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# Plant Disease Detection by Imaging Sensors – Parallels and Specific Demands for Precision Agriculture and Plant Phenotyping



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Precision agriculture and plant phenotyping are information- and technology-based domains with specific demands and challenges for the diagnosis and detection of plant diseases (Fig. 1). Precision agriculture is a crop management system based on the spatial and temporal variability in crop and soil factors within a field (Stafford 2000). This system aims to attain real-time, robust mapping systems for crop, soil, and environment variables to facilitate a management decision (Fig. 1). On the contrary, traditional agricultural management practices assume that parameters in crop fields are homogeneous, thereby leading to pesticide application and crop health management that is not clearly related to the existing disease management situation (Hillnhütter and Mahlein 2008). Considering that the occurrence of plant disease depends on specific environmental factors and that diseases often exhibit a heterogeneous distribution in fields, optical sensing techniques are useful in identifying primary disease foci and areas differing in disease severity in fields (Franke and Menz 2007). In combination with advanced methods of data analysis, these techniques can be used for targeted pest management programs in sustainable crop production. Site-specific and targeted applications of pesticides according to precision crop protection strategies results in potential reduction in pesticide use and can thus reduce the economic expense and ecological impact in agricultural crop production systems (Gebbers and Adamchuk 2010).

Whereas precision agriculture is aimed at examining spatial heterogeneities within crop stands, plant phenotyping assesses the appearance and performance of a genotype under distinct environmental conditions. During the plant breeding process, a large number of different genotypes are tested for disease and abiotic stress resistance, yield, produce quality, and many secondary traits (Fiorani and Schurr 2013). In disease resistance breeding, host pathogen interactions and the susceptibility of the breeding material must be evaluated efficiently. The target plant material must be tested by genotyping and phenotyping in an exhaustive and time consuming manner (Fig. 1). These investigations include manifold steps:

evaluations in different environments, under controlled and in natural field conditions as well as on single plants, individual organs, and even entire field canopies. In particular, plant phenotyping is labor and time consuming and as such is rather costly. Recently, phenotyping was used often as a synonym for noninvasive imaging and sensor-based analysis of anatomical, physiological, and biochemical plant properties (Guo and Zhu 2014; Walter et al. 2015).

Both precision agriculture and plant phenotyping have specific needs and challenges in regards to plant disease detection. In order to obtain objective and reliable automated diagnosis and detection of plant diseases, new approaches must be introduced and incorporated into traditional monitoring and rating systems. Optical sensors are promising tools for noninvasive disease detection and diagnosis (Fig. 2). There are an increasing number of imaging and noninvasive sensors available that can support diagnosis and plant disease detection. The progress in sensor and information technologies together with the expansion of geographic information systems opens new opportunities for precision agriculture and plant phenotyping.

However, a common drawback or limitation of optical sensors is the large amount and the complexity of the data collected. To be able to effectively utilize optical sensor data for diagnosis and disease detection, advanced data analysis and statistical methods are essential. The data has to include several important factors: (i) the detection of a disease at early points in time, (ii) the differentiation among different diseases, (iii) the separation of diseases caused by abiotic stresses, and (iv) the quantification of disease severity. These parameters need to be assessed with a level higher or equivalent to the accuracy attained with standard assessment methods and with a shorter computation time. In this context, data mining methods are constantly being introduced into plant science and are becoming a key technology. The current article will present recent results from the research community, indicate state-of-the-art methods, and discuss the future of sensor imaging and data analysis for the diagnosis and detection of diseases in crop production.

## Present and Future Trends in Plant Disease Detection

Accurate estimates of disease incidence, disease severity, and the negative effects of diseases on the quality and quantity of agricultural produce are important for field crop, horticulture, plant breeding, and for improving fungicide efficacy as well as for basic and applied plant research. Reliable and timely assessments of plant disease

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occurrence and spread are, in particular, the basis for planning targeted plant protection activities in field or greenhouse production and to forecast temporal and spatial disease spread in specific growing regions. Common methods for the diagnosis and detection of plant diseases include visual plant disease estimation by human raters, microscopic evaluation of morphology features to identify pathogens, as well as molecular, serological, and microbiological diagnostic techniques (Bock et al. 2010; Nutter 2001).

Microscopic methods utilize pathogen morphology (spores, mycelium, and fruiting bodies) for disease diagnosis. Speciation keys and identification schemes are available. Microbiological methods exist for pathogen isolation on selective, artificial media and molecular and serological methods can be readily applied for the diagnosis and detection of a pathogen. These methods are used by plant protection services and in both research and industrial development. During the recent decades, DNA-based and serological methods have revolutionized the identification and quantification of pathogens and diseases (Bock et al. 2010; Martinelli et al. 2014; Ward et al. 2004). Highly specific and rapid tests for growers and quarantine inspectors have been available since 1999. These tests can be applied directly at a field site, in a greenhouse, or in the production chain to assess and identify relevant plant pathogens. For example, a lateral flow-through version of ELISA can be used to detect *Phytophthora infestans* (late blight), *Ralstonia solanacearum* (Brown rot), *Erwinia amylovora* (Fire blight), *Pepino mosaic virus*, *Tomato mosaic virus*, *Potato virus Y*, and *Potato virus X* (Danks and Barker 2000). Using molecular and serological methods, pathogen strains or isolates can be identified that differ in their virulence or are resistant to a specific pesticide. However, the extent of pathogen development in a plant is not always correlated positively with disease intensity, because the amount of pathogen biomass is not necessarily proportional to the extent of visible disease symptoms (Nutter 2001).

Traditional, visual estimates identify a disease based on characteristic plant disease symptoms (e.g., lesions, blight, galls, tumors, cankers, wilts, rots, or damping-off) or visible signs of a pathogen (e.g., uredinospores of *Pucciniales*, mycelium or conidia of *Erysiphales*). Visual estimation is performed by trained experts and has been the subject of intensive research and investigation. Reliability and accuracy are benchmarks for the performance of visual assessment ratings. Visual estimation has become more accurate and reliable due to the availability of detailed guidelines and standards used for assessment training (Bock et al. 2010; Nutter 2001). Nevertheless,

visual estimation is always subject to an individual's experience and can be affected by temporal variation. This variation causes significant interrater variability and changes in interrater repeatability (Bock et al. 2008, 2010; Newton and Hackett 1994; Nutter 2001; Nutter et al. 1993; Steddom et al. 2005).

These time-consuming methods demand experienced individuals with well-developed skills in diagnosis and disease detection and are thus subject to human bias. In 1936, Riker and Riker emphasized the difficulties in diagnosing and detecting plant diseases. They gave an overview of the strengths and limitations of existing methods and concluded that:

“We need better methods for diagnosis; none of the methods given are to be considered as ‘standardized’. To think of them in such a way would put an end to efforts of improvement. They are useful only until better procedures can be developed.” (Riker and Riker 1936)

What Riker and Riker stated in 1936 is still valid and pertinent today—future-oriented plant protection needs new and innovative techniques to address forthcoming challenges and trends in agricultural production that require more precision than ever before due to consumer oversight. New and automated methods with high sensitivity, specificity, and reliability are therefore necessary to improve disease detection over and beyond that of visual estimation processes.

Intensive research has recently identified new, sensor-based methods for the detection, identification, and quantification of plant diseases (Hillnhütter et al. 2010; Mahlein et al. 2012a; Sankaran et al. 2010; West et al. 2003, 2010). These sensors assess the optical properties of plants within different regions of the electromagnetic spectrum and are able to utilize information beyond the visible range (Fig. 2). They enable the detection of early changes in plant physiology due to biotic stresses, because disease can cause modifications in tissue color, leaf shape, transpiration rate, canopy morphology, and plant density as well as variation in the interaction of solar radiation with plants (West et al. 2010). Currently the most promising techniques are sensors that measure reflectance, temperature, or fluorescence (Chaerle and Van der Straeten 2000; Mahlein et al. 2012a; Sankaran et al. 2010; West et al. 2003). Most of these spectral and thermal sensors were originally developed for the military, earth remote sensing, from satellites and aircraft, and in some cases for industrial use. According to Moore (1979), remote sensing is the use of reflected and emitted energy to measure the physical properties

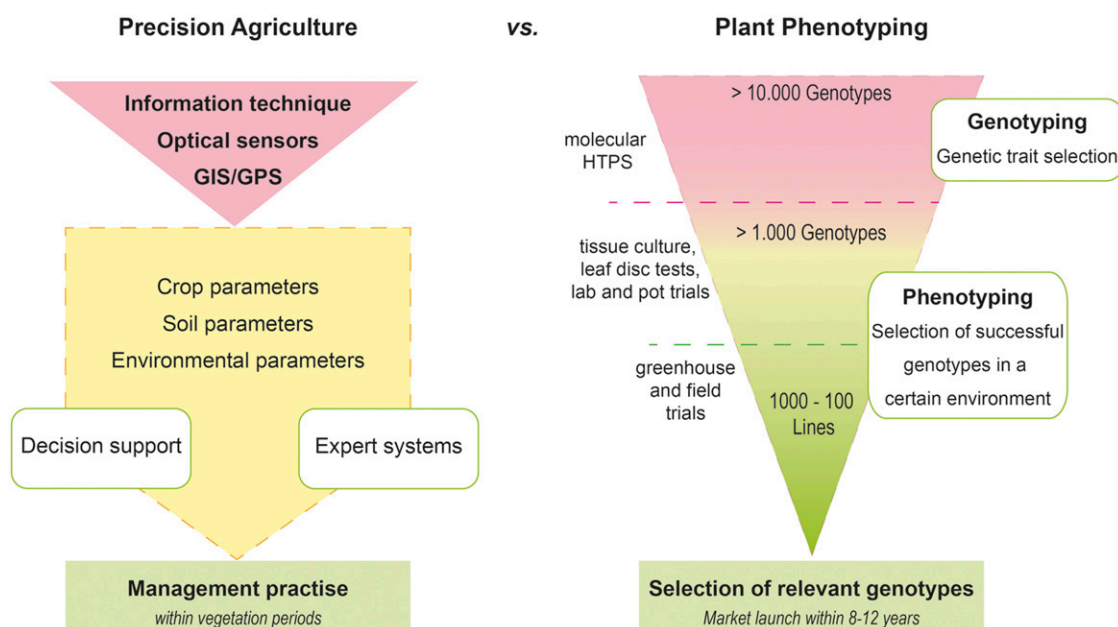


Fig. 1. Schematic diagrams of workflows and parameters in precision agriculture (left) and plant phenotyping (right).

of distant objects and their surroundings. The first remote sensing technique was aerial photography, invented with panchromatic films in World War I and with color infrared films for camouflage detection in World War II (Moore 1979). In the 1950s, multispectral systems, thermal infrared, radar, and sonar were invented and adapted for remote sensing (Fischer 1975). The first multispectral sensors for remote sensing by air- and spacecraft were tested in 1964. Hyperspectral imaging spectroscopy followed in the early 1980s (Campbell 2007). In plant sciences, remote sensing is a method used to obtain information from plants or crops without direct contact or invasive manipulation. The concept has been recently enlarged by proximal, close-range or small-scale sensing of plant material (Chaerle and Van der Straeten 2000; Mahlein et al. 2012b; Oerke et al. 2014). These sensors can be installed on multiple platforms (Fig. 2; digital microscopes, tractors, carriers, robots, high-throughput platforms, UAVs, zeppelins, aircrafts, satellites, etc.) or stationary sensors can be placed at strategic points.

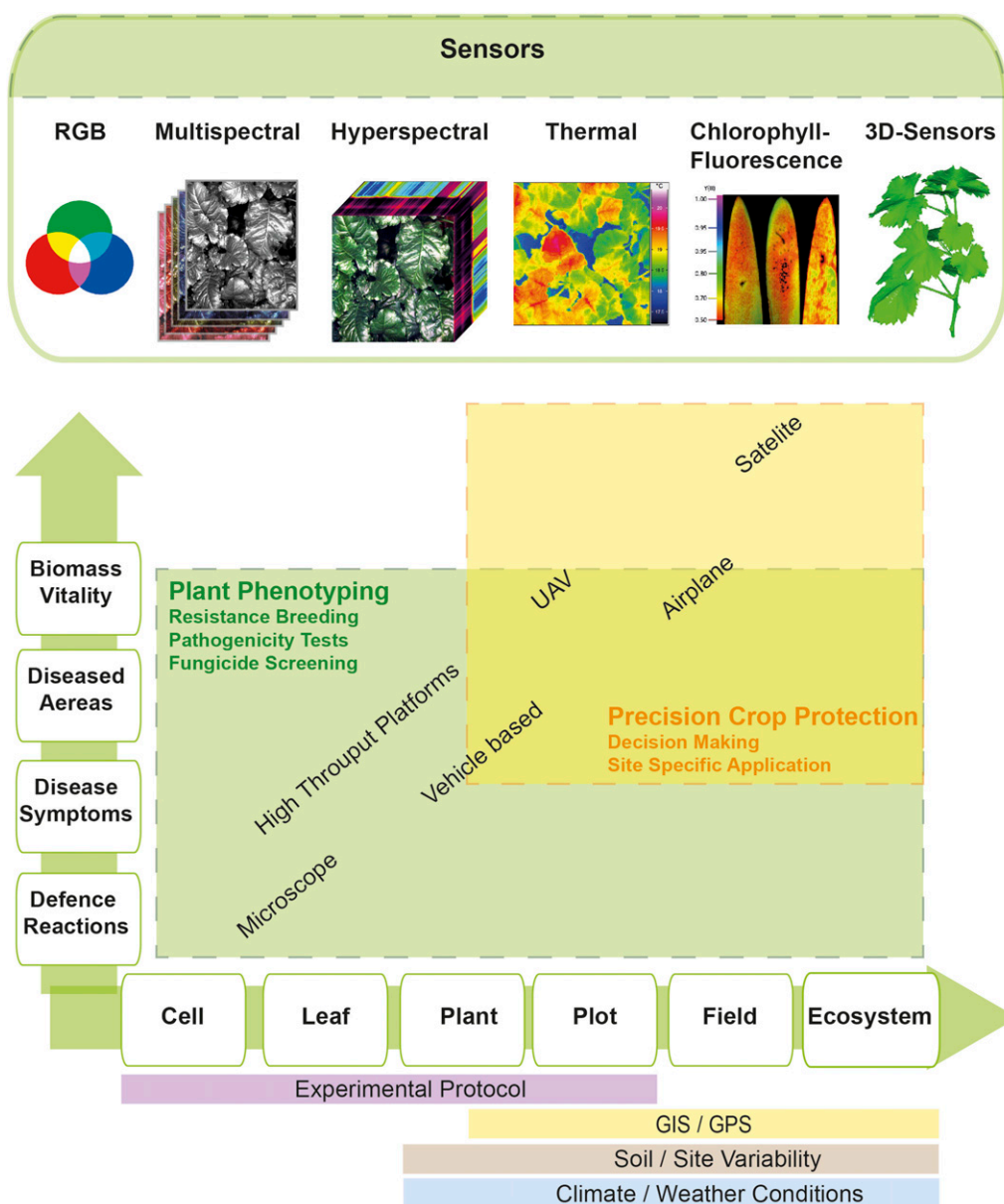
In terms of sensor use for disease detection in precision agriculture or plant phenotyping, individual specifications and conditions have to be considered (Figs. 1 and 2).

## New Methodology

A large number of more recent developments in different pathosystems using different types of highly sensitive sensors and multiple data analysis pipelines have been published and a few examples are summarized in Table 1 and illustrated in Figure 2. Extensive reviews give more detailed information on the different sensor systems listed (Bock et al. 2010; Mahlein et al. 2012a; Sankaran et al. 2010; West et al. 2003).

## Optical Sensors for Plant Disease Detection

**RGB-imaging.** Digital photographic images are important tools in plant pathology for assessing plant health. Digital cameras are easy to handle and are a simple source of RGB (red, green, and blue) digital images for disease detection, identification, and quantification. The technical parameters of these simple, handheld devices such as the light sensitivity of the photo sensor, spatial resolution, or optical and digital focus have improved significantly every year. Today, nearly every person, farmer or phytopathologist, carries modern and sophisticated digital camera sensors together with a mobile phone or tablet computer. Video cameras or scanners are alternative



**Fig. 2.** Overview of current sensor technologies used for the automated detection and identification of host-plant interactions. These sensors can be implemented in precision agriculture applications and plant phenotyping on different scales from single cells to entire ecosystems. Depending on the scale, different platforms can be operated and consequently different plant parameters can be observed (Oerke et al. 2014, modified).



methods for assessing digital images of different plant organs, from roots to inflorescences. RGB sensors are used on every scale of resolution for monitoring plants during the growing season.

RGB-color images with the red, green, and blue channels have been used to detect biotic stress in plants (Bock et al. 2010, Table 1). Along with color information in the RGB, LAB (L for lightness and A and B for the color-opponent dimensions, based on nonlinearly compressed coordinates), YCBCR (color compression scheme Y is the luma component and CB and CR are the blue-difference and red-difference chroma components, respectively), or HSV (hue, saturation, value) color space, the spatial information provides important characteristics of plant diseases (Bock et al. 2010). Furthermore, color, gray levels, texture, dispersion, connectivity, and shape parameters can be defined as features for the detection and identification of disease symptoms in plants (Camargo and Smith 2009; Neumann et al. 2014). Several research groups have used pattern recognition methods and machine learning to detect and to identify plant diseases from RGB images (Table 1; Camargo and Smith 2009; Neumann et al. 2014). In addition, systematic selection of relevant features from the RGB images increase classification accuracies (Behmann et al. 2014). Digital image analysis is a well-established technology used in plant disease assessment. Several software packages, such as ASSESS 2.0, "Leaf Doctor," Scion Image software, and custom-made modules are available (Bock et al. 2010; Pethybridge and Nelson 2015; Tucker and Chakraborty 1997; Wijekoon et al. 2008; <http://www.plant-image-analysis.org/>). In ASSESS 2.0, the color distribution of the images is analyzed in histograms, which are the basis for subsequent thresholding. The parameters for healthy and diseased areas can be adjusted by the user in a well-organized graphical user interface. In addition, disease severity can be extracted as diseased pixels or as a percentage after the

background is masked from the object of interest. ASSESS 2.0 is very practical for the evaluation of disease severity on single leaves and well-arranged images. Special attention must be given to the image acquisition step. Uniform focus, sharpness, and illumination are crucial for high accuracy and reliable results from automated image analysis. Furthermore, under natural conditions, the imaging angle (leaf orientation) and distance between the object and the sensor (pixel size) are additional influences on the image quality. Difficulty in detection and low levels of accuracy are often the result of heterogenic conditions and low image quality. The most important principle for success is a sound standardized imaging procedure that yields repeatable results.

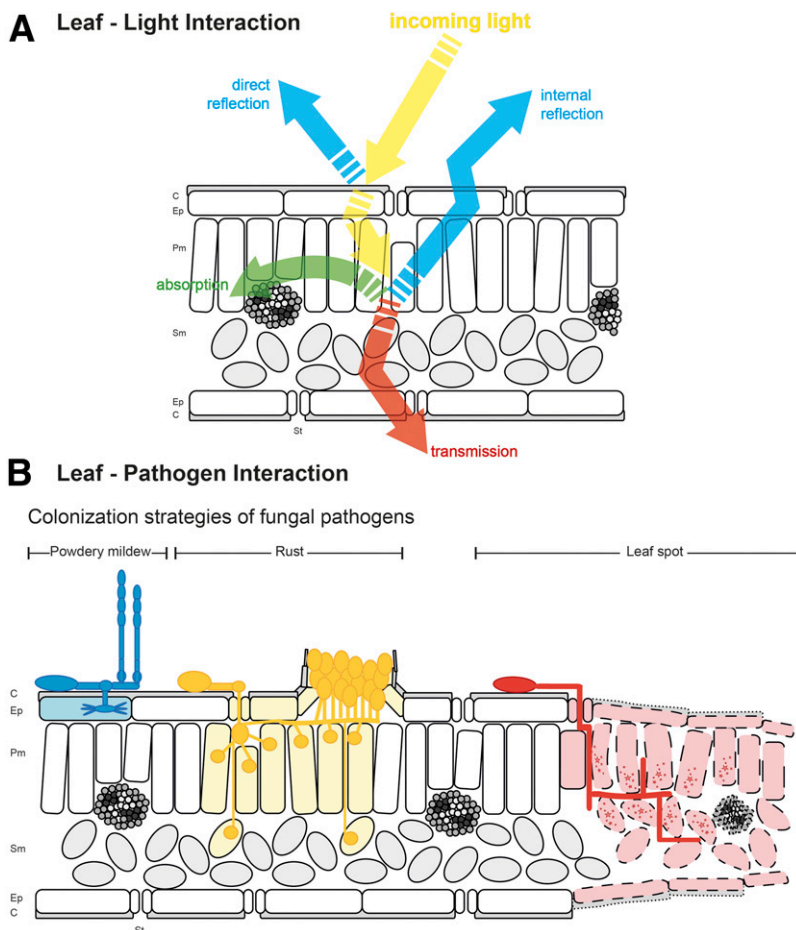
**Multi- and hyperspectral reflectance sensors.** Spectral sensors are generally categorized based on the spectral resolution (i.e., the number and width of measured wavebands), on their spatial scale, and on the type of detector, (i.e., imaging or nonimaging sensor systems). Multispectral sensors were the first spectral sensors invented. These sensors typically assess the spectral information of objects in several relatively broad wavebands. Multispectral imaging cameras may provide data, for instance, in the R, G, and B wavebands and in an additional near-infrared band. The evolution of modern hyperspectral sensors increased the complexity of the measured data by a spectral range of up to 350 to 2,500 nm and a possible narrow spectral resolution below 1 nm (Steiner et al. 2008). Contrary to nonimaging sensors, which average the spectral information over a certain area, hyperspectral imaging sensors provide spectral and spatial information for the imaged object. Hyperspectral data can be observed as huge matrices with spatial x- and y-axes, and the spectral information as reflectance intensity per waveband in the third dimension, z. The spatial resolution strongly depends on the distance between the sensor and the object. Thus, airborne or

**Table 1.** Examples of plant pathosystems and plant diseases assessed by optical sensors

Sensor	Crop	Disease / Pathogen	Reference
<b>RGB</b>	Cotton	Bacterial angular ( <i>Xanthomonas campestris</i> )	Camargo and Smith (2009)
		Ascochyta blight ( <i>Ascochyta gossypii</i> )	
	Sugar beet	Cercospora leaf spot ( <i>Cercospora beticola</i> ), Sugar beet rust ( <i>Uromyces betae</i> ), Ramularia leaf spot ( <i>Ramularia beticola</i> ), Phoma leaf spot ( <i>Phoma betae</i> ), bacterial leaf spot ( <i>Pseudomonas syringae</i> pv. <i>Aptata</i> )	Neumann et al. (2014)
	Grapefruit	Citrus canker ( <i>X. axonopodis</i> )	Bock et al. (2008)
	Tabaco	Anthrachnose ( <i>Colletotrichum destructivum</i> )	Wijekoon et al. (2008)
	Apple	Apple scab ( <i>Venturia inaequalis</i> )	Wijekoon et al. (2008)
	Canadian goldenrod	Rust ( <i>Coleosporium asterum</i> )	Wijekoon et al. (2008)
<b>Spectral sensors</b>	Barley	Net blotch ( <i>Pyrenophora teres</i> ), Brown rust ( <i>Puccinia hordei</i> ), Powdery mildew ( <i>Blumeria graminis hordei</i> )	Kuska et al. (2015); Wahabzada et al. (2015a)
	Wheat	Head blight ( <i>Fusarium graminearum</i> )	Bauriegel et al. (2011); Bravo et al. (2003); Huang et al. (2007); Moshou et al. (2004)
		Yellow rust ( <i>Puccinia striiformis</i> f. sp. <i>tritici</i> )	
	Sugar beet	Cercospora leaf spot ( <i>C. beticola</i> ), Sugar beet rust ( <i>U. betae</i> ), Powdery mildew ( <i>Erysiphe betae</i> ), Root rot ( <i>Rhizoctonia solani</i> ), Rhizomania ( <i>Beet necrotic yellow vein virus</i> )	Bergsträsser et al. (2015); Hillnhütter et al. (2011); Mahlein et al. (2010, 2012, 2013); Rumpf et al. (2010); Steddom et al. (2003, 2005)
	Tomato	Late blight ( <i>Phytophthora infestans</i> )	Wang et al. (2008)
	Apple	Apple scab ( <i>V. inaequalis</i> )	Delalieux et al. (2007)
	Tulip	Tulip breaking virus (TBV)	Polder et al. (2014)
	Sugar cane	Orange rust ( <i>Puccinia kuehnii</i> )	Apan et al. (2004)
	Sugar beet	Cercospora leaf spot ( <i>C. beticola</i> )	Chaerle et al. (2004)
	Cucumber	Downy mildew ( <i>Pseudoperonospora cubensis</i> ), Powdery mildew ( <i>Podosphaera xanthii</i> )	Berdugo et al. (2014); Oerke et al. (2006)
<b>Thermal sensors</b>	Apple	Apple scab ( <i>V. inaequalis</i> )	Oerke et al. (2011)
	Rosa	Downy mildew ( <i>Peronospora sparsa</i> )	Gomez (2014)
	Wheat	Leaf rust ( <i>Puccinia triticea</i> )	Bürling et al. (2011)
<b>Fluorescence imaging</b>		Powdery mildew ( <i>Blumeria graminis</i> f. sp. <i>tritici</i> )	
	Sugar beet	Cercospora leaf spot ( <i>C. beticola</i> )	Chaerle et al. (2004, 2007); Konanz et al. (2014)
	Bean	Common Bacterial Blight ( <i>Xanthomonas fuscans</i> subsp. <i>fuscans</i> )	Rousseau et al. (2013)
	Lettuce	Downy mildew ( <i>Bremia lactucae</i> )	Bauriegel et al. (2014); Brabandt et al. (2014)

spaceborne, far range systems have lower spatial resolution than near-range or microscopic systems. The spatial resolution has a strong influence on the detection of plant diseases or plant-pathogen interactions (Mahlein et al. 2012b; West et al. 2003). Airborne sensors are suitable for the detection of field patches that are diseased with soilborne pathogens (Hillnhütter et al. 2011) or in later stages of the diseases (Mahlein et al. 2012a; Mewes et al. 2011; Steddom et al. 2005). Sensors with a spatial resolution of approximately 1 m are hardly suitable for the detection of single symptoms or diseased leaves and plants; here, proximal sensor platforms are preferable (Oerke et al. 2014; West et al. 2003). Despite multiple studies (Table 1), the use of innovative hyperspectral imaging systems in plant pathology and in disease severity assessment is still in the research stage (Bock et al. 2010). The optical properties of leaves are characterized by (i) light transmission through a leaf, (ii) light that is absorbed by leaf chemicals (e.g., pigments, water, sugars, lignin, and amino acids), and (iii) light reflected from internal leaf structures or directly reflected from the leaf surface (Fig. 3A). Thus, reflectance of light from plants is a complex phenomenon dependent on multiple biophysical and biochemical interactions. The visible range (VIS 400 to 700 nm) is mainly influenced by leaf pigment content, the near-infrared reflectance (NIR 700 to 1,100 nm) depends on the leaf structure, internal scattering processes, and on the absorption by leaf water, and the short-wave infrared (1,100 to 2,500 nm) is influenced by the composition of leaf chemicals and water (Carter and Knapp 2001; Jacquemoud and Ustin 2001). Changes in reflectance due to plant pathogens and plant diseases can be explained by impairments in the leaf structure and chemical composition of the tissue during pathogenesis that is highly specific, e.g., succession of chlorotic and necrotic tissue or the

appearance of typical fungal structures, such as powdery mildew hyphae and conidia or rust uredospores (Fig. 3B and 4). Whereas biotrophic fungi such as powdery mildews or rusts have a relatively low impact on tissue structure and chlorophyll composition during early infection (Fig. 3B and 4), perthotrophic pathogens, such as those that cause leaf spots, often induce degradation of tissue due to pathogen-specific toxins or enzymes that ultimately results in necrotic lesions. In contrast, powdery mildews and rust fungi produce fungal structures on the leaf surface that can influence the optical properties of the plant-pathogen interaction. Complex and unique interactions are exemplarily visualized for sugar beet leaf diseases by raster electron microscopy and by semithin sections of diseased leaf parts (Fig. 4). These highly complex and unique disease patterns enable an identification of diseases based on the spectral properties of plants (Table 1, Fig. 5 and 6). Mahlein et al. (2010, 2013) demonstrated the differentiation of foliar pathogens of sugar beet based on leaf reflectance. Building on these results, Rumpf et al. (2010) was able to detect early *Cercospora* leaf spot, powdery mildew, and rust-diseased sugar beets before the appearance of visible symptoms. In other plant pathogen systems, noninvasive spectral data proved to be useful for the monitoring of *Fusarium graminearum* in wheat (Bauriegel et al. 2011), *Venturia inaequalis* in apple (Delalieux et al. 2007), or *Phytophthora infestans* in tomato (Wang et al. 2008). In proximal sensing, hyperspectral imaging also was shown to be useful for the assessment of mycotoxin-producing pathogens in maize (Del Fiore et al. 2010). Furthermore, Bravo et al. (2003) used in-field spectral images for the early detection of yellow rust infected wheat. Soilborne diseases were successfully discriminated by Hillnhütter et al. (2011), who looked at the symptoms caused by the nematode *Heterodera schachtii* and the soilborne fungus *Rhizoctonia solani* in sugar beet fields. In work



**Fig. 3. A,** The interaction of leaf tissue with light depends on structural and leaf chemical properties. **B,** During pathogenesis, leaf pathogens influence leaf structural and chemical properties, and by this the leaf optics are altered.

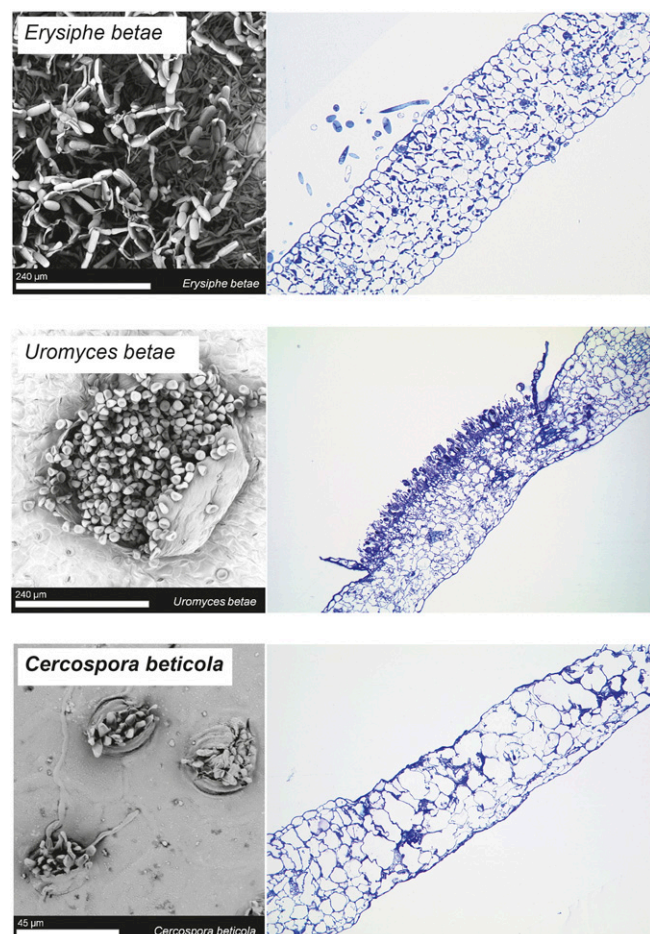
on sugarcane, Apan et al. (2004) were able to detect orange rust using EO-1 Hyperion hyperspectral imaging. Later, Huang et al. (2007) obtained reliable and accurate detection of yellow rust in wheat by ground based spectral measurements and airborne hyperspectral imaging. In addition to detection of plant diseases during the vegetation period, hyperspectral imaging is widely used for monitoring fruit health and quality. Canker lesions of citrus fruits (Qin et al. 2009), apple surface defects (Mehl et al. 2004), or rot of strawberries (ElMasry et al. 2007) can be identified by hyperspectral imaging sensors. These techniques are important in screening fruits and crops to avoid storage diseases.

**Thermal sensors.** Infrared thermography (IRT) assesses plant temperature and is correlated with plant water status (Jones et al. 2002), the microclimate in crop stands (Lenthe et al. 2007), and with changes in transpiration due to early infections by plant pathogens (Oerke et al. 2006). Emitted infrared radiation in the thermal infrared range from 8 to 12  $\mu\text{m}$  can be detected by thermographic and infrared cameras and is illustrated in false color images, where each image pixel contains the temperature value of the measured object. In plant science, IRT can be used at different temporal and spatial scales from airborne to small scale applications. However, it is often subject to environmental factors such as ambient temperature, sunlight, rainfall, or wind speed. The leaf temperature shows a close correlation to the plant transpiration (Jones 1992; Jones et al. 2002), which is affected by a diversity of pathogens in different ways. Whereas many foliar pathogens, such as leaf spots or rusts, induce local and well-defined changes, impairment by root pathogens (e.g., *Rhizoctonia solani* or *Pythium* spp.) or systemic infections (e.g., *Fusarium* spp.) often influences the transpiration rate and the water flow of the entire plant or

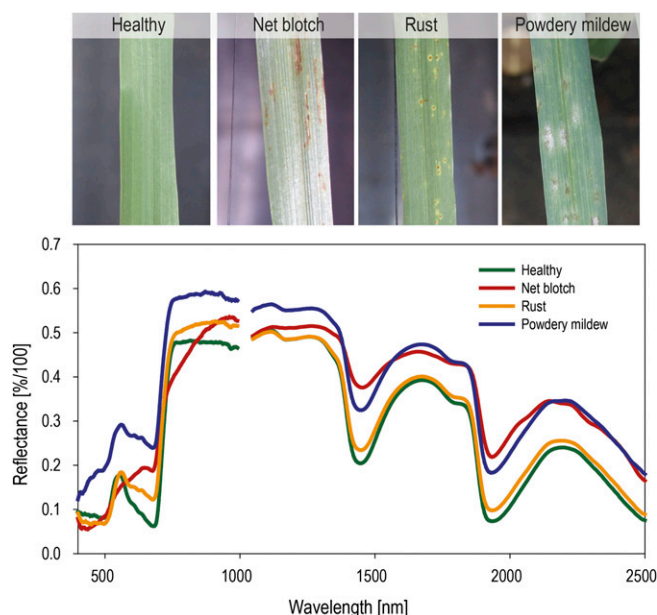
plant organs. Local temperature changes due to pathogen infection or to defense mechanisms have been reported for plant-virus interactions in tobacco and for *Cercospora beticola* in sugar beet by Chaerle et al. (2004). Oerke et al. (2006) monitored cucumber diseased with downy mildew (*Pseudoperonospora cubensis*) or scab disease in apple caused by *Venturia inaequalis* (Oerke et al. 2011) successfully by IRT. In the plant-pathosystem for apple and *V. inaequalis*, thermography was also able to visualize the spatial colonization of apple tissues by the pathogen over and beyond visible symptoms, where hyphae and conidia were only microscopically detectable (Oerke et al. 2011). Gomez (2014) monitored the infection and spread of *Peronospora sparsa* on different *Rosa* cultivars (Fig. 7). Applications in the field have been demonstrated by Nilsson (1991), with a high correlation between disease severity of various root and leaf diseases and in different crops (barley, wheat, oat, sugar beet, potatoes, etc.). For effective analysis of IRT images, the heterogeneity between and within leaves can be utilized. The mean temperature difference within single leaves, plants, and crop stands is an important indicator for the appearance of plant disease.

**Fluorescence imaging.** Various chlorophyll fluorescence parameters are used to estimate differences in the photosynthetic activity of plants. Chlorophyll fluorescence imaging instruments are commonly active sensors with an LED or laser light source that assesses photosynthetic electron transfer (Bauriegel et al. 2014; Chaerle et al. 2004; Murchie and Lawson 2013). This method has been used to study differences in the photosynthetic activity caused by biotic and abiotic stresses over the leaf area (Bürling et al. 2011; Scholes and Rolfe 2009). Combining fluorescence imaging with image analysis techniques has been shown to be useful for discrimination and quantification of fungal infections (Konanz et al. 2014). One disadvantage of current chlorophyll fluorescence imaging systems is that the preparation of the plants has to follow a strict protocol, and thus it is difficult to implement in normal agricultural greenhouses or field environments. Therefore, research has been directed at extracting fluorescence parameters from sun-induced reflectance in the field, which would have potential for plant disease assessment at the canopy or field level (Rossini et al. 2014).

**Sensors for assessing plant biomass and plant architecture.** Plant architecture and plant biomass can provide important information about the health status or the presence of a disease at the single plant scale or in fields. Different noninvasive sensors have been developed during previous years. The density of a crop stand can be an important parameter for planning targeted fungicide applications



**Fig. 4.** Detailed view of the host-pathogen interaction can be obtained by electron microscopy or semithin sections from light microscopy. This indicated the highly specific appearance and influence of different foliar diseases in the example of sugar beet leaf diseases.



**Fig. 5.** Characteristic spectral signatures of barley leaves diseased with net blotch, rust, and powdery mildew, respectively.



(Dammer et al. 2014). Photogrammetric solutions such as stereo cameras, 3D laser scanners, ultrasonography, or densitometry also have the potential to distinguish information about plant biomass or plant architecture (Busemeyer et al. 2013; Paulus et al. 2013; Wahabzada et al. 2015b). Single plant organs can be determined automatically and the volume information of, e.g., wheat panicles, can be determined and has been shown to have a high correlation to actual thousand kernel weight. Photogrammetric technology can, therefore, be applied for detection of wheat kernels infected with *Fusarium* spp. Stereo cameras and 3D laser scanners, for example, can provide data on color or reflectance intensity that can also be used for disease detection based on image or reflectance analysis, respectively (Paulus et al. 2014).

### Relevant Areas for Sensors in Plant Disease Detection

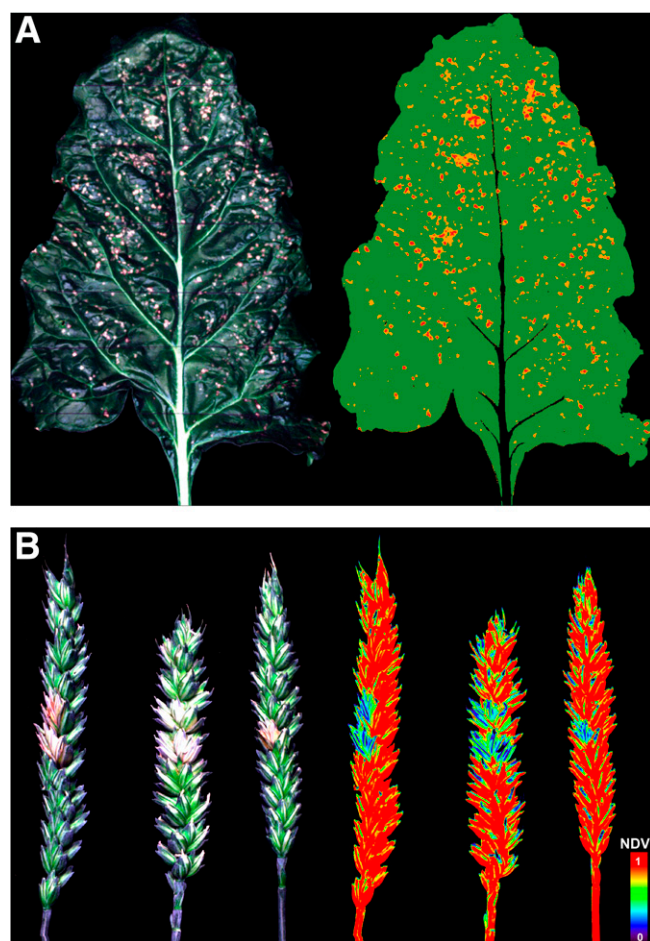
The large variety of sensor systems available to plant pathologists provides high resolution data of agricultural crop stands and can constitute the basis for early detection and identification of plant diseases. The great deal of progress in developing these technologies over the past 40 to 50 years and introducing them to agriculture and plant disease detection is impressive (Brenchley 1964; Jackson and Wallen 1975; Seelan et al. 2003; Nilsson 1995; West et al. 2003). Due to advances in precision agriculture and plant phenotyping, new and specific solutions for plant and crop science have been developed (Cobb et al. 2013; Fiorani et al. 2012; Furbank and Tester 2011; Steddom et al. 2005). The most successful sensors currently are being used for

noninvasive evaluation of crop nutrition status in the field. The development of new low-cost sensor solutions with satisfying performance available on the market is an important development for future practical applications in agriculture (Grieve et al. 2015; Paulus et al. 2014). However, to the best of my knowledge, sensors that can be used to specifically detect plant diseases are still not available on the market. The full potential of sensor based disease detection has still not been exploited. Instruments and technological solutions for field, greenhouse, and for phenotyping are available. However, these are highly specific and tailored prototypes and cannot be used on a broad scale.

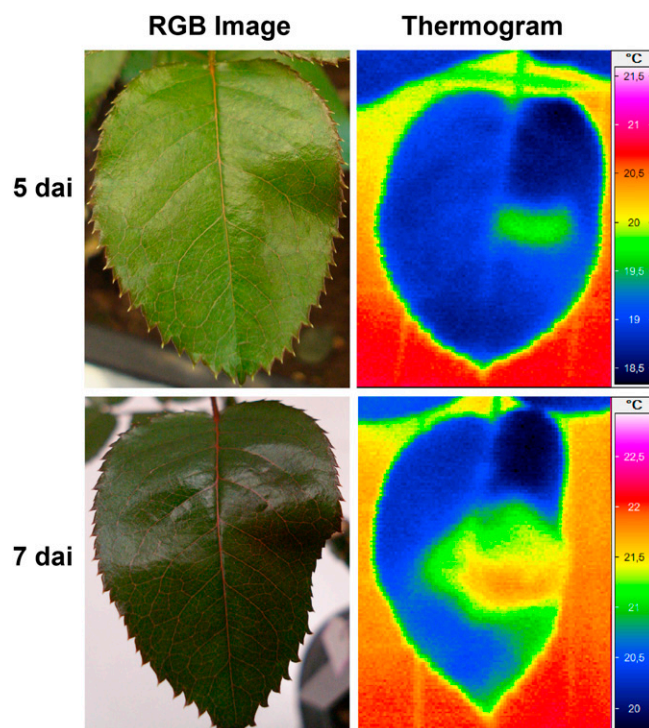
**Field systems.** Some advanced systems with potential applications in the field are the imaging platform for the detection of tulip breaking virus (TBV) infected tulip bulbs from Polder et al. (2014) or a prototype of a hyperspectral imaging platform for the detection of yellow rust (*Puccinia striiformis*) in wheat by Bravo et al. (2003). Polder et al. (2014) developed a robot with multispectral cameras and an online machine vision analysis pipeline. This research was motivated by a limited availability of technical experts for rating tulip bulbs. They were able to adjust and optimize this system to attain a level of accuracy equivalent to that obtained by experienced rating experts. With the hyperspectral imaging 'buggy' of Bravo et al. (2003), it was possible to detect and classify yellow rust diseased patches in wheat fields with a success rate of 96% under ambient light conditions. Their results are very encouraging for the development of cost-effective optical sensor platforms for an early and accurate detection of plant diseases in different crops.

**Resistance screening.** For plant phenotyping, different technical systems have been developed. The developments started with investigations of single plants under controlled conditions (Chaerle et al. 2007; Jansen et al. 2009). More recently, advanced field platforms have become more robust, enabling a holistic characterization of plant performance in multiple plots or of the entire canopy (Walter et al. 2015).

It has been shown that the level of susceptibility and/or resistance of different genotypes and varieties to a specific disease can be evaluated by optical sensors. Chaerle et al. (2007) were able to identify sugar beet lines with different levels of susceptibility to *C. beticola* by multispectral and fluorescence imaging. In addition, they were



**Fig. 6.** Disease detection of fungal plant diseases based on hyperspectral images. **A**, Supervised classification (spectral angle mapper) of *Cercospora* leaf spot on sugar beet. The green color denotes healthy leaf tissue, the yellow color the border of *Cercospora* leaf spot and the red color the necrotic center of *Cercospora* leaf spot. **B**, Spikelets, diseased by *Fusarium* head blight, can be visualized by calculation of the normalized difference vegetation index (Photo A: A.-K. Mahlein; photo B: A.-K. Mahlein and A. Al Masri).



**Fig. 7.** Monitoring of rose leaf colonization by *Peronospora sparsa* and symptom development of downy mildew in early stages (5 and 7 days after inoculation) of the disease by thermographic imaging. (Photo: S. Gomez).



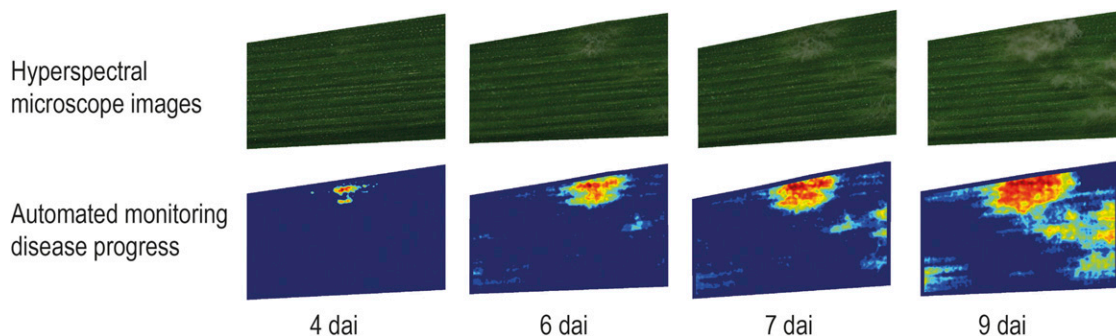
some of the first researchers to use automatized platforms for screening entire plants with sensors mounted on an xyz-robot system. Sugar beets were tested on different scales from small leaf discs, over detached leaf parts to entire leaves, fixed in a measuring grid. The attached leaf assay proved to be superior in revealing early, previsual symptoms by chlorophyll fluorescence and in discriminating between lines with different levels of susceptibility to *C. beticola* compared with detached leaf assays and in vitro tests. With the aim of high-throughput capacity, Rousseau et al. (2013) developed a thresholding approach based on the chlorophyll fluorescence parameters Fv/Fm on image pixels to evaluate symptoms caused by *Xanthomonas fuscans* subsp. *fuscans* on *Phaseolus vulgaris*. Visual observations by trained raters were reproducible and a modeling of the Fv/Fm distribution as a mixture of Gaussian distributions enabled a discrimination of various stages of symptom development. In this study, chlorophyll fluorescence underlined its potential to detect pre-symptomatic areas and could be an important tool for assessing quantitative resistance. Recent developments, such as the HyperART system for simultaneous measurements of leaf reflectance and transmission, were also invented (Bergsträsser et al. 2015). In a pilot application study, this sensor system demonstrated the potential to evaluate different levels of resistance/susceptibility of sugar beet to *C. beticola*. It has been shown that additional information from transmission measurements increases the detection sensitivity; however, the measurement protocol and procedure is rather complex and still requires substantial manual intervention.

**Assessment of plant defense reactions.** Most of the published work examining the detection of plant diseases is based on symptoms or presymptomatic physiological and biochemical changes. In resistance screening, researchers are interested in subtle defense reactions that are crucial for the ability of plants to prevent pathogen invasion or in their ability to impede pathogen development. According to Robinson (1969), host resistance is the ability of a plant genotype to hinder the growth and/or development of a pathogen. Complete resistance conditioned by a single gene (qualitative resistance) and incomplete resistance conditioned by multiple genes of partial effects (quantitative resistance) have long been recognized as two general categories of resistance (Poland et al. 2008). Plants respond to pathogen attack by activation of a large number of defense mechanisms (Glazebrook 2005). These mechanisms include production of antimicrobial metabolites and proteins, the physical reinforcement of cell walls through production of callose and lignin, and the induction of hypersensitive reactions (Hückelhoven 2007; Schulze-Lefert and Panstruga 2003; Voigt 2014). These changes occur at the tissue and cellular level after the first contact with a pathogen and are important for subsequent compatible or incompatible interactions, or considering the plant side, for susceptibility or resistance of a genotype. To assess these very early and marginal changes, specific sensors with a high spatial resolution must be developed. Douchkov et al. (2013) invented a so-called ‘microphenomic’ platform by combining high-throughput DNA cloning and single cell transformation protocols with automated microscopy and phenotyping. They were able to score fungal

penetration efficacy of *Blumeria graminis* f. sp. *hordei* on different barley genotypes. It is noteworthy that in this case, already detached and decolorized leaf parts were used for histologic studies. Penetration sites and powdery mildew haustoria as feeding organs could be automatically assessed and counted from digital microscopic RGB images. This highly invasive approach only enables evaluation of the interaction of a plant pathogen with the host plant at one single time point. Following pathogenesis over time would give further important information about the interaction of plant genotypes with pathogens. Generally, complete, partial, and incomplete resistance can be differentiated (Parlevliet 1979); a portion inhibits the initial penetration of a pathogen, while others induce a hypersensitive response to detract from the nutrition supply. However, in certain cases, the ability of a pathogen to sporulate is also inhibited (Glazebrook 2005). This aspect can only be considered through time series rating or noninvasive imaging approaches. An exact evaluation of resistance types has to be performed by multitemporal observation. A hyperspectral microscopic approach was recently developed by Kuska et al. (2015). The high spatial resolution of a pixel size of 7.5 µm coupled with a spectral resolution of the imaging sensor of 1 nm allowed the detection of subtle processes in time series after inoculation (Fig. 8). Evaluation of host-pathogen interactions over time and a discrimination of barley genotypes differing in susceptibility to powdery mildew were possible with this sensor-based and data driven phenotyping approach on a small scale level.

### The Big Data Challenge – Information Retrieval from High Dimensional Data

Regardless of all of the positive and future benefits of sensors for plant disease detection, we have to take into consideration that the interpretation of the sensor data are crucial (Barbedo 2013; Behmann et al. 2014; Wahabzada et al. 2015a). Most of the sensors do not measure plant physiological parameters directly but record a spectrum that is the sum of reflectance attributed to various plant characteristics and the measurement conditions (e.g., leaf inclination, illumination, and surface texture) (Gitelson et al. 2002; Jensen 2007). Additionally, the current quality and quantity of data that are made available from plant sensors has dramatically increased. The amount and dimension of sensor generated data can easily reach terabytes. This level of data with their underlying high spectral, spatial, and temporal resolution require the use of advanced methods of data handling, analysis, and interpretation (Bauckhage and Kersting 2013; Wahabzada et al. 2015a). In precision agriculture, we are dealing with observations of large areas or entire fields. It is important to process the data directly and online, resulting in a consequent plant protection measure (spray or not to spray). In plant phenotyping, the high number of genotypes or treatments being examined increases the dimensionality and complexity of the data produced (Fig. 1). Task-specific algorithms have to be developed to handle these data effectively. Behmann et al. (2014) discussed different data analysis methods of sensor data for disease detection. Up until today, there was not one superior method for all plant science and practical



**Fig. 8.** Powdery mildew progress on a susceptible barley genotype cv. Ingrid assessed by a hyperspectral microscope (Kuska et al. 2015). Using this small-scale approach, the phenotyping and differentiation of different genotypes is possible. (Photo: A.-K. Mahlein, M. Kuska, and M. Wahabzada).

agricultural applications. Supervised and unsupervised classification methods and clustering from remote sensing and data mining, such as k-means, artificial neural networks, self-organizing maps, or support vector machines, can be effective for detection, identification, and quantification of plant diseases from sensor data (Camargo and Smith 2009; Moshou et al. 2004; Rumpf et al. 2010; Wang et al. 2008). Principal component analysis has been successfully applied to monitor processes during pathogenesis in wheat infected with *F. graminearum* (Bauriegel et al. 2011). Different studies have demonstrated that the entire spectrum from 350 to 2,500 nm was not needed to detect crop stress due to fungal infections in the field because narrow spectral bands are highly correlated to each other (Bravo et al. 2003; Delalieux et al. 2007; Mewes et al. 2011). Depending on the object and aim, just a few regions of the spectral range may be of interest. Using a relatively small number of wavebands, detection equipment can be tailored to a specific crop and relevant plant-pathogen systems (Bravo et al. 2003; West et al. 2003). Subsequently, Mahlein et al. (2013) developed a method for extracting relevant wavebands from hyperspectral reflectance data and then combined these wavebands in spectral indices for specific detection of different foliar sugar beet diseases. These results can be technically implemented by imaging sensors based on narrowband LEDs and silicon C-MOS imaging detectors, reducing costs of spectral plant sensors dramatically (Grieve et al. 2015). Recently, within the context of the big data, new and innovative algorithms and machine learning methods are available. Wahabzada et al. (2015a) demonstrated the first data driven and automated analysis approach of hyperspectral imaging data from barley leaves diseased with foliar pathogens. It was possible to extract characteristic spectral signatures without human intervention and to follow symptom development during pathogenesis (Fig. 8).

### The Path to Success is Transdisciplinary Research!

The above review demonstrates that there are multiple opportunities and technical solutions for an automated, objective, and reproducible assessment of plant diseases by optical sensors (Fig. 2). Thermography and chlorophyll fluorescence are sensitive to early stress reactions of a plant; however, these sensors lack the potential to identify specific diseases. In this case, RGB-based and hyperspectral imaging techniques are preferable. It is to be expected that in the near future, smartphone and mobile phone based solutions, whether as built-in sensors or external sensor equipment, will significantly affect the availability and spread of knowledge based information and sensing techniques in plant disease detection, even for small-scale farmers. However, it must be said that most of the imaging technologies for this type of application are still in developmental phases. The time frame for a standardized introduction of this type of technology into agricultural practice in precision agriculture or plant phenotyping cannot yet be visualized. Despite similar tasks in plant disease detection, precision agriculture and plant phenotyping demand specific requirements and standards on imaging sensors (Fig. 1 and 2). Both domains are information-intensive, but the handling of the information and subsequent procedures differ. In precision agriculture, large scale areas have to be monitored and a plant protection measure has to take place in a short time frame (Stafford 2000). In the field, natural infection from aerial spores or diseases overwintering on debris serves as the source of inoculum. In plant phenotyping, all scales, from cell cultures to single plant organs to entire fields, are relevant (Granier and Vile 2014), and controlled inoculation with a certain amount of spores or infective units at a certain developmental stage is common. Often, a high number of repetitions with different genotypes and treatments have to be evaluated and characterized while considering morphological and physiological plant traits. In precision agriculture, special attention is placed on timing. Plant protection measures have to take place during a certain time frame to avoid economic losses due to plant pathogens. In plant phenotyping time series measurements, following a given protocol is of importance to monitor susceptibility or resistance of different genotypes. Thus, the platforms carrying a sensor and the follow-up data analysis have to be developed from different

perspectives. There are platforms such as UAVs, autonomous robots, and tractor-based sensors can be implemented in both domains. Another aspect not well-considered to date is the fusion of information and data from different sensors to provide a holistic view on plant-pathogen systems. The studies of Bravo et al. (2003), Berdugo et al. (2014), and Behmann et al. (2015) substantiated the additional benefit of sensor fusion approaches.

Because circumstances in plant phenotyping are often under better control, it is likely that sensor applications will prevail here first as compared with the time-consuming visual rating by technicians. For future research, it is indispensable to link complementary research fields, such as plant pathology, sensor development, informatics, and machine learning. Only a highly interdisciplinary approach with a close link to practical agriculture can lead to powerful solutions for diagnosis and disease detection with a high accuracy and sensitivity that will improve plant health management. In addition to this, there are many areas of research that needs to be more detailed:

- Characterization of different pathogen groups (fungi, bacteria, and viruses; soilborne or foliar pathogens) is necessary
- The impact of mixed infections on the optical properties of plants has to be investigated
- The interaction between foliar pathogens and soilborne pests such as nematodes has to be evaluated
- The interaction of biotic and abiotic stress has to be explored
- The impact on genomic, transcriptomic, and metabolic characteristics of plants to optical properties has to be investigated
- The connection to other knowledge based methods will provide a holistic perspective on plant systems (e.g., weather based prediction models)
- The potential for early infection before visible symptoms has not been fully exploited to date

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