



Comparing the performance of active and passive reflectance sensors to assess the normalized relative canopy temperature and grain yield of drought-stressed barley cultivars



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ABSTRACT

High-throughput precision phenotyping, using spectral reflectance measurements, has the potential to provide more information for making better-informed management decisions at the canopy scale in real time. Active and passive spectral reflectance sensors are available for ground-based remote sensing; however, they have not been compared in their performance for assessing the normalized relative canopy temperature (NRCT) and the grain yield of drought-stressed plants. In this study, five spectral passive and active reflectance sensors, including a hyperspectral passive sensor (HPS), a hyperspectral active sensor (HAS), an active flash sensor (AFS), the Crop Circle (CC) and the GreenSeeker (GS), were tested to assess the NRCT and grain yield of barley cultivars under mild and severe drought stress in 2012 and 2013. Simple linear regression and partial least squares regression models were used for analysing the spectral data. The results showed that the spectral indices of all sensors were more closely related to NRCT and grain yield under mild drought stress (R^2 up to 0.70, significant correlation at $p \leq 0.001$) than under severe drought stress (R^2 up to 0.53, significant correlation at $p \leq 0.001$). Closer relationships between three normalized water indices (NWI-1, NWI-3 and NWI-4) and NRCT and grain yield were obtained for the hyperspectral passive sensor compared to the same indices of the hyperspectral active sensor and the active flash sensor under both mild and severe drought stress. Multivariate analysis using partial least square regression improved the relationship (R^2 up to 0.77, significant correlation at $p \leq 0.001$) compared to the individual spectral indices and the single reflectance bands for each sensor. In conclusion, both the selection of adapted measurement devices and advanced statistical methods can improve assessments of NRCT and grain yield.

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1. Introduction

Spectral and thermal high-throughput technologies have the potential to provide quick and precise measurements of important physiological and agronomic traits for crop phenotyping in breeding nurseries (Hatfield et al., 2008; Mistele and Schmidhalter, 2010; Gutierrez et al., 2010; Elsayed et al., 2011). Proximal remote sensing systems for phenotyping on the field scale can be based on passive and active reflectance sensing. Passive sensor systems depend on sunlight as a source of light in contrast to active sensors, which are equipped with light-emitting components that provide radiation

in specific waveband regions. Therefore, active sensors are more independent of changing irradiation conditions (Kipp et al., 2014). Whereas passive sensors allow hyperspectral information to be obtained in the visible and near-infrared range currently, commercially available active sensors such as the GreenSeeker (NTech Industries Inc., Ukiah, California), the Crop Circle ACS-470[®] (Holland Scientific Inc., Lincoln, Nebraska) and an active flash sensor (AFS) (tec5 AG, Oberursel, Germany) are limited to comparatively few wavelengths according to the number and type of light sources and possible user-selectable filters (Erdle et al., 2011; Kipp et al., 2014). Active hyperspectral sensing allows for the evaluation and testing of the relationship of not yet identified wavelength combinations to relevant crop traits, which are nearly independent of the ambient environmental conditions (Erdle et al., 2011; Rischbeck et al., 2014), and has, therefore, been further tested in this study to investigate the relationship to the normalized relative canopy temperature and grain yield of drought stressed barley cultivars.

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Passive reflectance sensors have widely been used in the past to measure several canopy variables such as plant water status, biomass, leaf area index, nitrogen status or grain yield. Recently, active sensors have been used to estimate parameters such as the nitrogen status of wheat cultivars and maize hybrids (Tremblay et al., 2009; Shaver et al., 2010; Erdle et al., 2011; Winterhalter et al., 2013), the grain yield of wheat and maize (Inman et al., 2007; Marti et al., 2007), green biomass, the leaf area index and plant coverage in cereals (Trotter et al., 2008; Fitzgerald, 2010; Kipp et al., 2014).

Use of these sensors for identifying promising genotypes in a breeding program will be facilitated, if grain yield can be predicted before harvest (Royo et al., 2003). Early prediction of grain yield by spectral reflectance measurements prior to harvest could reduce phenotyping time and expenses compared to destructive measurements (Marti et al., 2007; Prasad et al., 2007).

In previous studies, several researchers suggested that the grain yield could be estimated using spectral reflectance during different growth stages (Peñuelas et al., 1997; Schmidhalter et al., 2001; Aparicio et al., 2002; Osborne et al., 2002; Babar et al., 2006; Marti et al., 2007; Prasad et al., 2007; Gutierrez et al., 2010); for example, the NDVI at the milk-grain stage was well correlated to the final wheat grain yield at two levels of nitrogen fertiliser application under rainfed and irrigated conditions. However, it was also observed that the NDVI $(R_{774} - R_{656}) / (R_{774} + R_{656})$ was also reasonably correlated to the grain yield at the onset of stem elongation (Marti et al., 2007). The normalized water index 1 (NWI-1; $(R_{970} - R_{900}) / (R_{970} + R_{900})$) and the normalized water index 2 (NWI-2; $(R_{970} - R_{850}) / (R_{970} + R_{850})$), as well as the normalized water index 3 (NWI-3; $(R_{970} - R_{920}) / (R_{970} + R_{920})$) and the normalized water index 4 (NWI-4; $(R_{970} - R_{880}) / (R_{970} + R_{880})$) from passive reflectance sensor measurements demonstrated great potential at differentiating high- and low-yielding genotypes in advanced lines of spring wheat under well-irrigated, water-stressed and high-temperature conditions in diverse trials (Gutierrez et al., 2010). Lobos et al. (2014) found that the normalized NWI-3 and the normalized difference vegetation index NDVI $(R_{830} - R_{660}) / (R_{830} + R_{660})$ were most closely related to the grain yield of wheat genotypes subjected to drought stress.

Prasad et al. (2007) found that the relationships of the grain yield of wheat cultivars with the water index (WI; (R_{970} / R_{900})) and the normalized water indices (NWI-1, NWI-2, NWI-3 and NWI-4) were stronger than with the red normalized difference vegetation index (RNDVI; $(R_{780} - R_{670}) / (R_{780} + R_{670})$) and the simple ratio (SR; (R_{970} / R_{680})). They performed better at identifying superior genotypes, either at any individual growth stage or in a combination of growth stages under rainfed conditions. Babar et al. (2006) found that under reduced irrigation, near infrared radiation (NIR)-based indices (WI, NWI-1, and NWI-2) resulted in the highest levels of association with grain yield.

The crop water stress index (CWSI) or the normalized relative canopy temperature (NRCT) is an important method to assess the drought stress based on leaf or canopy temperature. The use of leaf or canopy temperature to detect the drought stress is based on the principle that a plant's stomatal closure takes place during drought stress, which results in a decrease of energy dissipation and an increase in plant temperature (Idso et al., 1981; Patel et al., 2001). The CWSI quantifies the combined effects of soil water, atmospheric, and crop conditions on the crop water status, which ultimately affects the grain yield of cultivars. Nielsen and Anderson (1989) found that the CWSI was well related to the stomatal conductance, leaf water potential, leaf transpiration rate, available soil water, and leaf CO₂ exchange rate in sunflower. Several studies reported that there was a good relationship between the CWSI and grain yield (Irmak et al., 2000; Kashefipour et al., 2006; Zia et al., 2012).

Few studies have tried to relate spectral indices with the CWSI and canopy temperature. Zarco-Tejada et al. (2013) found that the CWSI was well related to a normalized Photochemical Reflectance Index $PRI_{norm} ((R_{570} - R_{531}) / (R_{570} + R_{531}) / (R_{800} - R_{670}) / (R_{800} + R_{670}))^{0.5} * (R_{700} / R_{670})$ and weaker relationships were obtained with the vegetation index NDVI $(R_{800} - R_{670}) / (R_{800} + R_{670})$ for vineyards at different measurement times.

Winterhalter et al. (2011) reported good relationships between the spectral index (R_{760} / R_{730}) and the canopy temperature (CT) for maize under irrigated and drought stress treatment. Observed associations between the NWI-3 and canopy temperature were consistent with the idea that genotypes with a better hydration status have a larger water flux and transpirative cooling (Gutierrez et al., 2010).

To the best of our knowledge, there is very little information available about the comparative assessments of the performance of passive and active sensing systems for assessing the grain yield and CWSI. Since the NRCT is similar to the CWSI, we preferred to the index "NRCT" in this study.

Therefore, the purpose of this work was to evaluate the performance of passive and active sensors to: i.e. (i) assess whether spectral indices can reflect changes in the NRCT of barley cultivars under drought stress conditions, (ii) assess the grain yield of barley cultivars under drought stress conditions, and (iii) compare the performance of spectral reflectance indices respective of the reflectance bands and the partial least square regression for retrieving such information from five spectral sensing systems by assessing the NRCT and grain yield of drought stressed barley cultivars.

2. Materials and methods

2.1. Field experiments and design

The field experiments were conducted at the Dürnast research station of the Technische Universität München in southwestern Germany (11°41'60" E, 48°23'60" N). Dürnast is characterized by a sub-oceanic climate, with mild cloudy winters and warm summers. The average yearly precipitation was 844 mm, and the average temperature was 8.3 °C. At the research station, two rain-out shelter facilities were used for conducting field trials under controlled drought stress. A rain-out shelter is a moving greenhouse mounted on tracks. Crops were grown under open sky, and only in case of rain does the shelter close to keep plants and the soil dry. There is a sensor that allows the rain-out shelter to move and cover the crops in case of light rain. The sensor is similar to a leaf wetness sensor. The rain-out shelter was kept closed during the autumn and winter to keep the soil dry. Spring barley was sown on 17 April 2012 and 18 April 2013. According to the residual mineral nitrogen contents in the soil measured after winter, additional mineral fertiliser was applied up to a level of 130 kg N ha⁻¹ before sowing in all trials. To reduce the lodging risk, especially of the historic cultivars, the growth regulator "Trinexapac" was applied at the initiation of the shooting (BBCH 31) in all trials.

Two types of experiments were conducted:

Mild drought stressed field trials were established in a randomized block design with five replicates in a rain-out shelter facility during the 2012 and 2013 seasons. The soil is a calcareous Cambisol consisting of silty loam. It has a field capacity of 42%, and the permanent wilting point is at 20%. The rooting depth of spring barley in this soil was assessed to be at 150 cm based on former soil sampling (Rischbeck et al., 2014). The rain-out shelter was kept closed during autumn and winter to keep the soil dry. Gravimetric soil samples were taken until a depth of 150 cm at the start and the

end of the spring barley season to assess soil water consumption in 19 plots in 2012 and in seven plots in 2013. In 2012 the average water consumption was 212 mm (182 mm of soil water and 30 mm of irrigation water). In 2013 the average water consumption was 288 mm (226 mm of soil water and 62 mm of irrigation water). Mild drought stress increased continuously throughout the season. The crop was irrigated after sowing for securing germination. Later in the season only little irrigation was applied, and continuous but mild drought stress developed. The plots consisted of eight rows spaced 15 cm apart, and each had a length of 1.8 m. In 2012, sixteen historical and modern German cultivars were grown, whereas in 2013, sixteen modern international cultivars were grown (Table 3).

Severe drought stressed field trials were established in a randomized block design with five replicates in a rain-out shelter facility in the 2012 and 2013 seasons. The re-filled soil under the rain-out shelter consists of a soil mixture (22.6% silt, 53.3% fine sand, 19.1% medium-sized sand and 5% coarse sand) at a depth of 0–20 cm and of sandy soil at a depth of 20–100 cm (60% medium sized sand, 30.6% fine sand, 3.4% coarse sand, 4.4% silt and 1.6% clay). Water was applied with spray nozzles mounted with 50 cm distance on a moving bar at 1 m above the soil surface. Irrigation was stopped from the 14th of June to 18th of June, 2012 during heading stage (BBCH 42–59) and from the 7th of June to 19th of June, 2013 during heading stage (BBCH 41–53). After this short but severe stress phase the crops were re-irrigated. This period was chosen to study the tolerance of barley cultivars to drought stress in the heading growth stage and to study drought stress effects on grain yield (Rischbeck et al., 2014).

The total amount of water added during the whole season was 114 mm in 2012 and 132 mm in 2013. The plots consisted of eight rows spaced 15 cm apart, and had a length of 1.8 m each. In 2012, nine German cultivars were grown, whereas in 2013, twelve international cultivars were grown (Table 3).

2.2. Field measurements

2.2.1. Thermal measurements

Canopy surface temperature in the field trial was determined by two HEITRONICS KT15.83D infrared thermometers (Heitronics GmbH, Wiesbaden, Germany). The thermal sensors have a spectral response in the range of 8–14 μm , their temperature resolution is 0.06 °C. The diameter of the Field of View is 0.7 mm, which enables point measurements. Rotating mirrors inside the case are used for referencing case temperature, which enables high stability of the environmental measurements. The infrared thermometers were mounted on a carrier vehicle (PhenoTrac 4 from the Chair of Plant Nutrition, TUM) pointing at the canopy from two opposed oblique views at an angle of 27° from nadir. Canopy temperature was assessed by averaging the temperatures measured by both devices. The oblique view increases the biomass fraction and decreases the soil fraction in the field of view. The thermometric devices were calibrated using a blackbody. Blackbody temperature was increased from 8 °C to 100 °C. The blackbody surface was measured by pointing the thermometric devices to it. Emissivity of the thermometric devices was set to 0.99. The thermometric devices showed a deviation from blackbody temperature of 7.2 mK/K (millikelvin/K), 15.7 mK/K for the second device used, respectively. Measurements dates are presented in Table 2.

2.2.2. Spectral reflectance measurements

The sensor systems tested were a hyperspectral passive reflectance sensor and four active reflectance sensors. The passive bidirectional reflectance sensor system (tec5, Oberursel, Germany) contained two units of a Zeiss MMS1 silicon diode array spectrometer. One unit was linked to a diffuser detecting solar radiation

as a reference signal. Simultaneously, the second unit measured the canopy reflectance, which analyzed the reflected radiation in 256 spectral channels and measured the reflectance and incident radiation simultaneously in the spectral detection range from 300 to 1000 nm, with a bandwidth of 3.3 nm, and from 1000 to 1700 nm, with a bandwidth of 6 nm (Mistele and Schmidhalter, 2008; Winterhalter et al., 2013). The spectrometer was connected with a view optic in one light fiber. The optical input was positioned at nadir direction and measured the canopy reflectance with an aperture of 12°, what resulted in a field of view (FOV) of about 0.14 m² at a height of 1 m above the canopy. The active sensors, as described by Erdle et al. (2011) and Winterhalter et al. (2013) were a GreenSeeker RT100 sensor, which uses two LEDs as a light source and detects the reflection of each in the VIS (656 nm, ~25 nm band width) and NIR (774, ~25 nm band width) spectral regions. The FOV of this device is a narrow strip of about 61 cm by 1.5 cm (0.00915 m²) at a height of 66 cm to 112 cm above the canopy (NTech Industries, 2007). A Crop Circle ACS 470 instrument, which emits white light (light source: ~400 to ~800 nm) with a selection of filters for wavelengths of 670, 730, and 760 nm used within this study. The FOV of the Crop Circle is an oval of ~32° by ~6° range (Holland-Scientific, 2008), resulting in approximately 0.064 m² at 1 m above the canopy. The third active device used was an active flash sensor (AFS) similar to the N-Sensor ALS® (YARA International, ASA) but limited to a single sensor and USB interface. The light source for this system was flashing xenon light, producing a spectral range of 650–1100 nm with 10 flashes per second and a circular field of view of approximately 0.15 m² at 1 m above the canopy. Four different wavelengths could be measured simultaneously. In this experiment, filters similar to those of the YARA ALS® system were chosen: 730, 760, 900, and 970 nm (Jasper et al., 2009; Erdle et al., 2011). Additionally, a hyperspectral active sensor (customized device, tec 5, Oberursel, Germany) was used, which collects information at 96 wavebands ranging from 380 to 1026 nm and has a spectral resolution of 7 nm. The optical input was positioned at nadir direction and measured the canopy reflectance with an aperture of 11.4°, resulting in a field of view (FOV) of about 0.13 m² at a height of 1 m above the canopy. The aperture of an optical system is the opening that determines the cone angle of a bundle of rays that enter the optics (Rischbeck et al., 2014). The light source of the active sensor is a xenon flash lamp. The device was initially calibrated using a white paper at a distance of 1 m and the surface area was >0.13 m². Reflectance is calculated by dividing the incoming light intensity by the light intensity reflected from the reference surface at each wavelength.

Active and passive optical sensor systems were mounted on a frame in front of a tractor-based measuring platform (the mobile multi-sensor phenotyping platform PhenoTrac 4) <http://www.pe.wzw.tum.de>, which was developed by the Chair of Plant Nutrition, Technische Universität München. The sensors were driven to measure the spectral reflectance of all plots. Independent of the sensor system, all devices were used at a nadir position above the canopy across the season. This height was chosen to constantly sense the plots' central area and avoid the unintended detection of the plot borders. While collecting information in the field, the sensor outputs were co-recorded along with the GPS coordinates from a RTK-GPS (real-time kinematic global positioning system) (Trimble, Sunnyvale, CA, USA). The frequency of recordings was primarily dependent on the output frequency of the RTK-GPS. For each position, the actual sensor output was co-referenced and recorded. Afterwards, readings within one plot were averaged to single values per plot.

Spectral reflectance measurements were taken mostly on sunny days in 2012 and 2013. Spectral measurements were taken on 25 May 2012 (BBCH 32), 18 June 2013 (BBCH 55) and 19 June 2013 (BBCH 56) in the mild drought stress trial as well as on 18 June

2012 (BBCH 60), 21 June 2012 (BBCH 63), and 19 June 2013 (BBCH 56) in the severe drought stress trial.

2.3. Plant traits

2.3.1. Grain yield

Plots in the rain-out shelter trials were harvested by hand. Total grain yield was weighed for each plot, samples were oven-dried to determine grain water content on a gravimetric basis and the yield was expressed as t/ha, normalized to a water content of 14%_w. Plot yields were averaged for each cultivar in each field trial.

2.3.2. Normalized relative canopy temperature

The normalized relative canopy temperature (NRCT) was calculated according to the formula by Jackson et al. (1981):

$$(T - T_{\min}) / (T_{\max} - T_{\min})$$

where T is the actual infrared temperature measured in the canopy, T_{\min} is the lowest temperature measured in the whole field trial (lower baseline) and T_{\max} is the highest temperature in the whole field trial (upper baseline). Jackson et al. (1981) used a wet bulb temperature as the lower baseline and a dry bulb temperature as the upper baseline, calculated based on air temperature and air vapour pressure deficit, whereas in this experiment canopy measurements were used. The advantage of this method is that no additional measurements other than infrared temperatures are necessary. Even in drought-stressed, large field trials usually temperatures of fully transpiring and non-transpiring canopies can be found and used as baselines. However, empirically measured temperatures can differ from calculated baselines. Conditions like incident radiation or wind speed impact surface temperature (Jones et al., 2009), but do not enter calculations of CWSI. Deviations of NRCT and CWSI can occur. The NRCT makes temperature measurements interpretable as a reduction in stomatal conductance and allows for the comparison of drought stress levels in different field trials under different environmental conditions.

2.4. Statistical analysis

2.4.1. Selection of spectral reflectance indices and single reflectance bands

In Table 1 thirteen spectral indices from different sources are listed with reference. They were selected, based on their reported relationships with water status, biomass and agronomic traits. The other three indices for this work listed in Table 1, were selected based on contour maps (Figs. 1 and 2). For the contour maps all possible combinations of binary, normalized spectral indices were calculated from spectral measurements with the hyperspectral passive (300–1200 nm) and the hyperspectral active reflectance sensor (370–1026 nm) for each plot in the severely and the mildly drought stressed field trial. Results were correlated with the NRCT (Fig. 1) and grain yield (Fig. 2) from the same plots. Contour maps are matrices of the coefficients of determination of these correlations. The selected normalized difference indices presented generally more stable relationships with the NRCT and the grain yield than other spectral indices at different dates in 2012 and 2013 (Figs. 1 and 2, Table 4). The R package “lattice” from the software R statistics version 3.0.2 (R foundation for statistical computing 2013) was used to produce the contour maps. From the hyperspectral reflectance readings, fifteen single wavebands (510, 560, 570, 656, 670, 740, 760, 774, 780, 810, 850, 880, 900, 920, and 970 nm) with high correlation to yield and the NRCT across plots (data not shown) were selected for modeling.

From the active multi-band sensors all measured single wavebands (Active Flash Sensor AFS_730, AFS_760, AFS_970 and AFS900;

Crop Circle CC_760, CC_670 and CC_730; Green Seeker GS_774 and GS_656) and normalized binary indices showing high correlations with NRCT and grain yield (Table 6) were selected and used for modeling.

2.4.2. Modeling of NRCT and grain yield

Sigmaplot for Windows v.12 (Systat software Inc., Chicago), and Microsoft Excel 2010 were used for the statistical analysis. Simple linear regressions were calculated to analyze the relationship between spectral indices listed in Table 1 with the NRCT and grain yield for the hyperspectral passive sensor (Table 4) and the hyperspectral active sensor (Table 5). Simple linear regressions were calculated to analyze the relationship between single reflectance bands and spectral indices with the NRCT and grain yield for the Active Flash Sensor, the Crop Circle and the Green Seeker (Table 6). Coefficients of determination and significance levels were determined; nominal alpha values of 0.05, 0.01 and 0.001 were used (Tables 4–6).

The Unscrambler X multivariate data analysis software version 10.2 (CAMO Software AS, Oslo) was used to calibrate and validate partial least square models. Single wavebands and indices derived from the same spectra usually contain redundant information (Sharabian et al., 2014). Partial Least Square Regression (PLSR) creates orthogonal latent variables across the input variables (indices and single wavebands) and relates them to the target variables (NRCT and grain yield). This is a way to cope with redundancy in the input variables. The PLSR searches the sensitive information from normalized difference indices or reflectance bands, for each spectral reflectance sensor. For the hyperspectral passive sensor indices listed in Table 4 were used as input variables in the PLSR models shown in Tables 7 and 8. For the hyperspectral active sensor the indices listed in Table 5, except the three normalized water indices, were used as input variables in the PLSR models shown in Tables 7 and 8. For the Active Flash Sensor, the Crop Circle and the GreenSeeker the single wavebands and indices presented in Table 6 were used as input variables for the PLSR models shown in Tables 7 and 8. For the models datasets from mild and severe drought stress conditions were used.

For determining model quality two different approaches of validation were used:

In Table 7 a (12 fold) cross validation approach was applied for the PLSR models. Calibration and validation quality of models is presented through adjusted coefficients of determination of calibration (R^2_{cal}) and validation (R^2_{val}) and root mean square errors for calibration (RMSEC) and for validation (RMSEV). Scatter plots of NRCT and grain yields predicted from calibration and validation models with observed data for 18. June 2013 are shown in Fig. 3.

In Table 8 a validation approach using fully independent data was used. PLSR models were calibrated using datasets at 18 June 2012 and 18 June 2013 and validated using data from another day in the same or a corresponding field trial. The quality of the validation models is presented through adjusted coefficients of determination and the slope and intercept of the linear regressions between observed and predicted values of the grain yield and the NRCT.

3. Results

3.1. Variation in the NRCT and grain yield of barley cultivars under mild and severe drought stress in 2012 and 2013

The highest value of the across trial average grain yield of 6.54 t/ha was recorded under mild drought stress in 2013. The highest across trial average NRCT value was 0.83 in 2013 under severe

Table 1
Device, formula, index abbreviation, references and field of view of different previously developed and new spectral indices used in this study.

Device	Formula	Index abbreviation	References	Field of view
Hyperspectral passive sensor	$(R_{730} - R_{670}) / (R_{730} + R_{670})$	HPS 730.670; HAS 730.670	This work	0.14 m ² at height 1 m
	$(R_{760} - R_{670}) / (R_{760} + R_{670})$	HPS_NDVI; HAS_NDVI	Kipp et al. (2014)	
Hyperspectral active sensor	$(R_{760} - R_{730}) / (R_{760} + R_{730})$	HPS 760.730; HAS 760.730	Barnes et al. (2000)	0.13 m ² at height 1 m
	$(R_{780} - R_{670}) / (R_{780} + R_{670})$	HPS_NDVI-1; HAS_NDVI-1	Rouse et al. (1974)	
	$(R_{970} - R_{920}) / (R_{970} + R_{920})$	HPS_NWI-3; HAS_NWI-3	Gutierrez et al. (2010)	
	$(R_{970} - R_{880}) / (R_{970} + R_{880})$	HPS_NWI-4; HAS_NWI-4	Prasad et al. (2007)	
	$(R_{970} - R_{900}) / (R_{970} + R_{900})$	HPS_NWI-1; HAS_NWI-1	Babar et al. (2006)	
	$(R_{780} - R_{510}) / (R_{780} + R_{510})$	HPS 780.510; HAS 780.510	Mistele et al. (2012)	
	$(R_{850} - R_{560}) / (R_{850} + R_{560})$	HPS 850.560; HAS 850.560	This work	
Crop Circle	$(R_{730} - R_{670}) / (R_{730} + R_{670})$	CC 730.670	Erdle et al. (2011)	0.064 m ² at height 1 m
	$(R_{760} - R_{730}) / (R_{760} + R_{730})$	CC 760.730	Barnes et al. (2000)	
	$(R_{760} - R_{670}) / (R_{760} + R_{670})$	CC_NDVI	Jasper et al. (2009)	
GreenSeeker Active flash sensor	$(R_{774} - R_{656}) / (R_{774} + R_{656})$	GS_NDVI	Erdle et al. (2011)	0.00915 m ² at height from 0.66 m to 1.12 m
	$(R_{970} - R_{900}) / (R_{970} + R_{900})$	AFS_NDW-1	Babar et al. (2006)	
	$(R_{760} - R_{730}) / (R_{760} + R_{730})$	AFS 760.730	Barnes et al. (2000)	

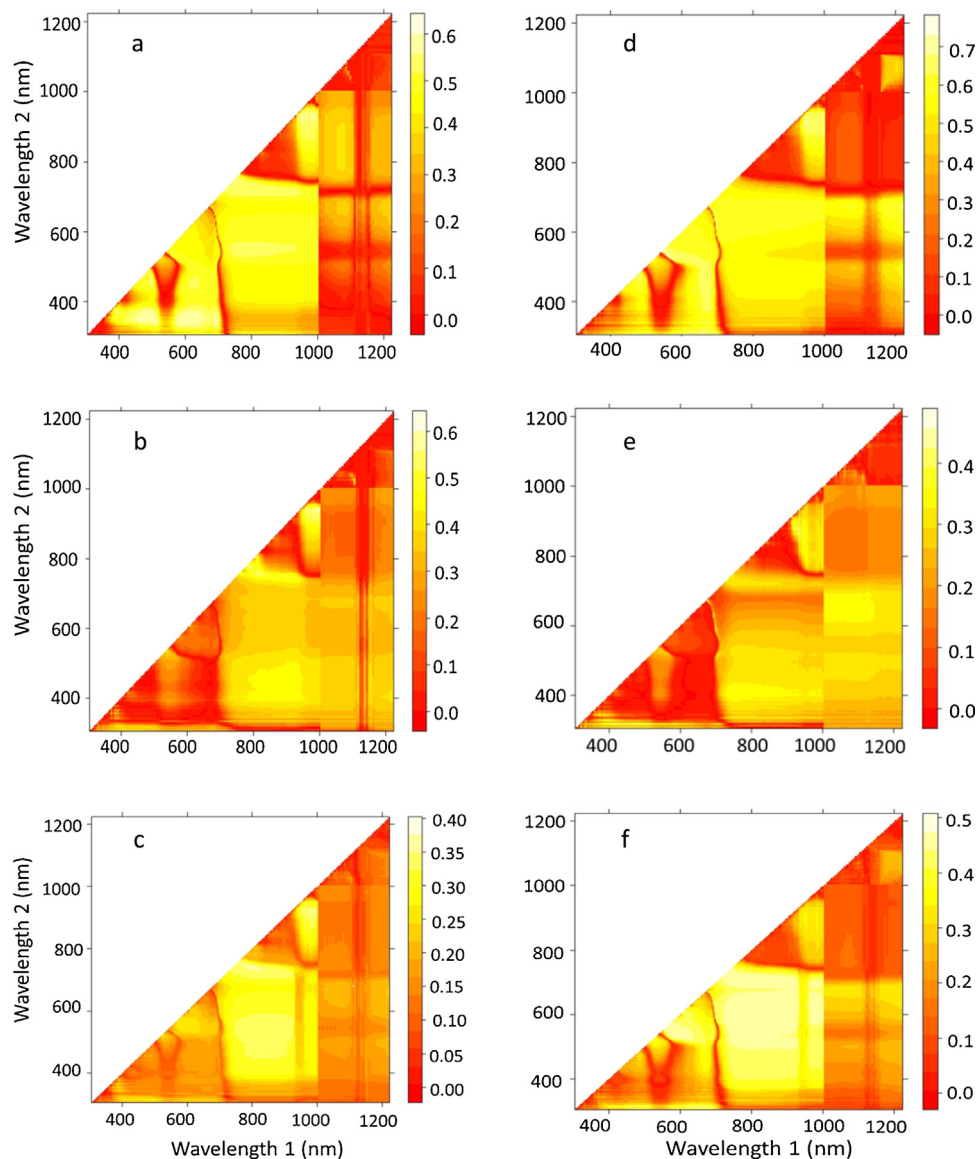


Fig. 1. Correlation matrices (contour maps) showing the coefficients of determination (R^2) for all dual wavelength combinations in the range of 300–1200 nm (as a normalized difference index) of the hyperspectral passive reflectance sensor with grain yield: (a) under mild stress on 18 June 2013 and (b) under severe stress at 21 June 2012, and with the NRCT: (d) under mild drought stress on 18 June 2013 and (e) under severe drought stress on 18 June 2012. Mean coefficients of determination (R^2) of the spectral indices with the grain yield for six measurement dates (c). Mean coefficients of determination (R^2) of the spectral indices with the NRCT for six measurement dates (f).

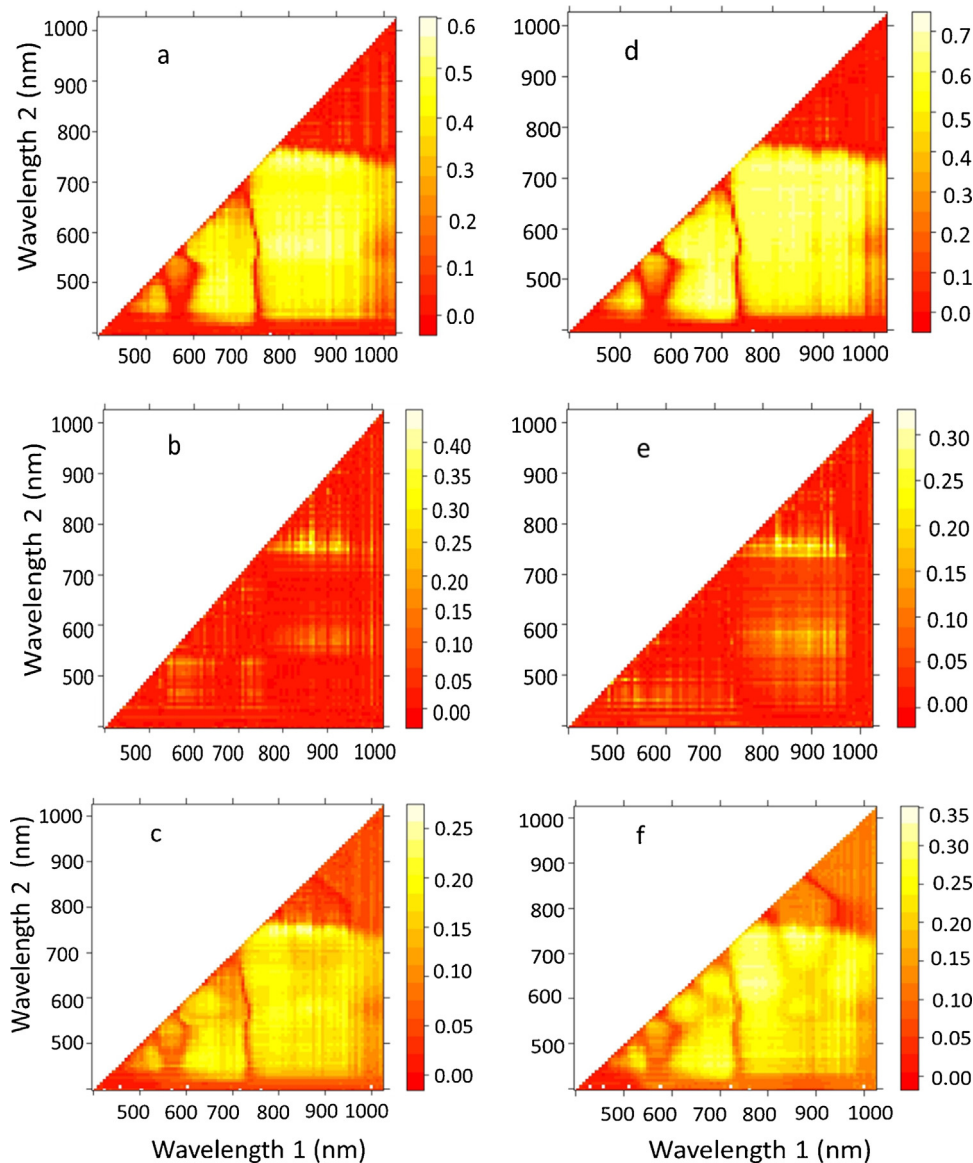


Fig. 2. Correlation matrices (contour map) showing the coefficients of determination (R^2) for all dual wavelength combinations in the range of 370–1026 nm (as a normalized difference index) of the hyperspectral active reflectance sensor with the grain yield: (a) under mild stress on 18 June 2013 and (b) under severe stress on 21 June 2012, and with the NRCT: (d) under mild drought stress on 18 June 2013 and (e) under severe drought stress on 18 June 2012. Mean coefficients of determination (R^2) of the spectral indices with the grain yield for six measurement dates (c). Mean coefficients of determination (R^2) of spectral indices with the NRCT for six measurement dates (f).

drought stress. The value of the mean NRCT for both years varied positively with the drought stress level. Statistically significant linear relationships between the NRCT and the grain yield were found on four out of six measurement dates ($R^2 \geq 0.18$, $p \leq 0.01$;

Table 2). Significant differences ($p \leq 0.05$) were found for the mean grain yield among the barley cultivars under mild drought stress in 2012 and 2013, as well as under severe drought stress in 2012 and 2013 (Table 3). The cultivar Barke presented the highest grain

Table 2

Minimum, maximum and mean of plots of the normalized relative canopy temperature (NRCT) and the grain yield of barley cultivars subjected to mild and severe drought stress for six measurement dates in 2012 and 2013. Coefficients of determination (R^2 values) of correlations between NRCT and grain yield are shown.

Measurement Date	No. of plots	Drought stress level	NRCT			Plot grain yield (t/ha)			R^2
			Min	Max	Mean	Min	Max	Mean	
25 May 2012	84	Mild	0.12	0.80	0.33	3.60	8.10	5.71	0.18**
18 June 2012	48	Severe	0.30	0.82	0.65	2.82	5.63	4.25	0.22***
21 June 2012	48	Severe	0.32	0.75	0.48				0.04
18 June 2013	84	Mild	0.20	0.88	0.51	3.63	10.55	6.54	0.36***
19 June 2013	84	Mild	0.12	0.69	0.35				0.35***
19 June 2013	46	Severe	0.72	0.92	0.83	1.31	5.62	2.95	0.06

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

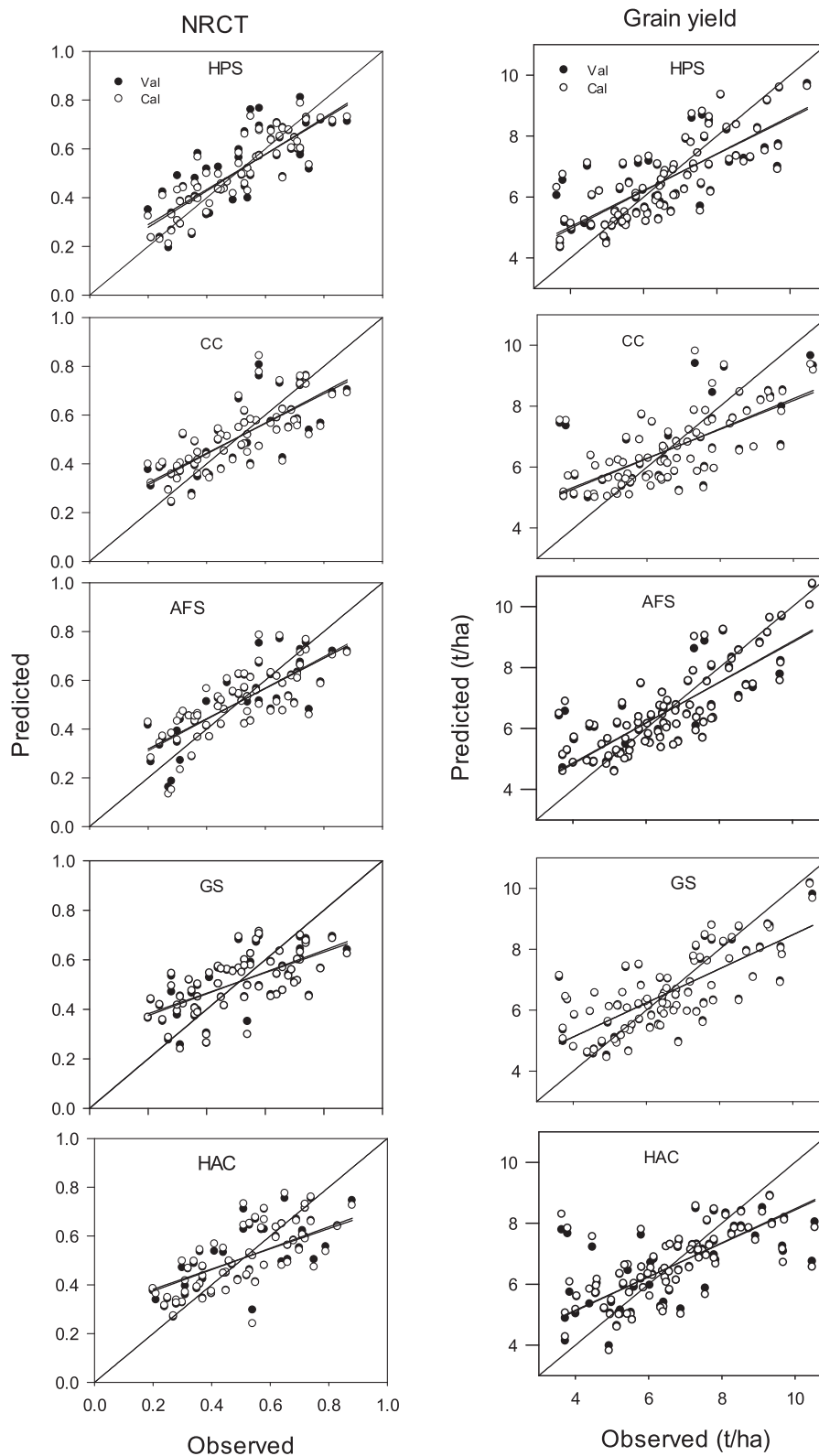


Fig. 3. Scatter plots and linear regressions between observed and predicted values of NRCT and grain yield of barley cultivars. Predicted values come from partial least square regressions using spectral indices and single wavebands (listed in Tables 4–6) from the sensor systems: Hyperspectral passive sensor (HPS); Crop Circle (CC); Active flash sensor (AFS); GreenSeeker (GS) and Hyperspectral active sensor (HAS) measured on 18 June 2013 in a mild drought stress field trial. 12-fold cross-validation was performed based on the same data.

yield of 6.43 t/ha under mild drought stress, and Beatrix presented the highest grain yield with 4.87 t/ha in 2012 under severe drought stress. Djamilia showed the highest grain yield of 7.87 t/ha under mild drought stress and 4.42 t/ha under severe drought stress in

2013. The range of the mean grain yields among the barley cultivars was 1.69 t/ha under mild drought stress and 1.67 t/ha under severe drought stress in 2012, and 2.62 t/ha under mild drought stress and 2.52 t/ha under severe drought stress in 2013 (Table 3).

Table 3

Average grain yields of barley cultivars under mild or severe drought stress in 2012 and 2013. Values with the same letter are not significantly different ($p \geq 0.05$) among cultivars according to Duncan's test. SD indicates standard deviation.

Mild drought stress in 2012			Severe drought stress in 2012			Mild drought stress in 2013			Severe drought stress in 2013		
Cultivars	Grain yield (t/ha)	SD (t/ha)	Cultivars	Grain yield (t/ha)	SD (t/ha)	Cultivars	Mean (t/ha)	SD (t/ha)	Cultivars	Grain yield (t/ha)	SD (t/ha)
Barke	6.43a	0.75	Beatrix	4.87a	0.37	Djamila	7.87a	1.90	Djamila	4.42a	0.89
Sissy	6.39a	0.79	Djamila	4.81a	0.69	Wiebke	7.68ab	1.91	Scarlett	3.95a	0.42
Beatrix	6.35a	0.32	Apex	4.67a	0.31	Extract	7.16ab	1.28	Quench	3.57ab	0.77
Trumpf	6.30a	0.84	Wiebke	4.64a	0.23	Ursa	6.93ab	1.13	Mutante	3.55ab	0.19
Streif	6.23ab	0.26	Eunova	4.61a	0.38	Quench	6.92ab	1.84	Grace	3.51ab	0.56
Wiebke	5.99abc	0.52	Ursa	4.04b	0.26	Eunova	6.91ab	1.04	Ursa	3.49ab	1.11
Victoriana	5.84abc	0.96	Barke	3.98b	0.43	Grace	6.71ab	1.64	Wiebke	2.66bc	0.68
Djamila	5.79abcd	1.36	Heils Franken	3.59bc	0.44	Trumpf	6.58ab	0.54	Sissy	2.33c	0.21
Pflugs Intensiv	5.71abcd	0.61	Bavaria	3.18c	0.26	Streif	6.47ab	1.58	IPZ24727	2.21c	0.72
Perun	5.59abcd	0.48				Gimpel	6.22ab	2.14	Trumpf	1.99c	0.87
Ursa	5.58abcd	0.66				Mutante	6.11ab	1.43	Streif	1.97c	0.77
Apex	5.52abcd	0.53				IPZ24727	5.95ab	1.28	Barke	1.90c	0.48
Heils Franken	5.24bcd	0.48				BRS195	5.84ab	2.36			
Isaria	5.03cd	0.75				Scarlett	5.56ab	0.87			
Eunova	4.78d	0.47				Sissy	5.55ab	2.13			
Bavaria	4.76d	0.59				Barke	5.26c	1.46			

3.2. Contour map analysis of the hyperspectral passive and active sensor data

The contour map analysis of the relationship between the grain yield respectively NRCT with normalized spectral indices under mild drought stress (Figs. 1a, d and 2a, d) revealed generally higher coefficients of determination (R^2 -values) than a contour map analysis done for the severe drought stress (Figs. 1b, e and 2b, e). The contours of the matrices of the hyperspectral passive sensor (Fig. 1) presented generally distinct relationships with the NRCT and the grain yield under mild and severe drought stress in the visible and near infrared area, and indicated strong relationships in the water absorption band (Fig. 1c and f, 970 nm). By contrast, the contours of the matrices of the hyperspectral active sensor (Fig. 2) presented generally distinct relationships in the visible and near infrared area only under mild drought stress (Fig. 2a and b), and showed no clear relationships to the water stress bands (Fig. 2c and f). The contours of the matrices from the active sensor (Fig. 2) were less distinct than those from the passive sensor (Fig. 1).

3.3. The relationship between the NRCT and grain yield with spectral reflectance indices and reflectance bands of different spectral sensing systems

Across the six measuring dates, spectral indices and single reflectance bands of five spectral systems were correlated with the NRCT and the grain yield of the barley cultivars. The obtained coefficients of determination (R^2) are shown in Tables 4–6. Linear relationships were chosen to assess the relationship between spectral indices and reflectance bands with the NRCT and grain yield. The closest significant relationships for both mild and severe stress were found for the hyperspectral passive sensor (R^2 values ranging from 0.1 to 0.67), the Crop Circle (R^2 values ranging from 0.08 to 0.72), the active flash sensor (R^2 values ranging from 0.1 to 0.67), and the GreenSeeker (R^2 values ranging from 0.11 to 0.57), respectively (Tables 4 and 6), and a less close relationship was obtained from the hyperspectral active sensor (R^2 values ranging from 0.1 to 0.63), specifically at severe drought stress as indicated in Table 5. In contrast to the hyperspectral active sensor, statistically significant linear relationships between the NRCT and the grain yield were obtained with three normalized water indices of the hyperspectral passive sensor (Tables 4 and 5).

3.4. Partial least squares regression analysis to predict NRCT and grain yield

In Table 7 the quality of the PLSR models is presented through adjusted coefficients of determination of calibration (R^2 cal) and validation (R^2 val) and root mean square errors (RMSE). Across all calibration data sets, the closest relationships for the NRCT ($R^2 = 0.76$) and for the grain yield ($R^2 = 0.69$) were recorded for the hyperspectral passive sensor. Across all validation data set formations, the highest coefficients of determination, with $R^2 = 0.70$ for the NRCT and $R^2 = 0.65$ for the grain yield, were recorded for the hyperspectral passive sensor and the active flash sensor, respectively. The closest relationships for both calibration and validation data sets were found for the hyperspectral passive sensor, the Crop Circle, and the Active flash sensor, and less close relationships were obtained for the hyperspectral active sensor and the GreenSeeker (Fig. 3 and Table 7). In the independent validation in Table 8 the highest coefficients of determination with $R^2 = 0.70$ for the NRCT were obtained with spectral indices and reflectance bands recorded from the Crop Circle. The highest coefficients of determination with $R^2 = 0.63$ for the grain yield were obtained for the spectral indices recorded for the hyperspectral passive sensor. The highest slope recorded was 0.91 for the NRCT and obtained for the hyperspectral active sensor (Table 8).

4. Discussion

Different passive and active reflectance sensors were used in this study to assess the NRCT and grain yield under mild and severe drought stress conditions for six measurement dates in 2012 and 2013. Drought stress affected both the NRCT and grain yield. Lobos et al. (2014) found that the exposure of wheat genotypes to two levels of drought stress (mild and severe drought stress) led to a noticeable decrease in grain yield. Patel et al. (2001) found that drought stress led to an increase in the crop water stress index. These results agree with our results obtained for barley cultivars (Tables 2 and 3), where the NRCT increased and the grain yield decreased with progressive drought stress in 2012 and 2013. Among the barley cultivars, significant differences in mean grain yield are found in the mild drought stress trials in 2012 and 2013, as well as in the severe drought stress trials in 2012 and 2013 (Table 3). These results agree with the findings of Samarah et al. (2009), who reported variations in the grain yield of four barley cultivars under

Table 4
Coefficients of determination of linear regressions of the NRCT and the grain yield with spectral indices of the hyperspectral passive sensor (HPS) (calculated as normalized difference indices) for barley cultivars subjected to mild and severe drought stress in 2012 and 2013.

Spectral indices of HPS	Parameters	25 May 2012 Mild stress	18 June 2012 Severe stress	21 June 2012 Severe stress	18 June 2013 Mild stress	19 June 2013 Mild stress	19 June 2013 Severe stress
730.670	NRCT	0.61 ^{***}	0.11 [*]	0.19 ^{**}	0.56 ^{***}	0.59 ^{***}	0.05
	Grain yield	0.19 ^{**}	0.01	0.29 ^{***}	0.44 ^{***}	0.42 ^{***}	0.24 ^{***}
NDVI	NRCT	0.59 ^{***}	0.16 ^{**}	0.27 ^{***}	0.59 ^{***}	0.60 ^{***}	0.06
	Grain yield	0.19 ^{**}	0.04	0.34 ^{***}	0.45 ^{***}	0.46 ^{***}	0.22 ^{***}
NDVI-1	NRCT	0.59 ^{***}	0.16 ^{**}	0.28 ^{***}	0.59 ^{***}	0.60 ^{***}	0.06
	Grain yield	0.19 ^{**}	0.05	0.35 ^{***}	0.45 ^{***}	0.47 ^{***}	0.22 ^{**}
760.730	NRCT	0.55 ^{***}	0.31 ^{***}	0.34 ^{**}	0.51 ^{**}	0.52 ^{**}	0.08
	Grain yield	0.16 ^{**}	0.35 ^{***}	0.35 ^{**}	0.56 ^{***}	0.55 ^{***}	0.07
780.510	NRCT	0.61 ^{***}	0.30 ^{***}	0.25 ^{***}	0.56 ^{***}	0.51 ^{**}	0.01
	Grain yield	0.18 ^{**}	0.23 ^{***}	0.41 ^{***}	0.48 ^{**}	0.44 ^{**}	0.24 ^{***}
850.560	NRCT	0.60 ^{***}	0.29 ^{**}	0.10 [*]	0.54 ^{**}	0.55 ^{***}	0.01
	Grain yield	0.19 ^{**}	0.28 ^{***}	0.37 ^{***}	0.52 ^{**}	0.53 ^{**}	0.13 [*]
810.740	NRCT	0.53 ^{***}	0.27 ^{***}	0.28 ^{**}	0.37 ^{***}	0.46 ^{***}	0.03
	Grain yield	0.16 ^{**}	0.51 ^{***}	0.43 ^{***}	0.52 ^{***}	0.52 ^{***}	0.04
NWI-1	NRCT	0.53 ^{***}	0.28 ^{**}	0.16 [*]	0.65 ^{***}	0.36 ^{***}	0.02
	Grain yield	0.22 ^{***}	0.52 ^{***}	0.49 ^{***}	0.55 ^{***}	0.38 ^{***}	0.14 [*]
NWI-3	NRCT	0.53 ^{***}	0.26 ^{**}	0.16 [*]	0.67 ^{***}	0.36 ^{***}	0.01
	Grain yield	0.23 ^{***}	0.53 ^{***}	0.51 ^{***}	0.57 ^{***}	0.37 ^{***}	0.15 [*]
NWI-4	NRCT	0.52 ^{***}	0.29 ^{**}	0.18 [*]	0.63 ^{***}	0.34 ^{***}	0.02
	Grain yield	0.23 ^{***}	0.49 ^{***}	0.43 ^{***}	0.54 ^{**}	0.37 ^{***}	0.14 [*]

* Statistically significant at $p \leq 0.05$.

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

irrigated, mild and severe drought stress. Negative significant linear relationships between the NRCT and grain yield were found ($R^2 \geq 0.18$, $p \leq 0.01$; Table 2) at four different measurement dates under mild and severe drought stress. These results agree with findings of Irmak et al. (2000), Kashefipour et al. (2006) and Zia et al. (2012), who found strong relationships between the CWSI and grain yield of corn, barley and wheat cultivars under irrigated, rainfed and drought stress conditions.

In this study, high-throughput passive and active sensing, as well as thermal near infrared sensing, was found to present some major advantages. Spectral and thermal measurements could be performed simultaneously and in a short time by using the mobile multi-sensor phenotyping platform, PhenoTrac 4. Fast measurements can reduce disturbances caused by shifting illumination

and temperature conditions. In several other studies (Ferrio et al., 2005; Inman et al., 2007; Prasad et al., 2007; Gutierrez et al., 2010) handheld sensors were used for spectral and temperature measurements for assessing grain yield and the crop water stress index or. This method is more time consuming and may, therefore, be more affected by external factors, such as ambient climatic conditions. The hyperspectral passive sensor (HPS) and the hyperspectral active sensor (HAS) used in this study offer a broad range of wavelengths in the visible and near infrared area that enabled the calculation of many spectral indices (Tables 4 and 5). In contrast, the other three active sensors, the Crop Circle (CC), active flash sensor (AFS) and Green Seeker (GS) use few wavebands according to the type of light sources and the number and type of filters (Table 6).

Table 5
Coefficients of determination of linear regressions of the NRCT and the grain yield with spectral indices of the hyperspectral active sensor (HAS) (calculated as normalized difference indices) for barley cultivars subjected to mild and severe drought stress in 2012 and 2013.

Spectral indices of HAS	Parameters	25 May 2012 Mild stress	18 June 2012 Severe stress	21 June 2012 Severe stress	18 June 2013 Mild stress	19 June 2013 Mild stress	19 June 2013 Severe stress
730.670	NRCT	0.49 ^{***}	0.02	0.00	0.54 ^{***}	0.62 ^{***}	0.03
	Grain yield	0.21 ^{***}	0.01	0.00	0.35 ^{***}	0.41 ^{***}	0.01
NDVI	NRCT	0.49 ^{***}	0.03	0.01	0.55 ^{***}	0.63 ^{***}	0.04
	Grain yield	0.21 ^{***}	0.00	0.01	0.38 ^{**}	0.42 ^{**}	0.11 [*]
NDVI1	NRCT	0.38 ^{***}	0.04	0.01	0.56 ^{***}	0.63 ^{***}	0.01
	Grain yield	0.18 ^{**}	0.00	0.01	0.37 ^{***}	0.43 ^{***}	0.10 [*]
760.730	NRCT	0.46 ^{***}	0.15 [*]	0.13 [*]	0.37 ^{***}	0.48 ^{***}	0.01
	Grain yield	0.19 ^{**}	0.11 [*]	0.14 [*]	0.43 ^{***}	0.20 ^{***}	0.21 ^{***}
780.510	NRCT	0.32 ^{***}	0.06	0.00	0.51 ^{***}	0.61 ^{***}	0.05
	Grain yield	0.20 ^{***}	0.00	0.00	0.36 ^{***}	0.44 ^{***}	0.17 ^{**}
850.560	NRCT	0.20 ^{**}	0.17 ^{**}	0.01	0.51 ^{***}	0.62 ^{***}	0.03
	Grain yield	0.07	0.03	0.06	0.38 ^{***}	0.50 ^{***}	0.01
810.740	NRCT	0.50 ^{***}	0.25 ^{***}	0.01	0.01	0.05	0.00
	Grain yield	0.10 [*]	0.12 ^{**}	0.05	0.00	0.20 ^{***}	0.03
NWI-1	NRCT	0.62 ^{***}	0.02	0.00	0.00	0.01	0.00
	Grain yield	0.19 ^{**}	0.01	0.00	0.05	0.00	0.02
NWI-3	NRCT	0.62 ^{***}	0.02	0.00	0.00	0.02	0.00
	Grain yield	0.18 ^{**}	0.00	0.00	0.05	0.00	0.02
NWI-4	NRCT	0.63 ^{***}	0.01	0.00	0.00	0.01	0.00
	Grain yield	0.19 ^{**}	0.01	0.01	0.06	0.00	0.03

* Statistically significant at $p \leq 0.05$.

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

Table 6
Coefficients of determination of linear regressions between reflectance bands and spectral indices obtained from three active sensors (Active flash sensor, AFS; Crop Circle, CC; GreenSeeker, GS) with the NRCT and grain yield of barley cultivars subjected to mild and severe drought stress in 2012 and 2013.

Drought stress levels	Date	Parameters	Active Flash Sensor						Crop Circle						GreenSeeker		
			AFS_730	AFS_760	AFS_970	AFS_900	AFS760_730	AFS_NW1	CC_760	CC_670	CC_730	CC730_670	CC760_730	CC_NDVI	GS_774	GS_656	GS_NDVI
Mild drought stress	25 May 2012	NRCT	0.04	0.40 ^{***}	0.39 ^{***}	0.31 ^{***}	0.62 ^{***}	0.51 ^{***}	0.60 ^{***}	0.60 ^{***}	0.17 ^{**}	0.72 ^{***}	0.70 ^{***}	0.73 ^{***}	0.38 ^{***}	0.42 ^{***}	0.57 ^{***}
		Grain yield	0.07	0.02	0.02	0.01	0.15 ^{**}	0.28 ^{***}	0.05	0.25 ^{***}	0.12 ^{**}	0.17 ^{**}	0.20 ^{***}	0.18 ^{**}	0.02	0.27 ^{***}	0.20 ^{***}
Severe drought stress	18 June 2012	NRCT	0.06	0.03	0.02	0.02	0.18 ^{**}	0.04	0.02	0.18 ^{**}	0.08	0.01	0.17 ^{**}	0.14 ^{**}	0.05	0.06	0.01
		Grain yield	0.36 ^{***}	0.32 ^{***}	0.27 ^{***}	0.29 ^{***}	0.07	0.01	0.20 ^{**}	0.26 ^{**}	0.04	0.10 [*]	0.32 ^{***}	0.03	0.31 ^{***}	0.16 ^{**}	0.16 ^{**}
Severe drought stress	21 June 2012	NRCT	0.08	0.04	0.02	0.03	0.16 ^{**}	0.08	0.03	0.26 ^{**}	0.13 ^{**}	0.00	0.20 ^{***}	0.07	0.06	0.13 ^{**}	0.00
		Grain yield	0.27 ^{***}	0.18 ^{**}	0.12 ^{**}	0.15 ^{**}	0.24 ^{**}	0.19 ^{**}	0.16 ^{**}	0.25 ^{***}	0.32 ^{***}	0.12 ^{**}	0.22 ^{**}	0.01	0.24 ^{***}	0.11 [*]	0.03
Mild drought stress	18 June 2013	NRCT	0.02	0.14 ^{**}	0.14 ^{**}	0.11 [*]	0.50 ^{***}	0.44 ^{***}	0.24 ^{***}	0.64 ^{***}	0.00	0.49 ^{***}	0.53 ^{***}	0.50 ^{***}	0.16 ^{***}	0.43 ^{***}	0.38 ^{***}
		Grain yield	0.25 ^{***}	0.43 ^{***}	0.49 ^{***}	0.47 ^{***}	0.60 ^{***}	0.40 ^{***}	0.44 ^{***}	0.38 ^{***}	0.10 ^{**}	0.46 ^{***}	0.48 ^{***}	0.49 ^{***}	0.52 ^{***}	0.26 ^{***}	0.56 ^{***}
Mild drought stress	19 June 2013	NRCT	0.10 [*]	0.28 ^{**}	0.32 ^{**}	0.29 ^{***}	0.58 ^{***}	0.54 ^{***}	0.32 ^{**}	0.70 ^{***}	0.00	0.56 ^{***}	0.60 ^{***}	0.58 ^{***}	0.28 ^{***}	0.36 ^{***}	0.43 ^{***}
		Grain yield	0.26 ^{***}	0.46 ^{***}	0.52 ^{***}	0.50 ^{***}	0.61 ^{***}	0.49 ^{***}	0.50 ^{***}	0.40 ^{***}	0.07 [*]	0.49 ^{***}	0.49 ^{***}	0.52 ^{***}	0.48 ^{***}	0.20	0.42 ^{***}
Severe drought stress	19 June 2013	NRCT	0.00	0.00	0.01	0.01	0.13 [*]	0.01	0.10 [*]	0.08	0.00	0.07	0.32 ^{***}	0.21 ^{***}	0.01	0.04	0.06
		Grain yield	0.02	0.00	0.00	0.00	0.13 [*]	0.03	0.00	0.27 ^{***}	0.06	0.08 [*]	0.10 [*]	0.12 [*]	0.00	0.07	0.04

* Statistically significant at $p \leq 0.05$.

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

Table 7
Calibration (R^2 cal and RMSEC) and 12-fold cross-validation (R^2 val and RMSEV) statistics of partial least square regression models of ten spectral indices of the hyperspectral passive reflectance sensor (listed in Table 1), seven spectral indices of the hyperspectral active reflectance sensor (listed in Table 2, except three normalized water indices) and of spectral indices and reflectance bands of the Active flash sensor, the Crop Circle and the GreenSeeker (listed in Table 6) with NRCT and grain yield in 2012 and 2013.

Drought stress levels	Date	Parameters	Hyperspectral passive sensor				Hyperspectral active sensor				Active flash sensor				Crop Circle				GreenSeeker			
			R^2 cal	RMSEC	R^2 val	RMSEV	R^2 cal	RMSEC	R^2 val	RMSEV	R^2 cal	RMSEC	R^2 val	RMSEV	R^2 cal	RMSEC	R^2 val	RMSEV	R^2 cal	RMSEC	R^2 val	RMSEV
Mild drought stress	25 May 2012	NRCT	0.72 ^{***}	0.007	0.63 ^{***}	0.008	0.64 ^{***}	0.008	0.58 ^{***}	0.009	0.66 ^{***}	0.008	0.56 ^{***}	0.008	0.77 ^{***}	0.006	0.74 ^{***}	0.007	0.57 ^{***}	0.009	0.53 ^{***}	0.009
		Grain yield	0.31 ^{***}	0.813	0.14 ^{**}	0.905	0.14 ^{**}	0.902	0.08 [*]	0.937	0.33 ^{***}	0.799	0.26 ^{***}	0.841	0.27 ^{***}	0.830	0.15 ^{**}	0.896	0.28 ^{***}	0.829	0.25 ^{**}	0.843
Severe drought stress	18 June 2012	NRCT	0.57 ^{***}	0.008	0.44 ^{***}	0.009	0.23 ^{**}	0.010	0.05	0.012	0.24 ^{**}	0.011	0.05	0.012	0.59 ^{***}	0.011	0.53 ^{***}	0.012	0.07	0.011	0.00	0.012
		Grain yield	0.69 ^{***}	0.404	0.60 ^{***}	0.501	0.30 ^{**}	0.690	0.08 [*]	0.801	0.61 ^{***}	0.492	0.43 ^{***}	0.594	0.53 ^{***}	1.147	0.50 ^{***}	1.167	0.33 ^{***}	0.650	0.26 ^{***}	0.680
Severe drought stress	21 June 2012	NRCT	0.45 ^{***}	0.003	0.25 ^{***}	0.008	0.16 ^{**}	0.009	0.12 ^{***}	0.01	0.41 ^{***}	0.007	0.29 ^{***}	0.008	0.26 ^{***}	0.008	0.14 ^{**}	0.009	0.13 ^{**}	0.009	0.04	0.010
		Grain yield	0.63 ^{***}	0.48	0.50 ^{***}	0.511	0.31 ^{**}	0.654	0.05	0.766	0.51 ^{***}	0.551	0.31 ^{**}	0.655	0.47 ^{***}	0.570	0.36 ^{***}	0.630	0.25 ^{***}	0.682	0.11 [*]	0.742
Mild drought stress	18 June 2013	NRCT	0.76 ^{***}	0.008	0.64 ^{***}	0.010	0.58 ^{***}	0.011	0.53 ^{***}	0.012	0.64 ^{***}	0.010	0.54 ^{***}	0.012	0.59 ^{***}	0.011	0.53 ^{***}	0.012	0.57 ^{***}	0.009	0.50 ^{***}	0.009
		Grain yield	0.62 ^{***}	0.992	0.58 ^{***}	1.034	0.41 ^{**}	1.269	0.39 ^{***}	1.293	0.67 ^{***}	0.951	0.62 ^{***}	1.012	0.53 ^{***}	1.137	0.50 ^{***}	1.167	0.57 ^{***}	1.081	0.54 ^{***}	1.123
Mild drought stress	19 June 2013	NRCT	0.75 ^{***}	0.008	0.70 ^{***}	0.008	0.64 ^{***}	0.009	0.62 ^{***}	0.01	0.62 ^{***}	0.009	0.55 ^{***}	0.010	0.71 ^{***}	0.008	0.67 ^{***}	0.009	0.43 ^{***}	0.011	0.41 ^{**}	0.012
		Grain yield	0.59 ^{***}	1.03	0.54 ^{***}	1.092	0.41 ^{**}	1.270	0.35 ^{**}	1.335	0.67 ^{***}	0.946	0.60 ^{***}	1.053	0.57 ^{***}	1.058	0.51 ^{***}	1.153	0.50 ^{***}	1.174	0.41 ^{**}	1.260
Severe drought stress	19 June 2013	NRCT	0.36 ^{***}	0.003	0.11 [*]	0.004	0.65 ^{***}	0.006	0.16 [*]	0.009	0.41 ^{***}	0.007	0.29 ^{***}	0.008	0.43 ^{***}	0.003	0.17 ^{**}	0.004	0.05	0.004	0.04	0.004
		Grain yield	0.27 ^{***}	0.932	0.11 [*]	1.029	0.26 ^{***}	1.029	0.17 ^{**}	1.088	0.21 ^{***}	0.907	0.04	1.108	0.25 ^{***}	0.945	0.16 ^{**}	0.997	0.08 [*]	1.049	0.04	1.066

* Statistically significant at $p \leq 0.05$.

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

Table 8
Partial least square models of ten spectral indices of the hyperspectral passive reflectance sensor (listed in Table 1), seven spectral indices of the hyperspectral active reflectance sensor (listed in Table 2, except three normalized water indices) and of spectral indices and reflectance bands of the Active flash sensor, the Crop Circle and the GreenSeeker (listed in Table 6) with NRCT and grain yield were calibrated on 18 June 2012 respectively on 18 June 2013. Calibration functions were validated with independent data measured on the indicated prediction dates. Coefficients of determination, slopes and intercepts of these linear validation functions between observed and predicted values of NRCT and grain yield are shown. Significance levels are shown.

Drought stress level	Calibrated date	Predicted date	Parameters	Hyperspectral passive sensor		Hyperspectral active sensor		Active flash sensor		Crop Circle		GreenSeeker		
				R ²	a	b	R ²	a	b	R ²	a	b	R ²	a
Mild drought stress in 2013	18 June 2013	19 June 2013	NRCT	0.63***	0.34	0.19	0.63***	0.28	0.45	0.51***	0.32	0.43***	0.74	0.19
Mild drought stress in 2012	18 June 2013	25 May 2012	Grain yield	0.51***	0.22	61.52	0.44***	0.38	32.33	0.49***	0.52	0.42***	0.55	29.42
Severe drought stress in 2012	18 June 2012	21 June 2012	NRCT	0.55***	0.77	0.32	0.63***	0.24	0.33	0.70***	0.49	0.54***	0.52	0.35
Severe drought stress in 2012	18 June 2012	19 June 2013	Grain yield	0.11*	0.45	25.24	0.14*	0.09	81.01	0.18	0.35	0.20***	0.36	36.02
Severe drought stress 2013	18 June 2012	18 June 2012	NRCT	0.05	0.10	0.54	0.03	0.08	0.54	0.08	0.10	0.06	-0.05	0.59
Mild and severe drought stress	18 June 2013	18 June 2012	Grain yield	0.52***	0.32	21.34	0.01	0.00	55.02	0.13	0.03	0.04	0.05	53.67
			NRCT	0.29***	0.24	0.43	0.01	0.04	0.54	0.22	0.17	0.00	-0.01	0.57
			Grain yield	0.42***	0.26	43.66	0.15**	0.13	43.2	0.07	0.06	0.03	0.06	45.79
			NRCT	0.21***	0.27	0.21	0.64***	0.54	0.27	0.59	0.35	0.43***	-0.07	0.58
			Grain yield	0.22***	0.06	40.57	0.02	0.02	60.63	0.53***	0.15	0.56***	-0.17	62.99

a is the slope of the line.

b is the intercept (where the line crosses the y-axis).

* Statistically significant at $p \leq 0.05$.

** Statistically significant at $p \leq 0.01$.

*** Statistically significant at $p \leq 0.001$.

The shapes of the contour maps of matrices from the hyperspectral active sensor showed more diffuse edges than those from the hyperspectral passive sensor (Figs. 1 and 2); this may be caused by vibrations of the carrier vehicle, which possibly affects the optics and spectral light allocation on the diode array of the active spectrometer.

On all six measurement days in the mild and severe drought stressed trials, significant relationships between three normalized water indices and the NRCT and the grain yield were found for the hyperspectral passive sensor (Table 4). By contrast, for the hyperspectral active sensor, on five out of six measurement days no significant relationships were found between three normalized water indices and the NRCT and the grain yield (Table 5). Generally, the three normalized water indices of the hyperspectral passive sensor indicated better relationships with the NRCT and the grain yield than the AFS_NWI-1 of the active flash sensor. The generally poorer performance of the active sensors may be caused by the lower penetration depth of the artificial light compared to natural sunlight. Less information on biomass, canopy structure and water status can be collected. These results agree with Jasper et al. (2009), who reported that the artificial light sources of active sensors penetrate less deeply into crop canopies compared with solar radiation. The impact of the canopy structure on the incoming light may, therefore, be different for active and passive sensors and may affect the suitability of spectral indices. Similarly, Winterhalter et al. (2013) reported that active reflectance sensors have difficulties in detecting nitrogen uptake in maize at large sensor-to-target distances because their light source is weaker than natural sunlight.

The contour maps of matrices of the hyperspectral passive and active sensor, as well as the spectral indices and spectral bands of the other three active sensors, showed closer relationships to the grain yield for the mild drought stress trial than for the severe drought stress trial. These results are in agreement with Lobos et al. (2014), who found that the normalized water index NWI-3: $(R_{970} - R_{920}) / (R_{970} + R_{920})$ and the normalized difference vegetation index NDVI: $(R_{830} - R_{660}) / (R_{830} + R_{660})$ indicated closer relationships (R^2 values of 0.66 and 0.62) with the grain yield of wheat cultivars under mild drought stress compared with severe drought stress (R^2 values of 0.58 and 0.40). Additionally, Gutierrez et al. (2010) found that the normalized water indices NWI-1: $(R_{970} - R_{900}) / (R_{970} + R_{900})$ and NWI-3: $(R_{970} - R_{920}) / (R_{970} + R_{920})$ of a hyperspectral passive sensor presented significant relationships with the grain yield of wheat genotypes under drought stress.

The active flash sensor AFS index 760_730, the Crop Circle CC index 760_730 and the hyperspectral passive sensor HPS index 760_730 presented significant relationships with the NRCT and the grain yield, and the coefficients of determination of each sensor were 0.13*–0.62***, 0.10*–0.72*** and 0.16**–0.56***, respectively. The Green Seeker (GS_NDVI), Crop Circle (CC_NDVI) and the hyperspectral active sensor (HAC_NDVI) showed weaker relationships with the NRCT and the grain yield under severe drought stress compared with the indices HPS_NDVI and HPS_NDVI-1 of the hyperspectral passive sensor. Perhaps one reason for this finding is that the GS_NDVI, CC_NDVI and HAS_NDVI were affected by the ambient air temperature (Kipp et al., 2014).

The relationships between the spectral indices and single reflectance bands of the five sensor systems used in this study with the NRCT and grain yield of barley cultivars were weaker in the trials under severe drought stress in 2013 than in 2012. Due to the higher temperatures, the plants were exposed to more severe drought stress in 2013 compared with 2012 and the plants become more dehydrated under severe drought stress compare to the plants under mild drought stress. In 2013 the mean values of the NRCT were higher (0.83) than in 2012 (0.48 and 0.65), whereas the grain yield was lower in 2013 (2.95 t/ha) than in 2012 (4.25 t/ha).

Spectral indices and spectral bands of the five spectral sensor systems generally showed weaker relationships with the grain yield under mild drought stress on 25 May 2012 at the stem elongation stage (BBCH 32) than during the heading phase on 18 June 2013 (BBCH 55) and 19 June 2013 (BBCH 56). Our results are in agreement with Gutierrez et al. (2010), who found significant relationships between the normalized water indices NWI-1 and NWI-3 and the grain yield of spring wheat from heading to the grain filling stages (R varied from -0.40^{**} to -0.67^{**}) under drought stress in three years. In a study by Mandal et al. (2007), the normalized difference vegetation index (NDVI), the green NDVI, and the soil adjusted vegetation index (SAVI) measured from booting to the anthesis stage were correlated with the grain yield of sorghum and the R^2 values ranged from 0.44 to 0.62. Four indices: the NDVI $(R_{810} - R_{680}) / (R_{810} + R_{680})$; the ratio vegetation index RVI (R_{810} / R_{680}) ; the GNDVI $(R_{810} - R_{560}) / (R_{810} + R_{560})$; and the green ratio vegetation index GRVI (R_{810} / R_{560}) showed good and significant relationships with the grain yield of wheat cultivars from booting until the mature growth stages under different nitrogen fertiliser levels (Xue et al., 2007).

To the best of our knowledge, no information about the relationship between spectral indices or reflectance bands and the NRCT was found. Only a small number of studies exist using solely passive reflectance sensors for detecting the canopy temperature or the CWSI (Gutierrez et al., 2010; Winterhalter et al., 2011; Zarco-Tejada et al., 2013). In this study the capability of both passive and active reflectance sensors to assess the NRCT, an indicator of drought stress, is shown.

Our results show significant relationships of ten spectral indices with the NRCT and grain yield, which were derived from NIR bands such as the normalized water index, or by combining the VIS and NIR bands such as the normalized difference vegetation index. The indices measured on five days under mild and severe drought stress by the hyperspectral passive sensor (Table 4) and by four active sensors under mild drought stress (Table 5) presented generally significant relationships with the NRCT.

Zarco-Tejada et al. (2013) found that the normalized photochemical reflectance index $PRI_{norm} (R_{570} - R_{531}) / (R_{570} + R_{531}) / (R_{800} - R_{670}) / (R_{800} + R_{670})^{0.5} * (R_{700} / R_{670})$ was better related ($R^2 = 0.79$) to the CWSI measured in a vineyard than the normalized difference vegetation index NDVI $(R_{800} - R_{670}) / (R_{800} + R_{670})$ ($R^2 = 0.3$ ns).

Compared with spectral indices, models based on a partial least square regression offered an improvement for assessing the NRCT and the grain yield. This is shown by the improvement of the coefficients of determination in Tables 4–6, and the results of the cross validation in Table 7.

Linear models, calibrated based on datasets from 18 June 2012 and 18 June 2013, were validated based on measurements in other years under mild and severe drought stress. Only models from the passive sensor, but not from the four active sensors, presented significant relationships between the observed and predicted grain yield under mild and severe drought stress. The slopes of the validation regression lines were generally less steep for the passive and active sensors than the optimum slope one.

Through all RMSE for predicting grain yield of active and passive sensors in Table 7, the RMSE is smaller than the range of cultivar yields under mild and severe drought stress. The range of cultivar yields was 1.67 under mild drought stress and 1.69 t/ha under severe drought stress in 2012. In 2013 the range of cultivar yields were 2.61 under mild drought stress and 2.52 t/ha under severe drought stress. The highest RMSE was 1.335 t/ha for the validation of the hyperspectral active sensor (mild drought stress, 19.6.2013). Also through all RMSE for predicting grain yield of active and passive sensors sometimes RMSE is smaller than differences in yields of single barley cultivars. For example, RMSE of hyperspectral

passive reflectance sensor in severe drought stress at 18.06.2012 is 0.501 t/ha (Table 7). According to this RMSE the hyperspectral passive reflectance sensor does not enable to differentiate between Beatrix and Djamila (diff. 0.06 t/ha), but enables to differentiate between Beatrix and Ursa (diff. 0.83 t/ha), Barke (diff. 0.89 t/ha), Heils Franken (diff. 1.28 t/ha) and Bavaria (diff. 1.69 t/ha). It can be concluded, that a differentiation of high and low-yielding cultivars but not of single cultivars showing similar yields is technically feasible with all tested sensors. At the most of measurement dates the RMSE of the passive and active sensors is smaller than the RMSE of the active reflectance sensors.

In conclusion, the results show that differences in the NRCT and the grain yield of barley cultivars can reliably be detected using spectral measurements from passive and active reflectance sensors under mild and severe stress. The spectral indices and single reflectance bands obtained from bands of passive and active sensors in mild drought stress trials yielded closer relationships with the NRCT and grain yield than those under severe drought stress. Partial least square regression can improve the assessment of the NRCT and the grain yield of barley cultivars compared to the spectral indices and reflectance bands.

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