



Optical detection of pigment contents in leaves: intercomparison of several handheld devices

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Abstract

Handheld devices have become popular to determine epidermal contents of UV-A absorbing compounds in intact leaves. For the comparability of the results between different labs, it is important that all instruments show the same or at least similar results. Therefore, the signals from seven different Dualex Scientific optical leaf clips from five laboratories were compared to each other, alongside a prototype of an LSA-2050 leaf state analyzer and a UV-A-PAM fluorometer. The values of the Flav index, which is used to quantify epidermal UV-A absorbing compounds such as flavonoids, corresponded well with each other ($r^2 > 0.989$), except for one instrument. A higher variability among instruments was found for the chlorophyll (Chl) index, again with one instrument showing a significant offset. Although one Dualex instrument showed an excellent linear calibration for the anthocyanin (Anth) index relative to extract concentrations when red hazelnut leaves were investigated ($r^2 = 0.969$), the Dualex instruments had all an offset against each other and at Chl index values below 30, erroneous Anth index values were observed. While Flav index results appeared quite reliable, it is recommended to calibrate a Dualex instrument against leaf extracts for exact determination of chlorophyll and anthocyanin contents. Simple tests for the functionality of a Dualex instrument are suggested.

Keywords Epidermal UV transmittance · Dualex · Chlorophyll fluorescence · Anthocyanins

1 Introduction

Epidermal UV screening by the accumulation of compounds such as flavonoids and other polyphenolics is an important and widespread mechanism of plants for protection against the detrimental effects of UV radiation. The formation of flavonoids is induced by several different environmental factors, among them high photosynthetically active radiation (PAR), (low) temperature or nitrogen deficiency (for a

review, see [1]). To follow this dynamic and highly variable response, especially under natural conditions, the repeated assessment of a large quantity of leaves is required. For determination of epidermal UV screening, a non-destructive method based on measuring chlorophyll fluorescence was developed by Bilger et al. [2]. The basic principle of the measuring technique relies on the fact that chlorophyll fluorescence can be excited by a UV-A excitation beam as well as by a red beam. While the UV-A beam can be absorbed by other compounds absorbing in this wavelength range, this is not the case for the red beam, which, hence, serves as a reference. The ratio between the two fluorescence signals is related to the amount of all compounds which absorb UV-A radiation and optically interfere with the UV-A excitation beam. In later years, this originally laboratory-based method was transferred to portable devices [3–8], allowing the rapid testing of leaves under field conditions. Due to its ease of use, the Dualex Scientific instrument (formerly manufactured by ForceA, Orsay, France, currently by Pessl Instruments, Weiz, Austria) has become a popular device for determining epidermal UV-A absorbance via the so-called Flav index. Besides characterising the acclimation

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of leaves to UV radiation, the Dualex instrument has also been introduced as an indicator of the demand for nitrogen fertilization in crop plants via the so-called nitrogen balance index (NBI), which is the quotient of the Flav index and the simultaneously determined chlorophyll content characterized by the Chl index [7, 9]. The rationale for this method is based on the fact that N deficiency leads to a reduction of chlorophyll and at the same time to an increase in flavonoid and phenylpropanoid formation [10]. The Dualex scientific instrument has been widely used, e.g., to study the effects of UV-B exposure to crop plants [11], the influence of natural light exposure on understorey plants [12], the annual time course of the Flav index in a *Vaccinium vitis-idaea* canopy [13] or as a tool to assess crop nitrogen status [14].

Given the wide application of the Dualex instrument, it is desirable to compare results among different investigations, i.e. among different instruments. However, no external reference standard is existing to test for proper functioning of the instrument. Instead, it is necessary to send the instrument to the manufacturer for renewed calibration. Therefore, we used the opportunity of a meeting of The International Association for Plant UV Research (“UV4Plants”) at Göttingen in March 2024 to arrange an intercomparison of four Dualex 4 scientific and one Dualex 3. Since at Kiel university two further Dualex 4 instruments were available, these were also included in the overall comparison. In addition, two additional optical devices were included in the study, the established UV-A-PAM fluorometer [3, 5] and a prototype of a recently developed instrument, the LSA-2050 leaf state analyzer. While the primary aim was to assess the reliability and comparability among instruments of the Flav index, the two further indices determined by the Dualex 4 instrument, characterizing chlorophyll (Chl index) and anthocyanin (Anth index) content were also incorporated in the study. The measurement of the Anth index involves the same principle as that used for the Flav index, i.e. comparison of the fluorescence yields excited at two different wavelengths. Instead of using UV-A excitation, the measuring beam for anthocyanin measurement is centred around 530 nm [6, 15]. Anthocyanins are formed in some species in young or senescent leaves and can serve as photoprotectants [16]. The Chl index is determined using a comparison of the transmittance of a red beam absorbed by chlorophyll and a beam in the near infrared, where chlorophyll no longer absorbs. Within a certain range, the chlorophyll content of a leaf is related to its photosynthetic capacity. The Chl index is also used in the assessment of N demand (see above). As measuring objects for the intercomparison, a series of *Vicia faba* leaves was chosen, which had been grown at a broad range of irradiance inducing a strong variability in leaf optical properties. Since inconsistent results were obtained in the measurements of the Anth index, its reliability for the

detection of anthocyanins was assessed in a separate experiment using leaves of a red hazelnut variety.

2 Materials and methods

Plants of *Vicia faba* L. cv. ‘Fuego’ were grown at Kiel University in a growth chamber (Johnson Controls, Mannheim, Germany) at a range of irradiances. The irradiance of photosynthetically active radiation (PAR) at the leaf level was manipulated by growing the plants at different heights in the chamber, thereby varying the distance from the lamps (CMT360LS WBH EYE Iwasaki Electric Co., Japan) and accordingly the photon irradiance (400–700 nm), which ranged from 25 to 450 $\mu\text{mol m}^{-2} \text{s}^{-1}$. Environmental conditions in the chamber were 21 °C air temperature, 60% relative humidity and a day/night cycle of 16/8 h. Varying irradiance was used as a tool to effectively manipulate the production of phenolic compounds in leaves, including accumulation of flavonoids that contribute to epidermal UV shielding. These changes can be assessed using the Dualex instrument through variations in the so-called “Flav” index. The plants were brought to Göttingen one day before the conference and kept in a growth chamber at a photon irradiance of 400 $\mu\text{mol m}^{-2} \text{s}^{-1}$ and 20 °C. Right before the intercomparison, 20 leaf discs (diameter 2 cm) were punched from leaves of different plants and placed individually in a numbered Petri dish on filter paper moistened with tap water to secure the comparability of all measuring results.

On 21.03. 2024, within one hour, the participants did the series of measurements on these discs, always placing the Dualex instrument in the centre of each disc. In addition to the Dualex instruments, measurements were also done using a UV-A-PAM fluorometer (Gademann Instruments, Würzburg, Germany) in combination with a MINI-PAM-Fluorometer (Walz GmbH, Effeltrich, Germany). The latter served as a source for a red measuring beam, which is missing in the UV-A-PAM fluorometer. Data from this combination were normalized to the fluorescence values obtained from a blue plastic foil (Walz) after subtracting the values obtained with a non-fluorescing green PVC foil. As a reference for 100% transmittance, epidermis-free abaxial leaf sides of *V. faba* were used. From the obtained UV-A transmittance values, absorbance was calculated, which is equivalent to the Flav index provided by the Dualex instruments. For the latter instruments, their internal calibration provided by the manufacturer was used.

Different leaves from the same *V. faba* plants were used for an additional comparison among three Dualex instruments at Kiel, which were included in the overall data set.

Measurements were also done using a prototype of an LSA-2050 instrument (Walz GmbH) using reference

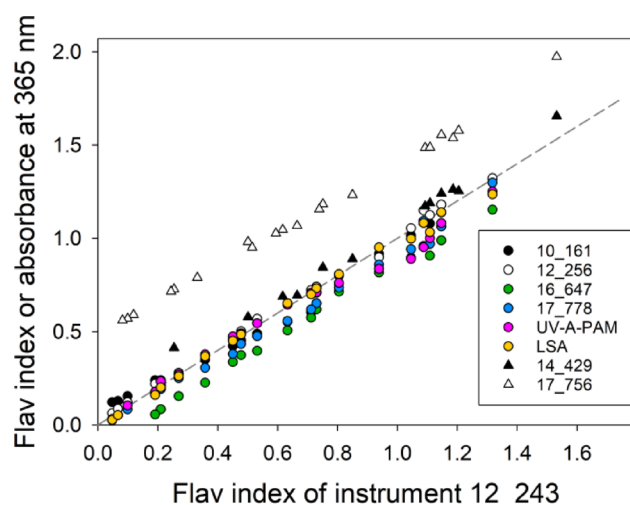


Fig. 1 Results from the measurements on 20 different leaf discs of *V. faba* grown at different irradiance as done with Dualex instruments identified by their serial numbers at Göttingen University (circles) and at Kiel (triangles). The data set includes also absorbance values determined by a UV-A-PAM fluorometer and an LSA-2050 leaf state analyzer. All data are related to the values obtained by the Dualex instrument with serial number 12_243. The dashed line represents identity between both axes

Table 1 Results of the regression analysis for the flavonoid indices (Flav) between the indicated instruments and instrument 12_243, as shown in Fig. 1

	slope a	std error a	intercept b	std error b	r ²
10_161	0.9370*	0.0227	0.0287	0.0165	0.9896
12_256	0.9974	0.0115	0.0158	0.0840	0.9976
14_429	0.9890	0.0307	0.0913*	0.0299	0.9904
16_647	0.9611*	0.0162	-0.1040*	0.0127	0.9958
17_756	0.9277*	0.0139	0.4787*	0.0111	0.9960
17_778	0.9520*	0.0191	-0.0243	0.0139	0.9928
UV-A-PAM	0.8981*	0.0223	0.0298	0.0162	0.9891
LSA-2050	0.9726	0.0162	0.0260*	0.0123	0.9956

Values for the slope and the y-axis intersection of the linear equation $y = ax + b$ are provided together with their respective standard errors. In addition, the regression coefficient r^2 is shown. * indicates a significant difference for the slope from 1.0 or for the intercept from 0.0, respectively, at an alpha error level of 5%

values from the abaxial side of a leaf from *Hylotelephium telephium* (L.) H. Ohba, provided in the instrument software. Further measurements were done at Kiel University on leaves from the same plants using two additional Dualex Scientific instruments.

In an additional experiment, leaves of a red-leaved variety of *Corylus avellana* L. cv. ‘Rotblättrige Zellernuss’ were collected in a local garden in Kiel. Discs with a diameter of 8 mm were punched from an area of the leaves with as little veins as possible using a cork borer. Dualex measurements were conducted from both leaf sides, followed by snap freezing the disks in an Eppendorf cap in liquid nitrogen. The caps contained 2 large (diameter 4 mm) and 3 smaller

(diameter 3 mm) glass beads (Roth GmbH, Karlsruhe, Germany). The discs were ground in a Retsch MM2 vibration mill (Retsch, Haan, Germany) for 4 minutes at 90% power after addition of 800 μ L cold methanol acidified with 1% concentrated HCl. After centrifugation in a cooled microcentrifuge (Fresco21, Thermo Fischer Scientific, Germany) at 11 600 x g, the supernatant was decanted and the pellet was resuspended in 350 μ L acidified methanol. The suspension was then homogenized on a thermomixer (Biometra TSC ThermoShaker, Analytik Jena, Germany) and this step was repeated after a further centrifugation. From the unified supernatants absorbance spectra from 750 to 300 nm were determined in a Cary 60 UV-VIS spectrophotometer (Agilent, Waldbronn, Germany). As a measure for anthocyanin content absorbance at 532 nm was used, after correction for the pheophytin content in the extract according to the formula:

$$(A_{532} - A_{750}) - 0.225 * (A_{653} - A_{750})$$

This formula was based on Mancinelli et al. [17] and Zirngibl et al. [18] and adjusted to the situation in the extracts of *C. avellana*. The absorption spectra of the anthocyanic extracts had relative absorbance maxima at 653 and 532 nm. The factor of 0.225 for the subtraction of pheophytin absorption at 532 nm was determined in leaf extracts from a green variety of *C. avellana*.

Regression analysis of the data was done using SigmaPlot 13 (Systat Software GmbH, Erkrath, Germany).

3 Results

The Flav values of the whole data set stretched over a wide range, from nearly 0 to over 1.2. Since the true value of epidermal UV-A absorbance is inherently unknown, data from all devices were plotted against the data of the Dualex instrument with the serial number 12_243 (Fig. 1). By choosing this device as reference it was possible to include a second data set on corresponding leaves of the same plants using two additional Dualex instruments. It is apparent that all devices, including also the two non-Dualex ones, were linearly correlated with high regression coefficients ($r^2 > 0.989$; $p < 0.001$) to device no. 12_243, and hence also to each other. The slopes were mostly close to 1, but only in three cases not significantly different from 1.0 (Table 1). The y-axis intercept was in four cases significantly different from zero, especially for 17_756, which was far off (Table 1). In fact, this observation had inspired the call for the intercomparison and was related to an apparent malfunction.

The chlorophyll measurements were compared among the devices as well. Following the previous procedure, one

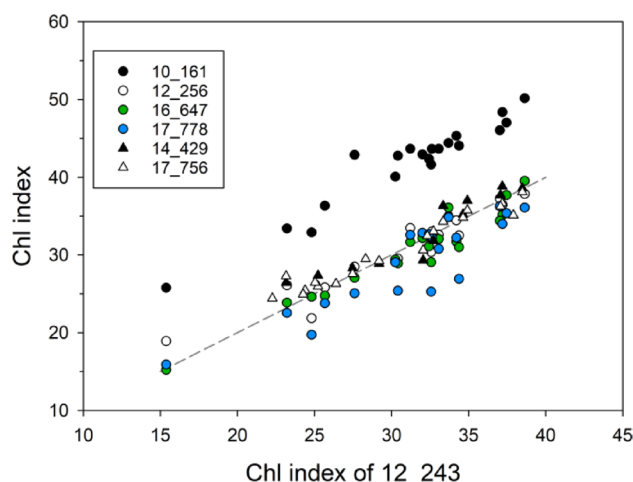


Fig. 2 The chlorophyll index (Chl) from six different devices as a function of the chlorophyll index determined with instrument 12_243. The dashed line indicates the function for identical indices

Table 2 Regression analysis results for the chlorophyll index (Chl) between the indicated instruments and instrument 12_243, as shown in Fig. 2

	slope a	std error a	intercept b	std error b	r ²
10_161	0.9932	0.0642	10.8774*	2.0321	0.930
12_256	0.8755*	0.0612	3.9488*	1.9372	0.919
14_429	0.9082	0.1064	3.8105	3.4544	0.8793
16_647	0.9445	0.0613	0.8969	1.9406	0.9296
17_756	0.8213*	0.0441	5.8128*	1.3574	0.9506
17_778	0.9294	0.1068	0.1724	3.3832	0.8079

Values for the slope and the y-axis intersection of the linear equation $y = ax + b$ are provided together with their respective standard errors. In addition, the regression coefficient r^2 is shown. * indicates a significant difference for the slope from 1.0 or for the intercept from 0.0, respectively, at an alpha error level of 5%

device was chosen as a reference for the other 6. It is apparent that 4 instruments produced similar values, whereas one consistently showed approx. 10 units higher values than the others (Fig. 2). The regression coefficients were clearly lower than those obtained for the Flav index (Tables 1 and 2). The LSA instrument is not included in this analysis since

at the time of the measurements, its chlorophyll indication was not yet finally calibrated.

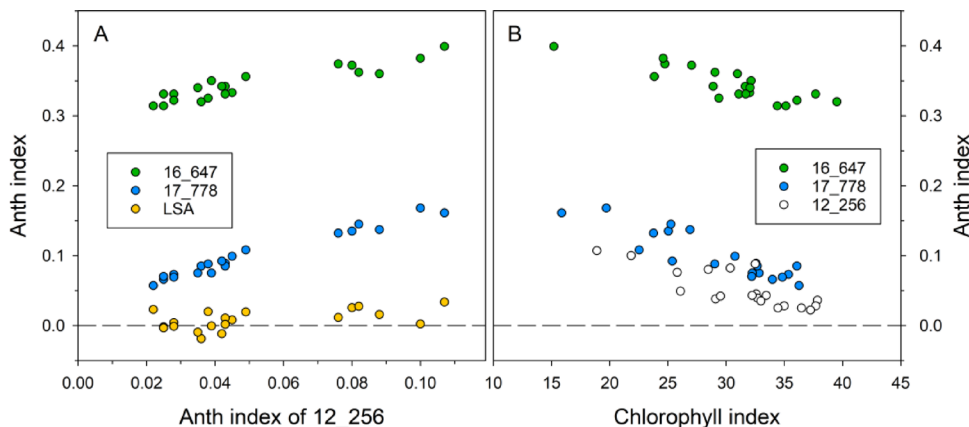
From Table 2 it can be concluded that three instruments had a significantly different offset in comparison to instrument 12_243, and two had a significantly lower slope, although at least for instrument 12_256, the data points were scattering along the line indicating equal values for both instruments considered.

In addition to the Flav index, most of the tested devices also determined an anthocyanin index (“Anth”) with the exception of instrument 10_161 and the UV-A-PAM fluorometer. While the Flav and related indices showed a satisfying correlation and uniformity, the same was not observed for the Anth index. Since the leaves were evenly green with no red colouration obviously visible to the human eye, it was already surprising that some instruments showed Anth values deviating from zero.

Figure 3A shows the relationships of the detected Anth values among the instruments, using this time instrument 12_256 as reference. Dualex 12_243 was excluded as it did not provide positive Anth values. For each single instrument, the Anth values ranged within an interval of about 0.1 units in the three Dualex instruments (Fig. 3B), whereas the corresponding index determined by the LSA-2050 displayed a smaller amplitude. However, the absolute levels of the detected Anth values varied strongly over a range of 0.4 units, whereas the slopes of the mutual relationships were similar for the Dualex instruments. The observed variation of the Anth values among the leaf samples was probably related to the different chlorophyll content of the samples. With decreasing chlorophyll contents, Anth values increased (Fig. 3B).

The variable performance of the Anth index of the Dualex instruments sparked an additional experiment to assess its general usefulness for anthocyanin detection, which was conducted with the Dualex device 12_243. The hazel leaves used for this experiment varied strongly in their red colouration, with the youngest leaves showing the highest anthocyanin contents and the lowest chlorophyll index values. The

Fig. 3 Anth index as determined by three different instruments, respectively, as a function of the Anth index determined by Dualex instrument 12_256 (A) or as a function of the chlorophyll index determined with the same instrument (B)



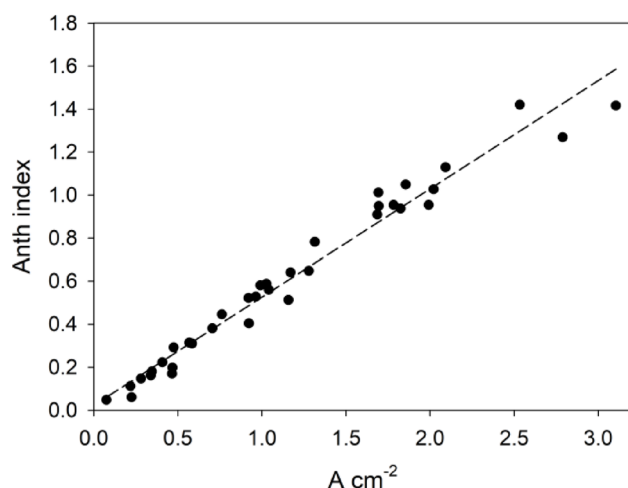


Fig. 4 Relationship of the sum of the Anth indices from the adaxial and abaxial sides of leaf discs of differently red leaves from *Corylus avellana* cv. ‘Rotblättrige Zellernuss’ and the anthocyanin absorbance at 532 nm of methanolic extracts from the same discs. The dashed line shows the regression line, $r^2=0.969$; $p<0.001$

latter index ranged from 20 to 42 units. Comparison of the Anth index, summarized over both leaf sides, and the absorbance of extracts from whole leaves showed a straight linear and proportional relationship, with a high correlation coefficient and a y-axis intercept not statistically different from 0 (Fig. 4). Leaves with data points close to the origin still had a red hue visible to the naked eye, and when two seemingly green leaves from a non-anthocyanic variety were measured, the Dualex instrument showed ‘NAN’, indicating an index value of zero or in the negative range.

4 Discussion

In general, the Dualex instruments showed a good or at least reasonable mutual agreement of the determined Flav values with one exception (17_756). This instrument was subsequently sent to the manufacturer (Pessl Instruments) for repair. Although the two other types of instruments (UV-A-PAM fluorometer and LSA-2050) had a different technical design, their data agreed well with those of the Dualex instruments (Fig. 1). The non-Dualex instruments allow users to set the reference value for 100% transmittance using measurements on epidermis free leaves or chloroplast suspensions placed on filter paper [4]. This feature enables independent calibration against external standards. Although calibration of the Dualex instruments must be done at the factory, the usage of epidermis-free leaves allows everybody to check the quality of the instrument. From easily available leaves of lamb’s lettuce (*Valerianella locusta* (L.) Laterr.) or spinach (*Spinacia oleracea* L.) and, as in the present study, from *V. faba*, the epidermis of the abaxial leaf

side can be easily removed on sufficiently large areas such that a test of zero screening is possible. Strong positive, but no negative deviations of the signal can be detected with this approach. The latter deviations can be assumed when even the intact abaxial sides of leaves of these species or from other shade grown plants show no numbers (‘NAN’). The satisfying agreement among the various instruments indicates that published Flav index data [e.g., 11–14] are generally quantitatively comparable.

In contrast to the Flav indices, chlorophyll indices showed a larger variability among instruments (Fig. 2; Table 2). Part of this variability may have been caused by the presence of veins in the leaf discs, which can negatively affect the reproducibility of measurements. We recommend using several measurements per leaf to account for this potential variability. A simple test for a large offset of the Chl index can be performed using fully senesced leaves, which should contain only traces of chlorophyll. Since chlorophyll content is relatively easily accessible using standard protocols [19], every user should calibrate his own instrument for quantitative chlorophyll determination. Within a certain range, the relationship between the optical index and true chlorophyll content may be species specific. It has also been recommended to determine specific calibration coefficients for monocots and dicots [7].

A satisfying agreement among the various instruments was not observed with the Anth index. In principle, two different problems were detected. First, different instruments had an offset of the values against each other (Fig. 3A), varying from close to or below zero (indicated by “NAN” by the instrument) up to 0.4. This shift is presumably caused by varying baseline reference values among the different instruments.

Second, the different instruments showed a similar dependency of the Anth index over the samples, apparently indicating consistently a slight increase in anthocyanin content along an unknown gradient over the leaves. However, in *V. faba* grown under benign conditions, we have never observed red colouration indicating anthocyanin accumulation and also the leaves investigated here were obviously green. A closer inspection of the data showed that the Anth values increased with decreasing chlorophyll content (Fig. 3B). Also, Nichelmann and Bilger [15] showed increasing F_{545}/F_{650} fluorescence excitation ratios with decreasing chlorophyll content of the sample, when suspensions of isolated chloroplasts were applied in varying concentration to filter paper. This behaviour can be explained by the fact that the measuring wavelengths are lying in regions of highly different extinction coefficients of chlorophyll. While the absorption of the red reference beam is still saturating even in the presence of reduced amounts of chlorophyll, the absorption in the green region, where the measuring

beam for anthocyanins is located, begins to decline with decreasing chlorophyll concentration. The varying absorption ratio between these wavelength regions has previously been named as sieve or package effect [20]. It leads with declining chlorophyll content to a reduced F_{530}/F_{650} ratio, which is interpreted by the instruments as anthocyanin signal. Therefore, Anth values indicated by a Dualex instrument on visibly green leaves with a chlorophyll index below ca. 30 units should be considered with utmost caution. On the other hand, this does not mean that the Dualex instrument is not well suited to detect anthocyanin in an obviously red coloured leaf. The excellent linear relationship between the anthocyanin content and Anth values in red leaves of *C. avellana* cv. ‘Rotblättrige Zellernuss’ up to Anth values above 1.0 (Fig. 4) proves the suitability of the instrument. This was observed despite Chl index values down to 20 in the leaves with the highest anthocyanin contents (data not shown). Since anthocyanin absorption *in vivo* is dependent on the pH value and other effects, such as complex formation and co-pigmentation with phenolic acids [21], the relationship between Anth index and anthocyanin content should be calibrated for the species used in the investigation.

This study profited from the parallel usage of several devices at identical objects. But how can a user who has only a single device available routinely check its quality? The results showed that the most critical difference among the devices was an offset in the signal. This is presumably due to a different reference value for the signal in the absence of pigments. In order to regularly check for this problem, we recommend a test of the signal in the absence of the pigments. This can be comparatively easily done with the visible pigments. Leaves with low chlorophyll content can be obtained by letting leaves senesce in darkness. Leaves without anthocyanins can be found everywhere. In contrast, leaves without UV-A screening compounds are very rare. In this case, epidermis-free shade grown leaves should be used as recommended above. Such tests can help to detect developing signal deviations. Should they occur, a calibration at the manufacturer is recommended.

5 Conclusions

With few exceptions, the instruments used in this study showed quite well comparable values, especially for the Flav index. This supports the role of this instrument as a workhorse in research characterizing leaf acclimation to various environmental conditions, among them nitrogen fertilization, high irradiance, low temperature and exposure to UV radiation, allowing a rapid orientation with large amounts of samples. Simple tests for instrument malfunctioning are

described. When determining chlorophyll contents, repeated measurements on a leaf are recommended.

Measurements of the Anth index should always be carefully evaluated and artefacts caused by too low chlorophyll contents (Chl index < 30) be taken into consideration. In our hands, visual inspection of the leaves for red colouration coincided well with Anth values when anthocyanins were indeed present. If there exists any doubt, the presence of anthocyanins should be verified by spectrophotometry on leaf extracts.

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Author contributions Wolfgang Bilger: Conceptualization, methodology, data analysis, writing – original draft. Gyula Czégény, Susanne Neugart, Jakub Nezval, Erhard Pfündel, Adriana Volná: Investigation, data analysis, writing – review and editing.

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Data availability The raw data used to generate the figures are available from the corresponding author upon due request.

Declarations

Conflict of interest E. Pfündel is employed by Walz GmbH, Effeltrich, Germany, the manufacturer of the LSA-2050 leaf state analyzer. All other authors declare they have no financial interests.

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References

1. Pescheck, F., & Bilger, W. (2020). UV-B resistance of plants grown in natural conditions. *Annual Plant Reviews*, 3, 337–398.
2. Bilger, W., Veit, M., Schreiber, L., & Schreiber, U. (1997). Measurement of leaf epidermal transmittance of UV radiation by chlorophyll fluorescence. *Physiologia Plantarum*, 101, 754–763.

3. Bilger, W., Johnsen, T., & Schreiber, U. (2001). UV-excited chlorophyll fluorescence as a tool for the assessment of UV-protection by the epidermis of plants. *Journal of Experimental Botany*, *52*, 2007–2014.
4. Goulas, Y., Cerovic, Z., Cartelat, A., & Moya, I. (2004). Dualex: a new instrument for field measurements of epidermal ultraviolet absorbance by chlorophyll fluorescence. *Applied Optics*, *43*, 4488–4496.
5. Kolb, C. A., Schreiber, U., Gademann, R., & Pfündel, E. E. (2005). UV-A screening in plants determined using a new portable fluorimeter. *Photosynthetica*, *43*, 371–377.
6. Pfündel, E. E., Ghazlen, N. B., Meyer, S., & Cerovic, Z. G. (2007). Investigating UV screening in leaves by two different types of portable UV fluorimeters reveals in vivo screening by anthocyanins and carotenoids. *Photosynthesis Research*, *93*, 205–221.
7. Cerovic, Z. G., Masdoumier, G., Ben Ghazlen, N., & Latouche, G. (2012). A new optical leaf-clip meter for simultaneous non-destructive assessment of leaf chlorophyll and epidermal flavonoids. *Physiologia Plantarum*, *146*, 251–260.
8. Julkunen-Tiitto, R., Nenadis, N., Neugart, S., Robson, M., Agati, G., Vepsäläinen, J., Zipoli, G., Nybakken, L., Winkler, B., & Jansen, M. A. (2015). Assessing the response of plant flavonoids to UV radiation: an overview of appropriate techniques. *Phytochemistry Reviews*, *14*, 273–297.
9. Cartelat, A., Cerovic, Z. G., Goulas, Y., Meyer, S., Lelarge, C., Prioul, J. L., Barbottin, A., Jeuffroy, M. H., Gate, P., Agati, G., & Moya, I. (2005). Optically assessed contents of leaf polyphenolics and chlorophyll as indicators of nitrogen deficiency in wheat (*Triticum aestivum* L.). *Field Crops Research*, *91*, 35–49.
10. Jones, C. G., & Hartley, S. E. (1999). A protein competition model of phenolic allocation. *Oikos*, *86*, 27–44.
11. Rodriguez-Calzada, T., Qian, M. J., Strid, A., Neugart, S., Schreiner, M., Torres-Pacheco, I., & Guevara-Gonzalez, R. G. (2019). Effect of UV-B radiation on morphology, phenolic compound production, gene expression, and subsequent drought stress responses in chili pepper (*Capsicum annuum* L.). *Plant Physiology And Biochemistry*, *134*, 94–102.
12. Hartikainen, S. M., Pieristè, M., Lassila, J., & Robson, T. M. (2020). Seasonal Patterns in Spectral Irradiance and Leaf UV-A Absorbance Under Forest Canopies. *Frontiers in Plant Science*, *10*, 1762.
13. Solanki, T., Aphalo, P. J., Neimane, S., Hartikainen, S. M., Pieristè, M., Shapiguzov, A., Porcar-Castell, A., Atherton, J., Heikkilä, A., & Robson, T. M. (2019). UV-screening and springtime recovery of photosynthetic capacity in leaves of *Vaccinium vitis-idaea* above and below the snow pack. *Plant Physiology and Biochemistry*, *134*, 40–52.
14. Padilla, F. M., Peña-Fleitas, M. T., Gallardo, M., & Thompson, R. B. (2014). Evaluation of optical sensor measurements of canopy reflectance and of leaf flavonols and chlorophyll contents to assess crop nitrogen status of muskmelon. *European Journal of Agronomy*, *58*, 39–52.
15. Nichelmann, L., & Bilger, W. (2017). Quantification of light screening by anthocyanins in leaves of *Berberis thunbergii*. *Planta* *246*, 1069–1082.
16. Grünig, N., Horz, J. M., & Pucker, B. (2025). Diversity and ecological functions of anthocyanins. *BMC Plant Biology*, *26*, 146.
17. Mancinelli, A. L., Huangyang, C. P., Lindquist, P., Anderson, O. R., & Rabino, I. (1975). Photocontrol of Anthocyanin Synthesis.3. Action of Streptomycin on Synthesis of Chlorophyll and Anthocyanin. *Plant Physiology*, *55*, 251–257.
18. Zirngibl, M. E., Araguirang, G. E., Kitashova, A., Jahnke, K., Rolka, T., Kühn, C., Nägele, T., & Richter, A. S. (2023). Triose phosphate export from chloroplasts and cellular sugar content regulate anthocyanin biosynthesis during high light acclimation. *Plant Communications*, *4*, 100423.
19. Porra, R. J., Thompson, W. A., & Kriedemann, P. E. (1989). Determination of accurate extinction coefficients and simultaneous equations for assaying chlorophylls a and b extracted with four different solvents: verification of the concentration of chlorophyll standards by atomic absorption spectroscopy. *Biochimica et Biophysica Acta*, *975*, 384–394.
20. Osborne, B. A., & Raven, J. A. (1986). Light-Absorption by Plants and Its Implications for Photosynthesis. *Biological Reviews of the Cambridge Philosophical Society*, *61*, 1–61.
21. Dangles, O. (2024). Anthocyanins as Natural Food Colorings: The Chemistry Behind and Challenges Still Ahead. *Journal of Agricultural and Food Chemistry*, *72*, 12356–12372.