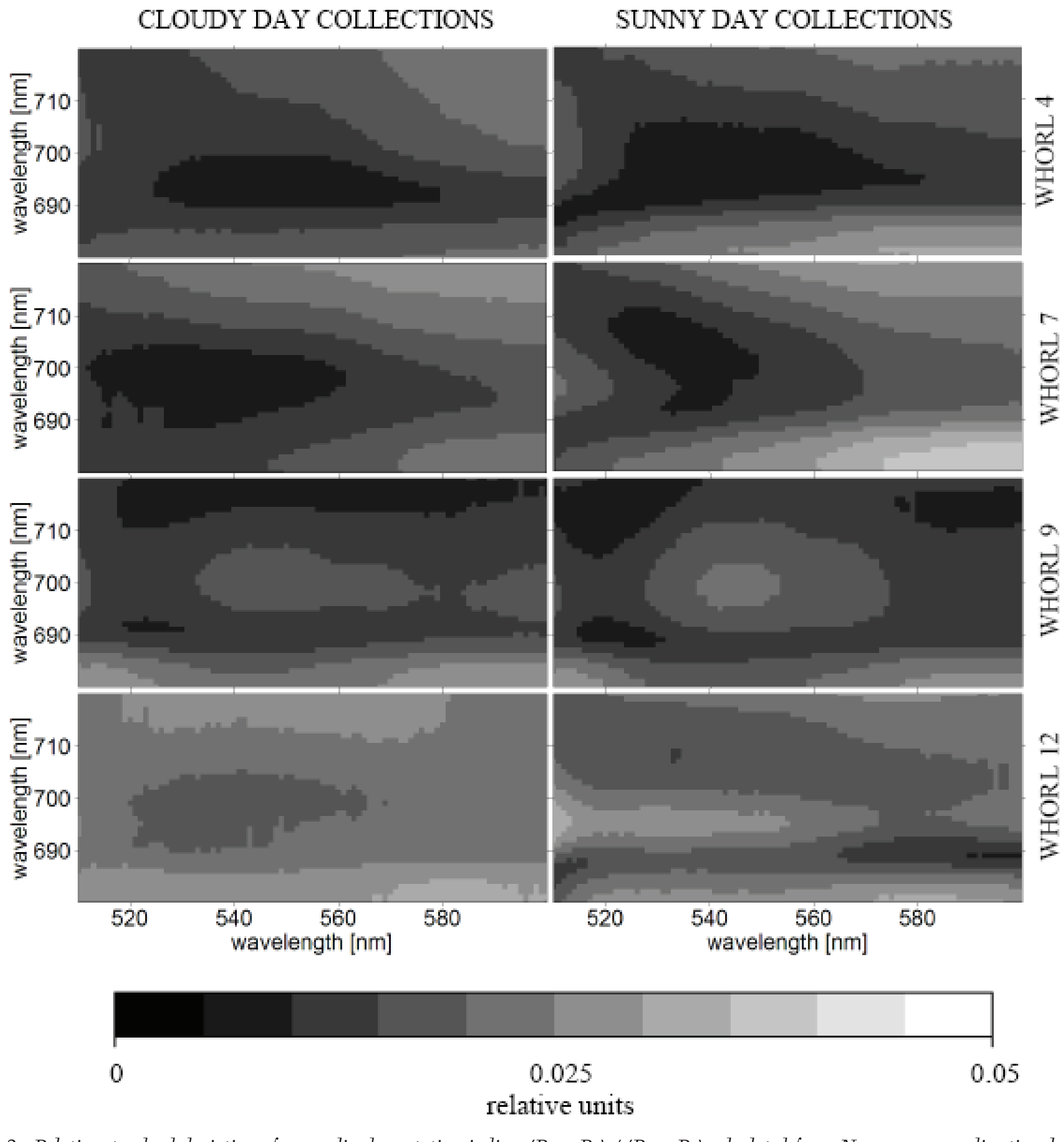
The hemispherical reflectance of the sample needles was computed according to the following equation:

$$R = \frac{R_{TOTAL}/R_{REF}}{1 - GF},$$

where R_{TOTAL} is the flux of radiation reflected from the sample in reflectance mode, R_{REF} is the flux of radiation reflected from a $BaSO_4$ reference standard in reference mode, GF is the gap fraction of the sample in reflectance mode. To compute the gap fraction between illuminated needles, needles inside the carrier were scanned using a double lamp and determination of the gap fraction was made by way of image processing, when the number of pixels of the total gap area between the sunlit needles was divided by the number of pixels of the total measured (illuminated) area. Our gap fraction for measurements in reflectance mode was on average 0.268 ± 0.05 on 16. 7. 2008 and 0.265 ± 0.05 on 30. 7. 2008.

Data analysis

To find the optical indicator characterizing particular types of needles, sunlit (4th and 7th whorl) needles are taken into account primarily. As they



2: *Relative standard deviation of normalized vegetation indices (R_x – R_y) / (R_x + R_y) calculated from Norway spruce directional-hemispherical reflectance. Each value represents a variation of 12 values calculated from the dataset obtained from each crown level throughout the day. The deviation for normalized ratios from the 4th whorl needles in the 1st row, 7th whorl needles in the 2nd row, 9th whorl needles in the 3rd row, and 12th whorl needles in the 4th row. The left column shows the dataset measured on a cloudy day, right column displays the dataset measured on a sunny day.*

are exposed to similar microclimate conditions during midday, when the stand is exposed to the highest portion of direct irradiation on either sunny or cloudy days, they are also expected to have a similar photosynthetic activity in a wider spatial scale. Thereby objective is to find optical indicator characteristic for needles in wider spatial scale within top of the tree crown. Transitional and shade needles on the other hand, are usually exposed to sunfleck occurrence even during midday. Vegetation index characterising needle type the most accurately, was chosen according to the analysis of relative standard deviations in normalized indices in needles belonging to diverse cathegories.

The determination coefficient (R^2) was computed to express the variation percentage of a dependent variable explained by an established regression to the independent variable. The significance of the statistical model was tested at probability levels $P < 0.1$, $P < 0.05$ and $P < 0.01$, using the analysis of variance (ANOVA).

Statistical differences between the means of leaf characteristics were determined using a two-sample F-test for variances, followed by a Student's t-test at level of significance $P < 0.05$. Based on the results of the F-test, the t-test, assuming either equal or unequal variances, was used.

All computations and tests were conducted in the statistical environment R (R Development Core Team, 2010).

RESULTS

Among all normalized indices computed from the reflectance spectra of the 4^{th} and 7^{th} whorl needles, that were referred to as sunlit, the ratio of reflectance at around 695 nm and reflectance between 525–560 nm reveal the lowest standard deviation (less than 1 % of parameter value) (Fig. 2). Throughout the crown the relative standard deviation in parameter value is increasing, being the biggest in the 12th whorl needles (over 3 % of parameter value).

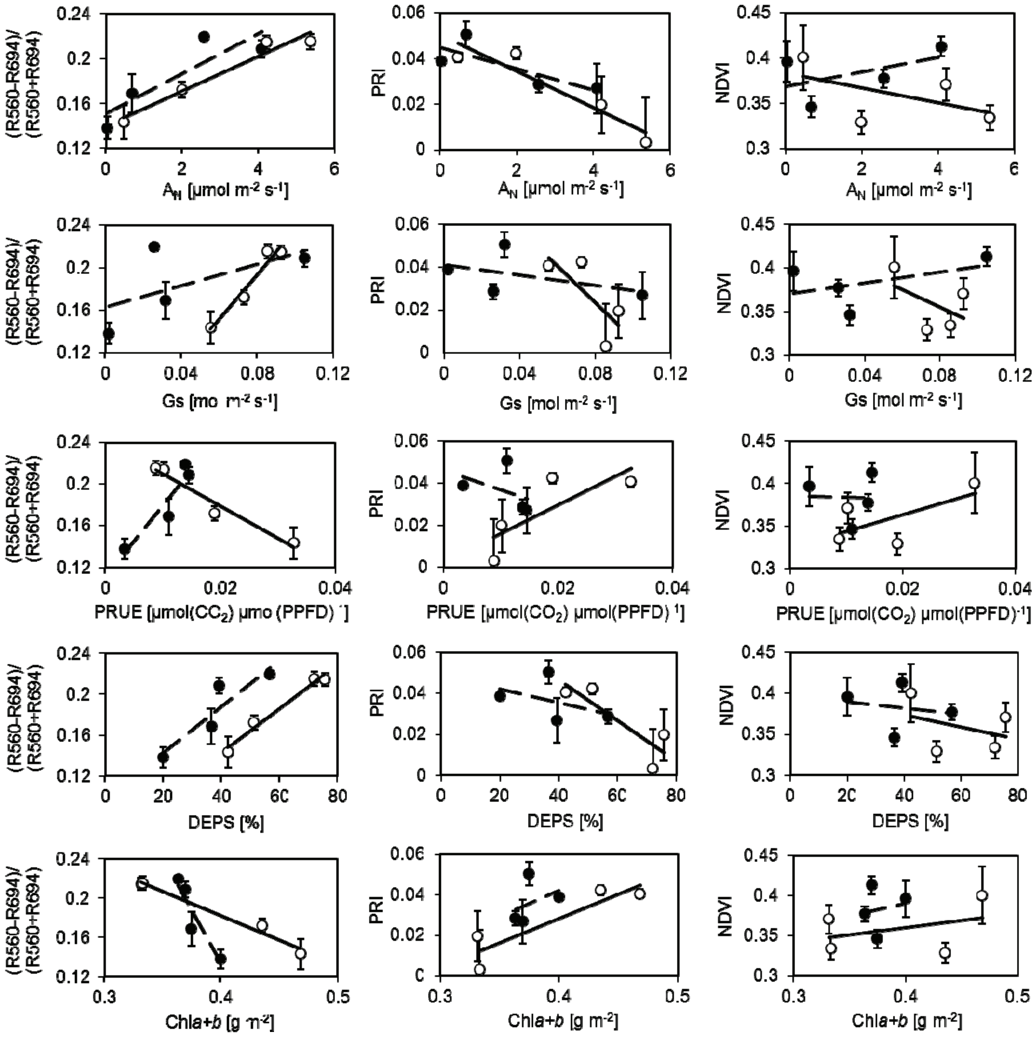
In case we intend to have a vegetation index characterizing the status of the vegetation, we suggest to use a ratio with reflectance at higher wavelengths, since the reflectance at 531 and also at higher wavebands is affected by the activity of the xanthophyll cycle and other protective processess. At conopy level, reflectance at 531 nm might vary by 6 % as a consequence of photoprotective processes actions (Hall *et al.*, 2008). The extent of the relative difference also depends on the capacity of the plant for thermal dissipation of excess light energy through the xanthophyll cycle (Peñuelas *et al.*, 1995), i.e. VAZ pool size. Adjacent wavebands above 531 nm are said to be affected by light scattering associated with the build-up of the thylakoid pH gradient (Gamon *et al.*, 1997; Nichol *et al.*, 2002). The denotation of these processes is the most pronounced up to 557 nm in Norway spruce, according to an independent analyses (yet unpublished data). Thus we have been using the R694 and R560 ratio in this study. Waveband at 560 nm has been also originally proposed as referential in PRI by Gamon *et al.* (1992) as this waveband is the least influenced by chlorophyll content.

The vertical graditionality of the normalized R694/R560 vegetation index of needles belonging to the south-oriented branches of spruce crown reflects intensity of their physiological processes during noon (Fig. 3, Tab. I) on sunny and cloudy days. Our 96-spectra database supports this conclusion, since the R694/R560 value of 9^{th} whorl needles never gets close to that of 7^{th} whorl needles ($P < 0.001$). The 12^{th} whorl needle index never reaches the parameter values of the 9^{th} whorl needles ($P < 0.001$). Reflectance in red region serves as reference, differentation is determined by green reflectance, i.e. green peak height.

For the photosynthesis rate, the stomatal conductance and even for DEPS reading via the red/green ratio, the slope of the line characterizing the mutual relationship of the variables is positive. The slope for the photosynthetic radiation use efficiency (PRUE)-R694/R560 index reverts under clear sky conditions, since under high light incidence PRUE in sunlit needles is lower during noon as a result of the LUE computation as A_N/PAR ratio. Graphs demonstrating the relationship between physiological characteristics and conventionally used indices PRI and NDVI computed from the needles' directional-hemispherical reflectance do not exhibit any significant correlation (Fig. 3). The determination coefficient for the relationship between R694/R560 and the intensity of photosynthesis-related processes recorded at time periods around noon is poor as well (not shown). This is not surprising, because even in sunlit parts of the crown SSW-oriented needles are shaded by branches obstructing the sunlight even in the morning and afternoon. This is also the case for the whole crown in case the needles belonging to branches in lower positions. The incoming radiation is determining for photosynthetic activity. At noon there should be a cleaner vertical gradient of sun penetration throughout the southern portion of the canopy, as a result of increased sunlight penetration into this part of canopy with regards to the position of the sun.

DISCUSSION

The exact red/green normalized vegetation index measured via a spectroradiometer attached to the tower has been found most suitable for tracking seasonal variations in the photosynthetic activity of rice fields (Inoue *et al.*, 2008). Drolet *et al.* (2005) found that the MODIS band 11 (centered at PRI's 531 nm) normalized to band 13 (centered at 667 nm) was more suitable for LUE prediction of forest stands in North American boreal forests compared to conventionally computed PRI using band 12 (centered at 557 nm). Drolet suggested that the better

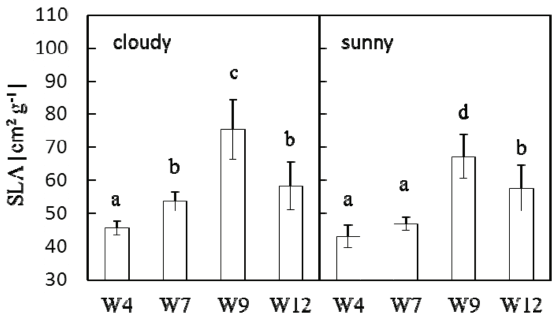


3: *Improvement over PRI and NDVI when using (R560 – R694) / (R560 + R694) for reading the intensity of physiological processes throughout the crown. In the bottom of the picture, parameter dependency on needle chlorophyll content is shown. Dependencies are shown for midday values of the photosynthesis rate (A_N), stomatal conductance (Gs), light use efficiency (PRUE) and DEPS. Each (R560 – R694) / (R560 + R694), PRI and NDVI value is an average of three reflectance measurements performed at the time of evaluation. Bright dots represent measurements on sunny days, dark dots show measurement on cloudy days.*

I: *Determination coefficients (R^2) found for the linear relationships between (R560 – R694) / (R560 + R694), PRI, NDVI and physiological parameters of interest measured on leaf level on sunny and cloudy day during the noon. Asterisks indicate level of statistical significance: *P < 0.1; **P < 0.05; ***P < 0.01. Abbreviations: A_N, assimilation rate under conditions of ambient irradiation, VPD etc.; Gs, stomatal conductance; PRUE, leaf level photosynthetic radiation use efficiency; DEPS, deepoxidation state of xantophyll cycle pigments.*

		AN	Gs	PRUE	DEPS	Chl*a*+*b*
R560/R694 ND	sunny	0.96**	0.95**	0.96**	0.98***	0.98***
	cloudy	0.77	0.36	0.88*	0.84*	0.88*
PRI	sunny	0.87*	0.54	0.64	0.73	0.81*
	cloudy	0.63	0.24	0.19	0.2	0.15
NDVI	sunny	0.29	0.24	0.42	0.13	0.03
	cloudy	0.27	0.23	0.01	0.04	0.13

correlation was a result of reduction in canopy chlorophyll content causing decrease in band 13 reflectance. Even though the position of MODIS band 13 isn't consistent with our reference with the position at 694 nm, there are certain commonalities in both results. MODIS band centered at 667 nm is covering the area of the maximal absorption feature of Chl *a* (670 nm). Therefore does not



4: *Mean values ± standard deviation of the morphometric parameter specific leaf area (SLA) of Norway Spruce needles in the level of 4th, 7th, 9th and 12th whorl (W4, W7, W9, W12 counted from the apex). The same letters indicate no statistical difference at P < 0.05, n = 12.*

necessarily have to be affected a lot by variations in the chlorophyll content. Our reference at 694 nm behaves in a similar way. More important for the relationship are differences in the green region. Possible dynamic in band 11 might have also affected relationship.

Our result is thus consistent with findings of other authors; nonetheless, it is difficult to generalize dependencies established between reflectance parameters and variables discovered using integrating sphere to remote observations. The canopy radiance is affected by the solar angle, incidence irradiance, energy transfer, canopy structure, leaf movement, soil reflectance, atmospheric effects and other factors. The exact background behind the robustness of the parameter has never been explained properly. Our results suggest that the ratio is insensitive to variations in the chlorophyll content, which varies quite significantly among the needles examined. Over a period of 14 days of measurements, the chlorophyll content changed by 0.05 g m^{-2} on average in sunlit needles ($P < 0.05$), whereas R694 and the green reflectance ratio remained constant. The mutual effect of leaf chlorophyll content and morphology, i.e. the amount of intercellular spaces seem to play a key role. These are properties commonly characterizing a leaf's optical properties in designated areas according to the widely used PROSPECT model (Jacquemoud and Baret, 1990; Malenovský *et al.*, 2006). Leaf morphology issue is partially resolved with specific leaf area (SLA) estimetes. SLA provides information about optical thickness of observed plant material and thereby can be also used to assess leaf structural parameter (Jacquemoud and Baret, 1990). Also SLA is apparently not at stable level in sunlit needles, being signifficantly higher in 7th whorl needles during cloudy period ($P < 0.05$). Structural and pigment attributes have a more pronounced and unpredictable effect on the PRI and NDVI values, lowering the performance of both spectral indices.

The ratio cannot be considered as an indicator of an absolute physiological signal. Photosynthesis is a process driven by the availability of nitrogen and water, whereas the optical properties of leaves respond to changes in the pigment content and morphology. The photosynthesis rate and related gas-exchange parameters assessed in sunlit needles at noon under clear sky conditions must be taken as orientational and does not necessarily have to be considered absolute. Insufficient soil moisture and a high vapour potential deficit may lead to a decrease of stomatal conductance to CO_2 diffusion followed by decreases of CO_2 concentrations at both the intercellular and chloroplast levels, causing the CO_2 uptake to decline (Urban *et al.*, 2007), particularly in sunlit needles. A mechanism causing the midday depression of photosynthesis has not yet been discovered. In our case, the midday depression of photosynthesis (A_N decreased from approximately 6–7 µmol(CO_2) m^{-2} s^{-1} at 10:00 to 4–5 µmol(CO_2) m^{-2} s^{-1} during midday) in sunlit needles on a sunny day was a result of stomata closure. Hence, red/green reflectance ratio should be rather concidered as indicator of long-term actions.

CONCLUSION

We tried to evaluate the performance of normalized vegetation indices as static physiological indicators. Among all possible variations, the ratio of reflectance in the red and green region came up as the best solution for judging the physiology throughout the Spruce crown. However, the distribution of the needles with the R560/R694 normalized index throughout the crown seems to be a consequence of long-term acclimation to light conditions. Since our finding is consistent with the discoveries of other authors who investigated stress response in reflectance, the result does not seem to be coincidental and is worth further investigations. Additionally, not all factors affecting the parameter value have been completely discerned. As needles are heterogenous in chlorophyll content, this factor seems to have the major effect on the value of parameter. In the green reflectance of canopies, more pronounced is canopy architecture than structure of individual leaves. A factor that hasn't been paid attention to very much yet, however many extraneous effects are affecting green reflectance of canopies. Therefore, attention should be paid to performance of the parameter to track ecophysiological variables of whole stands. Employing remote sensing techniques for larger plants might reveal a potential applicability of the spectral index for airborne/spaceborne imaging spectroscopy observations. Deeper studies should consider also angular distribution of parameter value.

SUMMARY

In this study we investigated empirical methods for rapid assessment of plant physiological activity via reflectance. Study is evaluating interaction between photons and plant materiál and is conducted at leaf level. The leaf reflectance of the Norway spruce needles sampled was measured using integrating sphere according to the method of Daughtry modified by Malenovský et al. Obtained experimental database of reflectance spectra was examined in order to identify vegetation index best suited for characterising particular needle types, i.e. sunny, transitional and shaded needles. Such parameter should also to certain extent describe physiological activity.

All possible types of normalized vegetation indices were computed from spectra measured with 1 nm resolution, and correlated with gas-exchange characteristics and needle photosynthetic pigment content in spatial scale. Our result demonstrate the potential to spectrally detect state of foliage with other means than conventional PRI and NDVI index. We document significant relationship between red/green reflectance ratio and physiological activity in cross crown during the noon. Due to well known dynamic in green reflectance, that is connected with photoprotective actions, index is presented as the ratio of reflectance at 560 nm and 694 nm. Additionaly, being always the highest in the uppermost part of the crown and the lowest in shaded part of the crown, parameter is strictly stratificated throughout the crown. However, we failed to determine the essence of the parameter. We were not able to identify, whether parameter value is primarily given by needle chlorophyll content or morphology.

Result is consistent with findings of other authors employing remote observations of plants, thus providing insight into problematic, that has not been studied well, yet. Study also suggests potential to estimate physiology with this indicator, but prior to serious application, principle of the parameter from geoinformatical point of view must be investigated. Remote sensing applications would pay attention to angular distribution of the parameter value and should also consider and properly correct extraneous effects on reflectance signal, e.g. state of atmosphere.

Acknowledgement

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