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Hyperspectral imaging for high-throughput vitality monitoring in ornamental plant production

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A R T I C L E I N F O

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A B S T R A C T

Ornamental heather (*Calluna vulgaris*) production is characterized by high risks such as occurrence of fungal diseases and plant losses. Given the general absence of formal research on this economically important production system, farmers depend on their own approaches to assess plant vitality. We provide a reproducible, affordable and transparent workflow for assessing ornamental plant vitality with spectroscopy data. We use hyperspectral imaging as a non-invasive alternative for monitoring plant performance by combining the long-term experience of experts with hyperspectral images taken with a portable hyperspectral camera. We tested a custom-made setup deployed in a horticultural production facility and screened thousands of heather plants over a period of 14 weeks during their development from cuttings to young plants under production conditions. The vitality of shoots and roots was classified by experts for comparison with spectral signatures of shoot tips of healthy and stressed plants. To identify wavelengths that allow distinguishing between healthy and stressed heather plants, we evaluated the datasets using Partial Least Squares regression. Reflectance in the green (519–575 nm) and red-edge (712–718 nm) region of the spectrum was identified as most important for classifying plants as healthy or stressed. We transferred the trained Partial Least Squares regression model to independent test data obtained on a different date, correctly classifying 98.1% of the heather plants. The setup we describe here is adjustable and can be used to measure different plant species. We identify challenges in data evaluation, point out promising evaluation approaches, and make our dataset available to facilitate further studies on plant vitality in horticultural production systems.

1. Introduction

Producers and retailers of ornamental plants have to produce plants of high quality, in order to be competitive in the marketplace (Gullino and Garibaldi, 2007). Fungal pathogens are a major risk factor in the quality of ornamental plants (Ruett et al., 2020b; Srivastava et al., 2018). Optimized disease management and pathogen detection approaches could reduce this risk and increase the stability of production (Daughtrey and Benson, 2005). Bud-flowering heathers (*Calluna vulgaris* L.) are ornamental plants of considerable economic importance (Borchert et al., 2012, 2009) that face a high risk of fungal infection. The market value of heather plants is thus strongly influenced by farmers' decisions on management measures that affect infections and product quality (Ruett et al., 2020b). Heather plants must be diligently monitored to ensure early detection of abiotic and biotic stresses. Although it can be time-consuming and require highly skilled employees, intensified monitoring has been identified as a promising optimization strategy in commercial heather production (Ruett et al., 2020b).

Non-invasive sensor technology has been proposed for early detection of abiotic (Lowe et al., 2017) and biotic stress symptoms on plants

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(Bauriegel et al., 2011). One of the greatest advantages of these sensor-based monitoring approaches is their capacity to allow non-destructive real-time measurements (Rascher et al., 2011) and their potential for the detection of plant-pathogen interactions (Mahlein et al., 2019). In ornamental plant production, non-invasive monitoring technologies are not yet established, but have the potential to contribute to or even replace time-consuming and costly manual assessments by experts.

Sensors can collect accurate information about current plant performance (Bohnenkamp et al., 2019), which allows improved assessment of plant vitality (Knauer et al., 2017). For instance, multispectral cameras have been shown to detect tulip virus diseases with a level of accuracy that was comparable to that of human experts (Polder et al., 2014). Red, green, blue (RGB) image analysis enabled successful rust detection on Canadian goldenrod (Wijekoon et al., 2008). Thermal sensors proved applicable for the early detection of downy mildew on roses, via detection of increased leaf temperature (Gomez, 2014). Optical sensors commonly applied in plant science cover the spectral range from the visible (VIS) to near infrared radiation (NIR) (400–1000 nm) (Lowe et al., 2017). Hyperspectral imaging in the VIS/NIR region using hyperspectral sensors has been shown to be a suitable method for detecting plant stress earlier than the naked eye of experts (Behmann et al., 2014). Hyperspectral sensors are particularly promising tools for optimized monitoring, since they allow detailed assessment of plant health status and monitoring of changes in plant physiology (Mahlein, 2016). Hyperspectral sensors have also been shown to detect the water status and chlorophyll content of sunflower leaves (Neto et al., 2017), powdery mildew on barley canopy (Behmann et al., 2018) and bacterial contamination of spinach leaves (Teena et al., 2013).

Due to these capabilities, hyperspectral sensors have high potential for early detection of stress and thus improved timing of plant protection procedures (Kuska and Mahlein, 2018). In commercial heather production, such sensor tools have not yet been applied.

Hyperspectral imaging of detached heather shoots under controlled illumination conditions have been shown to allow for a precise assessment of their photosynthetic pigment and anthocyanin content (Mac Arthur and Malthus, 2012). Similarly, canopy-level spectral reflectance measurements of heather moorlands enabled the non-invasive determination of leaf pigments (Nichol and Grace, 2010), and RGB-images taken by unmanned aerial vehicles (UAVs) have been used to map flowering phenology of heathland ecosystems (Neumann et al., 2020). However, the potential of hyperspectral approaches to monitor risk factors in commercial heather production has not yet been evaluated.

Here, we explore the potential of non-invasive hyperspectral sensor technology for continuous evaluation of plant vitality over time. The aim of our study was to provide a reproducible high-throughput measurement design, accompanied by a detailed description of all data processing steps, including a Partial Least Squares regression (PLSR) based sensitivity analysis to identify the most suitable wavelengths for stress detection in heathers. We developed a novel setup with a hyperspectral sensor, which was used for weekly imaging of 3276 heather plants. In addition, we used a high-resolution camera to take photographs of the heather plants. These datasets were used to test whether spectral measurements of heather plants contain useful information about the plants' vitality status. All related models and data are published open–access for further attempts at classification (Ruett et al., 2020a). The novel approach of hyperspectral monitoring in commercial heather production outlined here may be adapted for other plant production lines and thus contribute to the development of efficient sensor-based vitality monitoring approaches that facilitate plant health management by farmers.

2. Materials and methods

In order to produce vigorous plants, heather producers use stock plants, from which vegetative clones are cut and planted into specialized trays. Farmers try to compensate for likely plant losses by planting ~10% more cuttings than the number of plants that are needed to fulfill their production targets. Depending on the variety, plant losses vary greatly, with losses up to 30% or more that can threaten the operation and contractual obligations of a farm. Because of the high risk of plant losses during the initial cultivation phase, we focused our analysis on this stage.

2.1. Plant material and growing conditions

The experiment was carried out in a commercial production system for ornamental heather plants (Europlant Canders GmbH, Straelen, Germany). All analyses were applied to heather plants of the variety 'Sanne' (Beautyladies®, Edens Creations, Oldenbroek, Netherlands). A total of 3276 cuttings were planted into 12 trays (273 cuttings each) in Baltic peat with a pH of 4 and an electrical conductivity (EC) of 0.1 mS cm^{-1}. Plants were cultivated in greenhouses (mean temperature of 15.8 °C, with a standard deviation of 6.4 °C; mean relative humidity: 82.5%, with a standard deviation of 16.9%) from 19 March 2019 until 25 June 2019 under commercial growing conditions following the farm's standard crop management practices.

2.2. Experimental layout and weekly measurement protocol

Hyperspectral and RGB images were taken nearly simultaneously to gather a dataset combining expert assessment of heather performance and hyperspectral images of the respective plants. Hyperspectral data were acquired with a hyperspectral imaging sensor device, and plant status was documented photographically with a digital single-lens reflex camera (Fig. 1).

The measurements were taken once a week over a period of 14 weeks from 26 March to 18 June 2019, covering plant development between the time the cuttings were planted and the young plant stage. Plant positions were recorded as tray (1–12), columns (A–U) and rows (1–13).

2.3. RGB imaging and expert assessment

We took the RGB images immediately after the hyperspectral imaging. For acquiring RGB image data with high spatial resolution, a Nikon D7500 camera (Nikon GmbH, Düsseldorf, Germany; Fig. 1) equipped with a 35 mm standard lens (Nikon GmbH, Düsseldorf, Germany) was mounted on a tripod (Manfrotto Vitec Imaging Solutions, Cassola, Italy) with a height of 101 cm above the plant samples. The setting allowed a resolution of 230 × 230 pixels per plant plot (2 x 2 cm) and a total of 273 plant plots per RGB image. A halogen lamp with 220 W and 5600/3200 K (ARRI AG, Munich, Germany) was used for homogeneous illumination of the experimental RGB imaging setup. The angle of the halogen lamp was set to 30° with a distance of 162 cm to the tray surface.

RGB images of all trays on all sampling dates were evaluated by two heather experts, who classified all shoots according to their perceived vitality. The heather experts focused their visual assessment on vitality traits such as shoot color, leaf structure, canopy density, and plant size. Expert #1 was a specialist in ornamental plant cultivation, and Expert #2 was an extension officer for local heather farmers. Both experts had more than 20 years of experience in heather cultivation. Experts were asked to pay particular attention to fungal disease symptoms, which often occur in heather cultivation, but which were unfortunately not detected in our study. Experts compared their classifications, adjusting and harmonizing their judgments to classify each plant. For each measurement day, experts identified 'Healthy', 'Dead', and 'Shoot Stress' plants (Table 1). On the pricking (transplanting) day (25 June 2019), experts identified 17 plants that were excluded from further cultivation due to insufficient rooting. Although the experts showed great expertise, we cannot take the experts' classification as absolute truth, since visual assessment of images can also lead to errors. Nevertheless, this approach was the most promising to incorporate long-term experiences from the

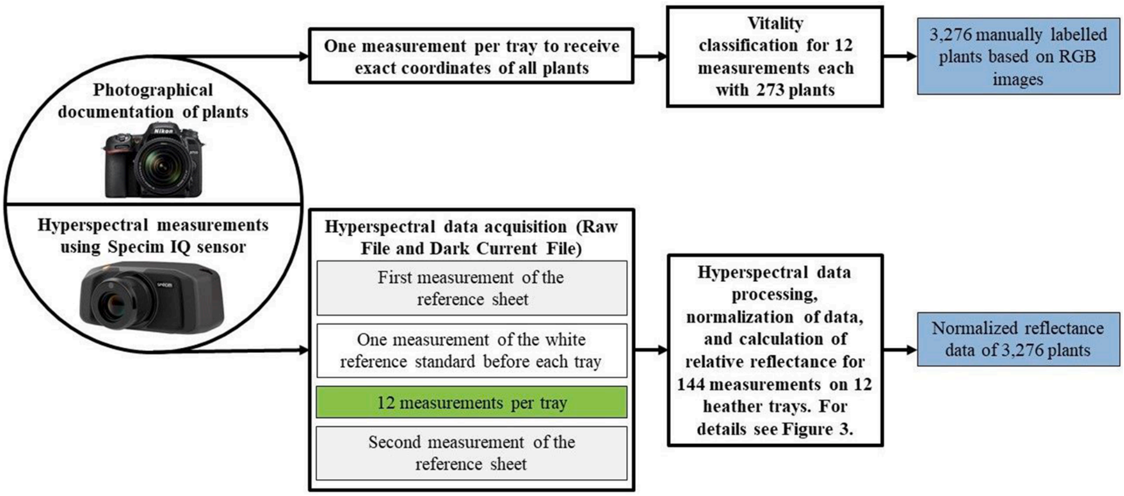

Fig. 1. Experimental layout and protocol for weekly measurements. Twelve trays with 273 plants each were photographed using a Nikon D7500 camera and imaged using the 'Specim IQ' hyperspectral imaging push-broom sensor. Output data from the vitality classification and from processed hyperspectral images are available as open access data (Ruett et al., 2020a).

Table 1
Expert assessment of health status of initially 3276 heather plants from one week after planting cuttings to pricking (transplanting) of young plants. One plant disappeared on day 14 and another plant on day 28 after planting, reducing the final plant number to 3274.

Days after planting	Healthy	Dead	Shoot Stress
7	3265	0	10
14*	3274	0	1
21	3267	0	8
28*	3250	0	24
35	3226	0	48
42	3173	1	100
49	3191	3	80
56	3247	5	22
63	3259	5	10
84	3257	5	12

* The two plants that disappeared may have been removed from the greenhouse by birds that sometimes snatch young heather plants for building nests.

practice of heather production into our study.

2.4. Hyperspectral imaging setup

The portable hyperspectral camera 'Specim IQ' (Specim Spectral Imaging Ltd., Oulu, Finland) was used to capture hyperspectral images (HS images). The HS sensor captures 204 spectral bands at wavelengths ranging from 397.32 to 1003.58 nm.

We designed an experimental hyperspectral imaging setup that allowed us to record image data from plants under controlled conditions. Our custom-made setup was constructed using aluminum tubes to mount the sensor above a standard particle board, on which sample trays could be placed. During measurements, the room was kept dark, with all incident light blocked. Two halogen lamps (500 W and 3200 K, Bresser GmbH, Rhede, Germany) with aluminum reflectors were installed on top of the setup to illuminate the samples in the VIS/NIR wavelength range. The halogen lamps were equipped with light diffusers to ensure homogeneous lighting of the whole sample tray (the full set-up is described in Table S1). We optimized conditions for data acquisition by adjusting the positions of sensor, tray surface and light source (Fig. 2). Each image covered an area of 512 × 512 pixels containing 36 cuttings on 17 x 17 cm. The hyperspectral imaging setup is adjustable to accommodate larger plants.

Each hyperspectral imaging session was initiated by setting the integration time of the HS sensor to 30 ms. An uncalibrated 10 x 10 cm Restan white reference standard (Image Engineering, Frechen, Germany) was used to calibrate the HS sensor at the beginning of each

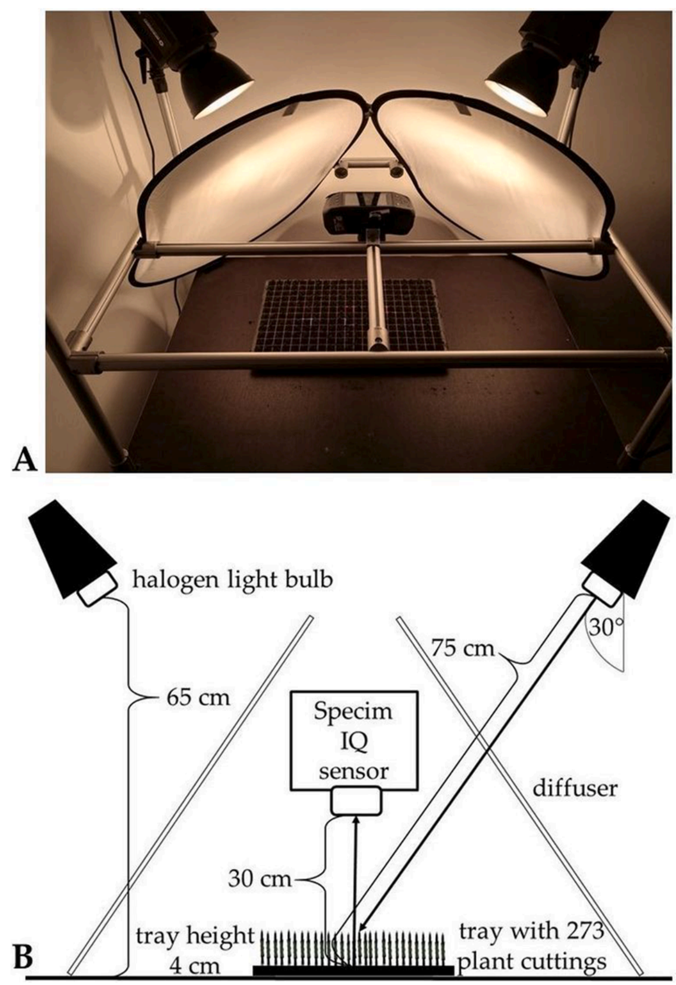

Fig. 2. Overview of the hyperspectral imaging setup using the Specim IQ sensor, two halogen lamps and two diffusors mounted on an aluminum frame (A), and conceptual drawing of the measurement setup (B).

measurement day. The Restan white reference standard was used for spectral calibration. As the Restan standard was too small to fill the entire sensor image and thus not suitable for the spatial correction of heterogeneities, we additionally measured a white photo cardboard (folia paper Max Bringmann KG, Wendelstein, Germany) (70 x 50 cm) at the beginning and at the end of each measuring day in the same position where we had positioned the tray with plant cuttings before (see Fig. 2). The white photo cardboard is hereafter referred to as reference sheet and the uncalibrated Restan white reference standard as white reference standard. Capturing the reflectance of such a reference sheet allows for the correction of spectral, spatial, and temporal variation in illumination conditions during the measurement.

2.5. Establishing a reference sheet to correct for spatial variation in illumination

A white reference covering the complete image area is essential for the HS image processing to correct for spatial variability in illumination conditions. We established a method to correct for spatial variation in illumination conditions using a white photo cardboard as reference sheet, after confirming that it had homogeneous spectral reflectance properties. Spectral reflectance was measured at six different positions with an 'ASD FieldSpec 4' point spectroradiometer (Analytical Spectral Devices, PANalytical B.V., Boulder, CO, USA) to test for spatial homogeneity of reflectance. At each position, ten measurements were recorded and averaged to reduce noise. The spectral reflectance of the reference sheet was very homogeneous with a mean of 87.3% and a standard deviation of 2.1%. For the uncalibrated Restan white reference standard the reflection was slightly lower (mean of 82.5%) and even showed greater variation (standard deviation of 2.8%) than the reference sheet.

We then compared both measurements with the spectral properties of a calibrated 95% Zenith Polymer white reference standard (SphereOptics GmbH, Herrsching, Germany) to precisely determine the spectral properties of the reference sheet and the white reference standard. The reflectance of the white reference standard varied between 80 and 87% in the spectral range of interest (450–900 nm). The reflectance of the reference sheet was, at 80–100%, consistently higher than that of the white reference standard. The spatial homogeneity and generally high reflectance of the reference sheet indicated that it was well suited to compute wavelength-specific correction factors for each spatial pixel in the HS images with the additional benefit of being more applicable under commercial conditions compared to a calibrated or uncalibrated white reference standard. Computed wavelength-specific correction factors were applied to the images of the reference sheet in the data processing procedure, which were then used for the processing of raw heather images. We were thus able to correct for spatial heterogeneity in illumination conditions and to apply spectral correction when calculating the spectral reflectance of heather plants.

2.6. Hyperspectral image processing

Hyperspectral data were processed in the R programming language (R Development Core Team, 2021). The package caTools (Tuszynski, 2020) was used to load spectral raw data into the R environment. To correct for spatial heterogeneity in illumination, raw data of the first hyperspectral reference sheet measurements were multiplied with the correction factors determined from the spectral measurements of the photo cardboard in the laboratory (cf. 2.5.). Then we applied a Gaussian filter from the package EBImage (Pau et al., 2010) with a kernel size of 3 pixels to reduce the pixel-to-pixel variability (noise) in the first hyperspectral reference sheet measurement. Due to the homogeneous surface of the reference sheet we detected little noise, all of which was removed by the filter. The same procedure was applied for the second hyperspectral imaging of the reference sheet after each measurement. We determined the relative differences between the recorded data from the first and second hyperspectral imaging in the wavelength region of 450–900 nm (mean relative difference: 7%, with standard deviation of 2.4%). The similarity of both measurements was used as an indicator for stable illumination conditions over the measurement period of 5 and 6 h during each measurement day. We then calculated the mean of the two reference sheet measurements to generate a white reference measurement (averaged reference sheet image), which was used in the further processing of the hyperspectral imaging of heather cuttings. This procedure was applied to the data of each measurement to correct for the spatial variation in illumination. As a next step, we subtracted the Dark Frame file, which is an image that the camera takes with the shutter closed to determine the Dark Current of the camera. The result was divided by the difference of the averaged reference sheet image and the averaged Dark Current of the reference sheet image to obtain reflectance. Fig. 3 illustrates the process of transferring data from raw digital numbers (DNs) to spectral reflectance (see the *R* script in S1).

To illustrate spectral signatures of the final processed data files, we manually defined pixels of sample plants as the regions of interest (ROI) within the HS image using the image processing software ENVI 4.7 (Exelis VIS, Boulder, CO, USA). We selected three to six central pixels on the shoot tips of each plant. Then we used the 'Grow' function in ENVI to automatically include shoot tip pixels with similar reflectance. Pixel numbers were increased until a minimum of 100 pixels per ROI were marked, excluding background and border pixels (mixed pixels) from the evaluation (Fig. 4). Spectral reflectance of the ROI was averaged and used for data analysis.

2.7. Identification of relevant wavelengths for classification

Partial Least Squares regression (PLSR) was applied to classify plants as healthy or stressed based on their average spectral signatures of the total ROI. We used hyperspectral images of 100 healthy and 100 stressed plants on day 42 after planting to train the algorithm. A sensitivity analysis revealed the relative importance of spectral reflectance at each wavelength for classification (Variable Importance in the Projection, VIP). A five-fold cross validation helped us to overcome potential inaccuracies related to training Machine Learning (ML) algorithms on a rather small training dataset (Hastie et al., 2009). We tested the trained algorithm on a second dataset containing 80 stressed plants and 80 randomly selected healthy neighboring plants that were measured 49 days after planting, resulting in a total of 160 observations for testing. The analysis was carried out using the 'caret' (Classification And Regression Training) (Kuhn, 2020) package for *R* and a comprehensive guide for code development (Pierobon, 2018). The PLSR code, the training dataset and the test dataset are accessible online (Ruett et al., 2020a).

3. Results

We screened the performance of 3276 heather plants over 14 weeks from cutting to young plant stage. During this time, 187 plants showed shoot stress symptoms, 5 died, and 17 plants showed insufficient rooting, with only 5 of them also showing shoot stress symptoms. In total, 5.7% of the plants showed stress symptoms.

3.1. Hyperspectral reflectance data annotated with vitality scores

To compare the spectral reflectance of ornamental heather plants with different stress symptoms, we chose five plants from one of the sample trays that differed in vitality based on the expert assessment (Fig. 5).

All plants were initially classified as healthy on day 35 after planting, but four plants showed signs of shoot or root stress classified as 'Temporary Shoot Stress', 'Dead', 'Shoot Stress', and 'Root + Shoot Stress' (Fig. 5). 'Healthy' plants showed a typical spectrum with a green peak around 550 nm and high reflectance in the NIR region from 750 to 900

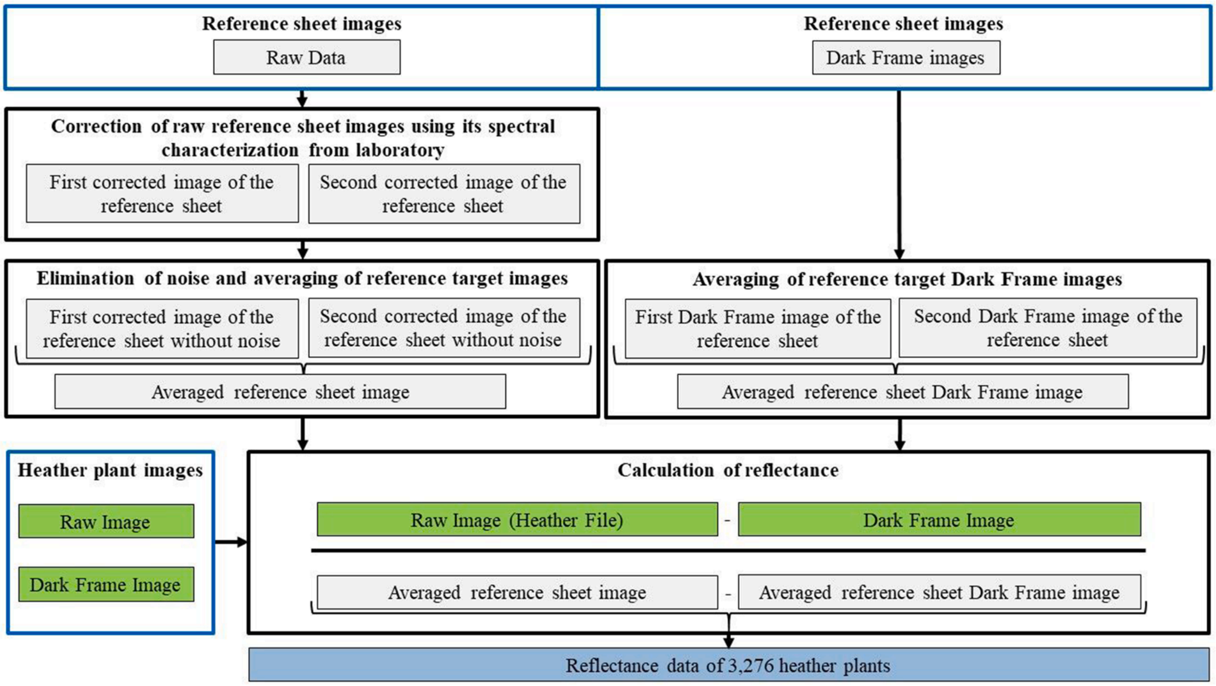

Fig. 3. Illustration of the data processing procedure to compensate for spatial heterogeneity in illumination conditions using a reference sheet. For each measurement day, the raw images of the reference sheets taken by the HS sensor were used to correct for spatial variation in illumination conditions of the hyperspectral images of trays with heather cuttings. Reflectance data is used for further analyses to detect variation between plants. Data processing started with raw images of the reference sheets obtained by the HS sensor. Reference sheet images and heather images were then passed through different processing steps. The final outputs of the processing procedure were a set of spatially and spectrally corrected reflectance data.

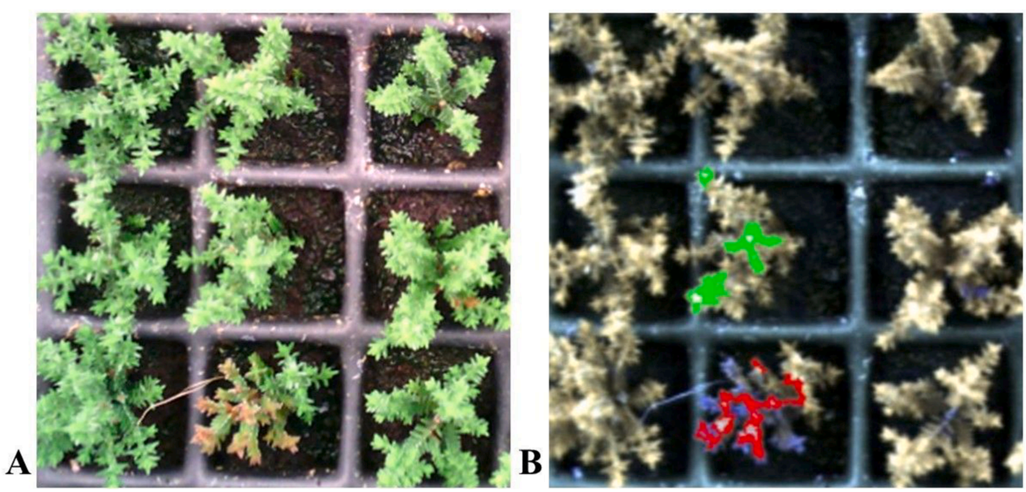

Fig. 4. RGB image of heather plants (A) and their counterpart HS images (B). The selected ROI shows green pixels for the plant classified as healthy and red pixels for the plant classified as stressed on day 63 after planting. For illustration of HS images, we selected the following spectral bands to set the RGB color space: $R = 539.75$ nm, $G = 525.10$ nm, $B = 616.34$ nm.

nm. ‘Temporary Shoot Stress’ was associated with a minor reduction of reflectance in the NIR region. The spectral reflectance of the ‘Dead’ plant did not show the typical green peak and featured a broader range of spectral reflectance values in the NIR region compared to ‘Healthy’ plants. ‘Shoot Stress’ and ‘Root + Shoot Stress’ produced a similar spectral curve over the same time period, with a lower green peak compared to ‘Healthy’ plants at the end of the experiment. ‘Root + Shoot Stress’ led to a slightly higher green peak compared to ‘Shoot Stress’. Stress also affected plant development, with ‘Healthy’ plants being larger and without signs of the reddish leaf color that was associated with ‘Dead’ plants, based on photographs taken on day 63 after planting.

3.2. *Classification and most important spectral regions*

We applied PLSR to classify healthy and stressed heather plants. We identified the most important wavelengths contributing to classification using variable importance in the projection (VIP) (Fig. 6).

Normalized reflectance of training data reveals higher spectral reflectance of healthy plants in the green and NIR regions of the spectrum, while only minor differences in reflection occur in the blue and red to red-edge region (Fig. 6A). The pattern of VIP scores is mostly in line with visual assessment of the spectra: the most important parts of the spectra for discriminating between healthy and stressed plants are located in the green region from 519 to 575 nm of the spectrum. VIP

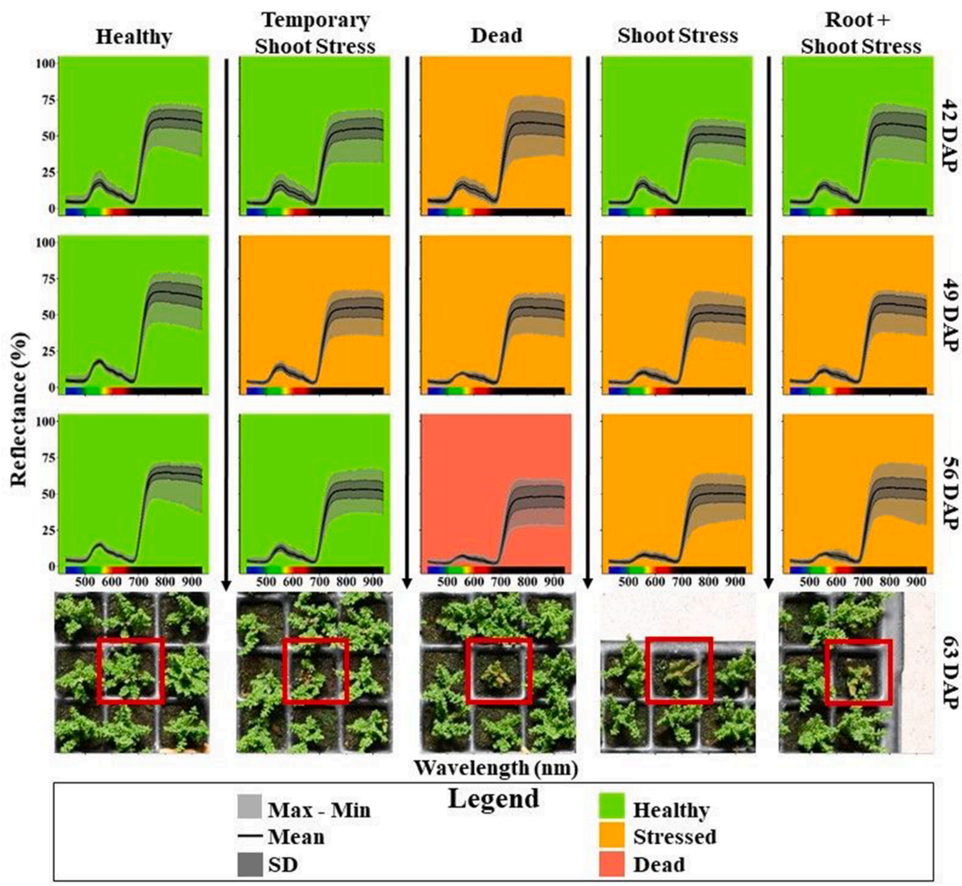

Fig. 5. Reflectance at wavelengths between 450 and 900 nm of groups of heather plants assessed by experts, which we annotated with vitality scores as 'Healthy', 'Temporary Shoot Stress', 'Dead', 'Shoot Stress', and 'Root + Shoot Stress' over three points in time from day 42 after planting (42 DAP) to day 56 after planting (56 DAP). Vitality, as assessed by experts, is indicated by the background color of each plot (Healthy = Green, Stressed = Orange and Dead = Red). Plants with insufficient root development in the 'Root + Shoot Stress' group were identified by root assessment on the pricking (transplanting) date and discarded. The graphs illustrate the mean (black lines), a confidence interval (mean ± 1 standard deviation; dark grey area) and the range between maximum (Max) and minimum (Min) (light grey area) of the spectral reflectance for each wavelength. Photographic images (bottom row) were taken on day 63 after planting. Red frames indicate the heather plants that were classified.

scores also show high importance in the red-edge region between 712 and 718 nm. Red light has a minor contribution to health classification while the contribution of blue light is negligible (Fig. 6B). When applying the model to the training data, we achieved an accuracy of 97.5% in discriminating between healthy and stressed heathers. Model coefficients reveal that relatively high reflectance of green light from 519 to 575 nm and low reflection in the red-edge region from 712 to 718 nm are characteristic of healthy heather plants and most important for classification (Fig. 6C). Validating the model on test data, we achieved even slightly higher accuracy of 98.1%, with only three out of 80 plants labeled as healthy by experts being incorrectly classified as stressed based on their reflectance, while all 80 stressed plants were correctly classified as stressed (Fig. 6D).

4. Discussion

Automated screening of ornamental plants is of rising interest (Polder et al., 2014), as monitoring plant quality can support operational decisions (Parsons et al., 2009). Highly reliable and precise methods are needed to establish automated screening processes focusing on stress and disease detection (Mahlein et al., 2018). In heather production, plants are frequently assessed visually by experts to detect stress and diseases. However, manual classification and interpretation by humans is a complex task (Laskin and McDermid, 2016), with assessments being strongly dependent on experience (Giuffrida et al., 2018). More importantly, expert assessments are time-consuming and costly and thus limited in the number of plants that can be covered (Kuska et al., 2015). Optical sensors have been shown to allow estimating the nitrogen and chlorophyll content of the ornamental plants *Chrysanthemum* (Bracke et al., 2019) and *Justicia brandegeana* (Freidenreich et al., 2019) at the leaf and canopy level. We used a hyperspectral sensor to image plants in commercial heather production and tested its suitability to identify plants that were classified as stressed by human experts. Automated quality evaluation of ornamental plants using suitable sensors and data processing pipelines might have the potential to complement existing monitoring strategies.

4.1. *Interpretation of hyperspectral reflectance data annotated with vitality scores*

The quality of ornamental plants strongly depends on shoot vitality, root number, and root function (Druege, 2020). The spectral signatures of 'Dead', 'Shoot Stress' and 'Root + Shoot Stress' show a minor reduction in the green peak compared to 'Healthy' and 'Temporary Shoot Stress' that can be related to the slightly browner shoot color observed for stressed shoots by naked-eye observation compared to the bright green shoot color for healthy shoots (e.g. RGB images from day 63 after planting in Fig. 5). Such changes in leaf color and spectral reflectance of stressed plants can be related to a lower chlorophyll content, as observed for heather plants by Mac Arthur and Malthus (2012). Wang et al. (2020) also demonstrated that hyperspectral imaging can be used to assess the foliar chlorophyll content of control and formaldehyde-treated plants with reduced chlorophyll content in 15 ornamental plant species.

4.2. *Classification of heather plants using PLSR*

Lohr et al. (2016, 2017) used NIR techniques to develop a model assessing the quality of *Pelargonium* and *Chrysanthemum* plants. In our study, application of a PLSR model on a spectrum from 450 to 900 nm facilitated identification of the most important wavelengths for classifying healthy and stressed heathers (Fig. 6C), achieving correct classification of 98.1% of test data (Fig. 6D). We detected important wavelengths in the red-edge region (Fig. 6C) with high VIP scores from

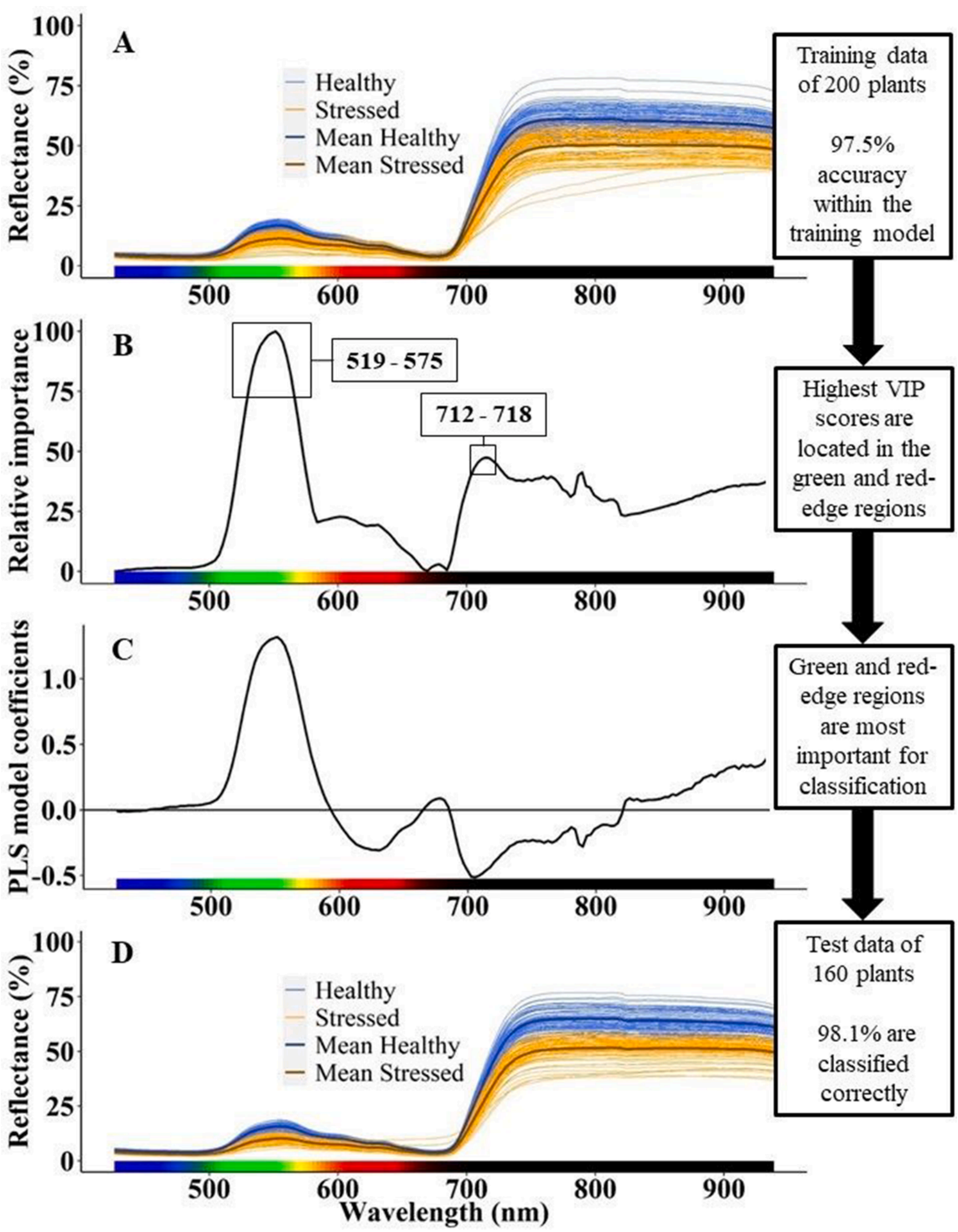

Fig. 6. Hyperspectral dataset used for deriving the PLSR model to classify healthy (thin blue lines) and stressed (thin orange lines) heather plants in the spectral range from 450 to 900 nm. Bold dark blue and dark orange lines show the mean reflectance of healthy and stressed plants, respectively. (A) Reflectance data of 100 healthy and 100 stressed plants from 42 days after planting were used to train the PLSR model. (B) The variable importance in the projection (VIP score = black line) illustrates the relative importance of spectral reflectance at a given wavelength for the classification as healthy or stressed, scaled to 100. (C) Model coefficients (black line) indicate correlation of reflectance at the respective wavelength with model outcome of healthy. (D) Reflectance data of 80 healthy and 80 stressed plants from 49 days after planting was used to test the PLSR model.

712 to 718 nm (Fig. 6B). The observed low reflectance (= a high absorption) of radiation in the red-edge range is a well-known sign of high chlorophyll concentrations in plant tissues (Filella and Penuelas, 1994; Gitelson et al., 1996; Ju et al., 2010). Radiation with wavelengths greater than 718 nm did not carry as much information relevant for discrimination of healthy and stressed plants as radiation around 550 and 715 nm.

Studies have shown that PLSR application on hyperspectral data sets is a suitable method for identifying wavelengths that are correlated with certain biological indicators (Luedeling et al., 2009). For example, PLSR has been used successfully to detect important wavelengths related to the canopy chlorophyll content in temperate forests in Germany (Hoeppner et al., 2020). The VIP score indicated the greatest potential for discriminating between healthy and stressed plants for spectral reflectance in the green spectral domain from 519 to 575 nm (Fig. 6B), with model coefficients specifying the most important wavelengths in this domain (Fig. 6C). Low reflectance of radiation in the green region at 550 nm, as observed for stressed heather plants (Fig. 6A and D), is a typical sign of enhanced anthocyanin assimilation, a stress response in higher plants (Chalker-Scott, 1999; Merzlyak et al., 2008). Similar results were obtained by Cotrozzi and Couture (2020), who analyzed spectral measurements of stressed lettuce plants using PLSR and also identified the green spectral domain as closely related to chlorophyll content, confirming the importance of these spectral bands for stress detection. A similar trend was detected by Wilson et al. (2004), who identified reduced reflectance in the green spectral domain for stressed corn leaves that were exposed to heavy metals in comparison to healthy corn leaves.

Although the overall accuracy of our classification was high (> 98 %), we took a closer look at the three plants that were classified incorrectly. All three were classified as stressed based on their reflectance patterns, while experts labeled them as healthy, not only on the day of hyperspectral data acquisition, but throughout the entire production cycle. From a farmers' point of view, erroneous classification of a healthy plant as stressed is less dangerous than the reverse case, as stressed plants that remain in the stand may serve as entry point for pathogens, potentially threatening the entire plant population. We assume that machine learning workflows should be designed to classify plants with unclear signatures rather as stressed, to avoid missing mildly stressed plants. Such conservative approaches are in line with farmers' cultivation approaches, reflecting that farmers are often risk-averse (Iyer et al., 2020) and may prefer models that rather err on the side of caution than miss potential sources of infection.

Our results indicate that multispectral cameras capturing the reflectance in the green and red region of the spectrum would be just as suited to classify plant vitality. Multispectral cameras are preferred for applied approaches compared to hyperspectral cameras because of their lower price and lower requirements for data analysis (Grieve et al., 2015; Mahlein et al., 2018).

4.3. Alternative approaches to assess plant vitality in ornamental plant production

Classical approaches like vegetation indices do not consider the full spectrum, but usually focus on just a few pre-defined wavelengths (Wahabzada et al., 2016). Advanced classifiers use all spectral information and are able to deal with the high dimensionality of hyperspectral data to detect the most important wavelengths (Paulus and Mahlein, 2020). Advanced approaches like PLSR allowed classification of healthy and stressed heather plants with the advantage to identify the most important spectral regions for classification. Based on the analysis of all spectral information of heather plants, it appears that certain classifiers may classify stress symptoms more precisely than others. Non-linear classifiers such as Support Vector Machines (SVMs) have been successful in detecting abiotic stress in plants (Zhang et al., 2018), and they have shown better performance for water-, nitrogen-, and weed- stress detection in sugarbeet than other machine learning methods such as decision trees (Khanna et al., 2019). Such SVM approaches have shown promise in early detection of stress and diseases (Thomas et al., 2018), but they can be highly time-consuming in terms of data pre-processing (Piiroinen et al., 2017). Neural Networks (NNs) may overcome such limitations by directly using data without requiring much pre-processing (Singh et al., 2018). Golhani et al. (2018) describe NNs as the approach with the highest potential for precise plant diagnosis due to its speed and high accuracy. NNs could be more efficient compared to SVM approaches when applied to our heather data, if higher speed and classification accuracy are actually achieved. Like the PLSR we applied here, these tools have shown their potential to classify plants based on hyperspectral data by incorporating the totality of measured spectra.

In recent years, Machine Learning (ML) approaches have emerged as powerful tools to solve classification problems using hyperspectral data, but there are also challenges in ML that need to be considered. Many ML methods can be described as black box models, because computational steps that detect patterns in datasets are often not well understood by users of such methods (Lipton, 2018). Another challenge is the collinearity of adjacent spectral bands in hyperspectral data (Coburn et al., 2018). Plants' spectral bands carry similar information that can overlap (as shown in Fig. 6A and D). The possibility that ML approaches may detect correlations in data that have no biological significance presents a major risk (Azodi et al., 2020). However, since the changes in reflectance patterns identified for heather plants by our approach are typical stress responses and physiologically well described and explained in the literature, we are confident that we did not find artificial correlations, but actual explainable changes that are caused by the plants' health status.

Inclusion of feature-based procedures that consider plant structure may hold promise for classifying plants according to their vitality. Object-based image analysis techniques consider shape and texture information within groups of pixels (Blaschke, 2010; Roscher et al., 2016). The substantial diversity in the appearance of plants, the highly branched shoot structure and the small leaf size of heather present challenges to hyperspectral sensing, but they may hold potential for feature-based classification procedures. Shoots of heather plants show considerable variation in size and color (images in Fig. 5). Low shoot biomass indicates weak growth, which can indicate low vitality. Compared to healthy shoots, stressed shoots are smaller and therefore represented by fewer pixels (images in Fig. 5). Combinations of spatial and spectral analyses have shown promise for plant phenotyping (Behmann et al., 2016). In this context, high spatial resolution in hyperspectral images can facilitate the analysis of shoot structures, allowing for a more detailed vitality assessment than hyperspectral data analysis alone (Behmann et al., 2016). If reduced vitality can be identified from shoot structure attributes of heather plants, object-based methods may increase the reliability of plant health assessments. The manual selection of pixels is a bottleneck that makes our method in its current state unattractive for farmers to apply. Identification of plant structure in theory allows to create a mask for hyperspectral data to automatically select pixels that are representative of a certain plant and thereby overcome that bottleneck. A combination of hyperspectral imaging with advanced evaluation methods might improve assessments of spectroscopic data (Mahlein et al., 2018). We anticipate that promising ML and feature-based methods may allow easy application of HS sensors and multispectral cameras for plant health status classification.

4.4. Outlook for the use of hyperspectral sensors in ornamental plant production

The hyperspectral imaging setup described in this study was designed for experiments at a horticultural production site. It was easy to use without intensive instructions, but required controlled illumination conditions. Hyperspectral images were repeatedly captured throughout the cultivation period to develop a hyperspectral vitality assessment classification. Such frequent measurements will not be needed for practical approaches. To establish automated plant classification by hyper- or multispectral sensors in farming routines, sensors should be integrated into farm machinery that face all plants regularly, such as pricking (transplanting) robots. Thereby, stressed plants unlikely to develop to marketable plants could be automatically discarded by robots (Polder et al., 2014). Such frequent monitoring could save resources and lower the risk of spreading fungal infections (Ruett et al., 2020b).

Before automated processes for stress detection with HS sensors can be fully applied in ornamental production practice, several challenges have to be investigated. Engineering challenges must be solved, such as how to conduct measurements under variable illumination conditions in the greenhouse. To facilitate fast data processing for simultaneous classification, the data analysis used in this study (Ruett et al., 2020a) must be further optimized and automated. From an economic perspective, the feasibility of applying sensors at production scales should be estimated by cost-benefit analyses under realistic application scenarios. In addition, biological variation between plant species, cultivars and growth stages requires determining individual classification thresholds for the respective plant type and growth stage of interest

Future research should thus focus on effective stress detection algorithms and low-cost sensors that are versatile enough to be applied to different plant species and perform under various environmental conditions, to facilitate the development of sensor-based technologies to the point of commercial applicability.

5. Conclusions

In commercial heather production, as in other intensive ornamental plant industries, plant quality and early detection of stress are critical determinants of economic success. We developed a testing procedure in which we integrated camera and sensor technologies, the setup of which was deployed in a horticultural production facility, to determine whether spectral reflectance measurements can support classification of heather plants with different health status. The hyperspectral imaging setup was specifically designed to resolve even subtle differences in reflection. Based on our experimental dataset we were able to classify healthy and stressed heather plants with an accuracy of 98.1% using PLSR. We identified reflectance in the green (519–575 nm) and red-edge (712–718 nm) regions as most important for classification. The setup and research design hold promise for experimental measurements on ornamental plants under controlled conditions, since they enable high-resolution measurements of small plant samples, clean data acquisition, and transparent data processing procedures. The resulting data set will be available for further studies on plant vitality. Future research should focus on the implementation of hyperspectral monitoring approaches in commercial plant production processes under greenhouse conditions.

CRediT authorship contribution statement

Marius Ruett: Data curation, Formal analysis, Investigation, Methodology, Software, Validation, Visualization, Writing – original draft, Writing – review & editing. **Laura Verena Junker-Frohn:** Data curation, Formal analysis, Investigation, Methodology, Supervision, Validation, Writing – review & editing. **Bastian Siegmann:** Data curation, Formal analysis, Investigation, Software, Validation, Writing – review & editing. **Jan Ellenberger:** Formal analysis, Software, Writing – review & editing. **Hannah Jaenicke:** Funding acquisition, Project administration. **Cory Whitney:** Data curation, Resources, Writing – review & editing. **Eike Luedeling:** Conceptualization, Funding acquisition, Project administration, Software, Supervision, Writing – review & editing. **Peter Tiede-Arlt:** Conceptualization, Funding acquisition, Project administration, Supervision. **Uwe Rascher:** Conceptualization, Funding acquisition, Project administration, Software, Supervision, Writing – review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Supplementary materials

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