REVIEW AND INTERPRETATION

Approaches, applications, and future directions for hyperspectral vegetation studies: An emphasis on yield-limiting factors in wheat

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Abstract
Hyperspectral instruments acquire spectral information in many narrow, contiguous bands throughout the visible, near-infrared and shortwave regions of the electromagnetic spectrum. Hyperspectral techniques are becoming very powerful tools for characterizing plants and nondestructively quantifying their chemical and physical properties because of their ability to provide layered trait information within the same spectral region. However, to effectively make use of hyperspectral sensing, an understanding of the theory behind these techniques, the power, and the limitations of the resulting data is required. This article presents an overview of hyperspectral sensing in regard to principles, instrumentation, processing methods, and current applications, specifically focusing on the quantification of yield-limiting factors in wheat (Triticum aestivum L.). The spectral properties of plants across the electromagnetic spectrum are first described to achieve a better understanding of plant–light interactions. Basic information about different imaging approaches is provided as are the necessary considerations for the analysis of hyperspectral data. Some of the major technical challenges associated with hyperspectral imaging as well as future directions are discussed. Finally, as an example crop, the use of hyperspectral techniques for quantifying yield-limiting factors in wheat is presented.

1 | INTRODUCTION

The accurate quantification of crop growth parameters throughout the growing season is important to assess crop performance and provides an opportunity for improving grain yields and quality through approaches such as site-specific application of fertilizers (Hansen & Schjoerring, 2003). Traditional methods of measuring growth parameters are reliable but are labor-intensive, time-consuming, and are not spatially explicit or extensive (Furbank & Tester, 2011; Mishra et al., 2017). The need for rapid, nondestructive measurements for understanding plant health and performance has driven the widespread implementation of different optical sensor technologies. Hyperspectral methods are one such emerging field becoming more commonly utilized as a nondestructive and rapid option.

Hyperspectral methods measure the light intensity across many narrow, contiguous spectral bands across the visible (VIS), near infrared (NIR), and shortwave infrared (SWIR)
regions of the electromagnetic spectrum. Hyperspectral methods have the vast and superior advantage over other optical techniques in that they can provide layered trait information within the same spectral region. For example, the red edge inflection point (REIP), the maximum of the first derivative reflectance between red and NIR wavelengths, can give an indication of chlorophyll a, chlorophyll b, leaf area index (LAI), and relative water content (RWC) simultaneously (Delegido et al., 2013; Delegido, Verrelst, Alonso, & Moreno, 2011; Filella & Peñuelas, 1994; Gitelson & Merzylyak, 1994; Horler, Dockray, & Barber, 1983).

Modern hyperspectral sensors can provide a narrow spectral sampling interval of 1 nm or less. Recent work has utilized hyperspectral sensing to quantify crop characteristics such as foliar chemistry (Onoyama, Ryu, Suguri, & Iida, 2013; Vigneau, Ecarnot, Rabatel, & Roumet, 2011; Yu, Lenz-Wiedemann, Chen, & Bareth, 2014; Zhang, Liu, He, & Gong, 2013), disease presence (Mahlein, Steiner, Hillnhütter, Dehne, & Oerke, 2012; Zhang, Wu, You, & Zhang, 2017) and drought stress (Römer et al., 2012; Mo et al., 2015). Authors have also documented the use of hyperspectral imaging systems incorporated in field phenotyping systems (Busemeyer et al., 2013; Virlet, Sabermanesh, Sadeghi-Tehrani, & Hawkesford, 2016) and high-throughput phenotyping platforms (Feng et al., 2017; Ge, Bai, Stoerger, & Schnable, 2016; Pandey, Ge, Stoerger, & Schnable, 2017) for broad-scale and close-up analysis of plant traits.

2 | SPECTRAL PROPERTIES OF VEGETATION

Light is the primary source of energy for plant processes, the most notable being photosynthesis. When light is incident on a leaf surface, it is either reflected, absorbed, or transmitted. The degree to which it is reflected, absorbed, or transmitted varies with the wavelength of radiation, the angle of incidence, the surface roughness, cellular structure, and the chemistry of the leaves themselves (Kumar, Schmidt, Dury, & Skidmore, 2001). The component of light that gets absorbed by vegetation is a result of the molecular vibration and stretching of specific molecular bonds (Coates, 2006; Stuart, 2004). The chemistry of a leaf predominantly consists of four elements: H, C, O, and N. The reflectance characteristics of a leaf are therefore strongly influenced by the interaction of light with C–O, O–H, C–H, and N–H bonds (Curran, 1989).

The general reflectance spectrum for green vegetation in the range of 350–2500 nm is similar for all species. The vegetation spectrum can be divided into three distinct wavelength domains defined by the main parameters governing the reflectance in that region: the VIS (350–700 nm), the NIR (700–1100 nm), and the SWIR (1,100–2,500 nm) (Figure 1). The spectral characteristics of vegetation in the visible region are dominated by the absorption of light by photosynthetic pigments such as chlorophyll or carotenoids (Blackburn, 2007; Roy, 1989). In the NIR, light absorption is due to leaf cell structure and light scattering, whereas in the SWIR light absorption is controlled by water content and leaf biochemistry (Sahoo, Ray, & Manjunath, 2015).

Across the visible region, vegetation reflectance is generally low due to the absorption by leaf pigments, including chlorophyll, carotenoids, xanthophylls, and polyphenols. Leaf pigments such as chlorophyll absorb light of certain wavelengths. This causes electron transitions within the pigment, releasing energy for photochemical reactions. However, only light of certain energy can cause these electron transitions meaning that plant pigments only absorb light at some wavelengths and not others (Gates, Keegan, Schleter, & Weidner, 1965).

Absorption troughs caused by chlorophyll are located at approximately 420, 470, and 660 nm (Curran, 1989; Sahoo et al., 2015). These strong absorption bands create a reflectance peak in the green region at about 550 nm. Carotenoids absorb at shorter wavelengths between 440 and 480 nm. If normal plant growth is interrupted, for example, if a plant is subjected to stress, it may decrease or cease chlorophyll production. This results in lower chlorophyll absorptions in the blue (450–495 nm) and red (620–720 nm) wavelengths.

Perhaps the most distinctive feature of the vegetation reflectance spectrum occurs at the transition from VIS to NIR and is known as the “red edge” (Krezhova, Maneva, Moskova, & Krezhov, 2015). This is viewed as a dramatic increase in reflectance in the spectral region between 680 and 720 nm. In the NIR region, reflectance is strongly influenced by water content, leaf development, cell structure, and the repeated scattering of light from cell interfaces (Carter, 1991; Gausman & Allen, 1973; Sahoo et al., 2015). Near infrared reflectance reaches high levels because leaf pigments and cellulose are almost transparent in this region resulting in a very low absorption (Sahoo et al., 2015). As opposed to VIS wavelengths, the energy of NIR light is not sufficient to cause photochemical reactions and is therefore not absorbed by pigments. Near infrared reflectance is further increased

Core Ideas

- Hyperspectral techniques becoming popular tools for characterizing plant properties.
- Nondestructive quantification of plant parameters possible through spectral acquisition.
- Present an overview of hyperspectral principles, instrumentation, processing and applications.
- Hyperspectral data for quantifying yield-limiting factors in wheat is reviewed.
by internal scattering between hydrated plant cells and intercellular air; the amount of light reflected in the NIR increases with the number of inter-cell spaces and cell layers within the leaf (Guyot, 1990; Sahoo et al., 2015). Strong absorption bands due to liquid and vapor water in the plant occur at approximately 970 and 1200 nm (Curran, 1989; Sims & Gamon, 2003).

In the SWIR region, reflectance properties of vegetation are mainly governed by water content and the biochemistry of leaves. Major atmospheric water absorption bands occur at 1450 and 1940 nm (Curran, 1989; Sahoo et al., 2015). However, in contrast to pigments where absorptions are caused by electron transitions, water absorptions are caused by vibrations and rotations of water molecules (Kumar et al., 2001). Protein, cellulose, lignin, and starch can also influence reflectance properties in the SWIR but the absorption peaks associated with these compounds are often weak if present at all: they are often obscured by the overall water absorption in fresh leaves and only become evident at lower water contents or in dry plant material (Curran, 1989; Sahoo et al., 2015).

In fresh leaves, water absorption features strongly obscure the absorption features of other biochemical components (Goetz, Gao, Wessman, & Bowman, 1990; Kokaly & Clark, 1999). In contrast to healthy, green leaves, the spectra of dried leaf material lack the strong water and chlorophyll absorptions that dominate green leaf spectra. Chlorophylls degrade rapidly during leaf senescence (Sanger, 1971) resulting in decreased absorptions, and hence increased reflectance, in red and blue wavelengths. Other pigments such as carotenes and xanthophylls become the dominant pigments in the leaves. These pigments absorb blue light but reflect in the green and red wavelengths, giving rise to the yellow color of senescing leaves. Senescent vegetation may also show numerous weak absorption features in the NIR associated with lignin, tannins, cellulose, pectins, and waxes (Elvidge, 1990).

Measured reflectance of vegetation is further complicated when taking measurements of canopies rather than single leaves. Absorption features can be concealed when measurements are made at the canopy scale because of its structure, leaf orientation, external illumination, atmospheric absorption, and background influences (Al Makdessi, Ecarnot, Roumet, & Rabatel, 2019; Axelsson, Skidmore, Schlerf, Fauzi, & Verhoef, 2013; Pinter et al., 2003). Therefore, the spectral signatures of crop canopies in the field can be significantly different to those taken of single green leaves measured in the laboratory.

### 3 | Hyperspectral Data

Hyperspectral sensing can involve a number of diverse instruments, platforms, and analysis techniques. The type of sensor, platform, and analyses utilized will be strongly influenced by the application, desired outcome, and the required spatial scale and spectral resolution.

#### 3.1 | Nonimaging Sensors

Nonimaging instruments record the spectral signature of a single point with high spectral resolution. If the field-of-view contains materials with different spectral properties, then the spectrum measured is an average (Behmann, Steinrücke, & Plümner, 2014). There are several commercial systems that acquire spectral information in narrow wavebands. These include the ASD FieldSpec (Analytical Spectral Devices), GreenSeeker (Trimble), N-sensor ALS (Yara International...
ASA), and CropCircle (Holland Scientific). Although most of these sensors acquire spectral information at only a few specific wavelengths to calculate classic vegetation indices, the ASD FieldSpec can measure the entire optical spectrum from 350 to 2500 nm, which can then be related to specific crop properties such as nutrient levels, disease presence, water content, and plant structure (El-Hendawy et al., 2019b; Ge et al., 2019; Mahajan, Sahoo, Pandey, Gupta, & Kumar, 2014; Yendrek et al., 2017).

Nonimaging instruments have been extensively used to quantify hyperspectral properties of wheat (Triticum aestivum L.) both in the field (El-Hendawy et al., 2019b; Mahajan et al., 2014; Pimstein, Karsieli, Bansal, & Bonfil, 2011) and in laboratory settings (Bannari, Khurshid, Staenz, & Schwarz, 2007; Das et al., 2017). Quite often, measurements collected under laboratory conditions are made proximally (i.e., in direct contact with a leaf) and rely on an active light source. They provide their own illumination and hence are not dependent on ambient lighting and atmospheric conditions. For this reason, they are often used to explore how varying plant status affects the reflectance of a sample under controlled conditions. As opposed to laboratory spectra, field data are more often collected at some distance from the target, with strong dependence on ambient light conditions. Collecting distal data increases the field of view and is useful for collecting information across a crop canopy rather than a single leaf. Nonimaging instruments can be mounted on simple platforms to collect measurements (Bai et al., 2019; Comar et al., 2012; Dreccer, Barnes, & Meder, 2014; Hansen & Schjoerring, 2003) but are typically used as hand-held devices.

3.2 | Hyperspectral imaging cameras

Imaging instruments, or cameras, provide visual images similar to conventional red–green–blue (RGB) photography but with much more detailed spectral information across a broader range of wavelengths. They combine the key features of both spectroscopic data and RGB imagery through the simultaneous acquisition of both spectral and spatial information. While nonimaging instruments provide spectral information that allows an understanding of plant biochemistry, imaging technology improves upon this by capturing a spatially explicit, visual representation of the target’s spectral information. This is preferable for many applications because the spatial dimension can provide extra information on shape, gradient, or texture of the target (ElMasry, Kamruzzaman, Sun, & Allen, 2012). Therefore, imaging systems are often used on remote sensing platforms for landscape assessment of vegetation as well as in plant phenotyping systems for high-throughput analysis of individual plants (Haboudane, Miller, Pattey, Zarco-Tejada, & Strachan, 2004; Pandey et al., 2017). There are many hyperspectral cameras currently available on the market. Some of the most common include: PIKA II (Resonon, Inc.), HySpex VNIR (HySpex), FX10 and FX17 (Specim Spectral Imaging Ltd.), and Micro-Hyperspec (Headwall Photonics Inc.). It is also common for researchers and engineers to develop their own systems based on their specific requirements.

A typical hyperspectral imaging system contains a light source, objective lenses, an imaging spectrograph, and an area detector (Figure 2). The light source generates the light that illuminates or excites the target. The most common lights installed in such systems are halogen lamps but as the need for cheaper and more powerful light sources has increased, light emitting diode (LED) technology, lasers and tunable sources have also been incorporated (Eitel, Magney, Vierling, & Ditmar, 2014; Mahlein et al., 2015; Mishra et al., 2017). The objective lenses focus the incoming light onto the area detector. As the light travels through the objective lenses, the imaging spectrograph disperses the broadband light into separate, discrete wavelengths. After the light has traveled through the dispersion device, it reaches an area detector, which measures the intensity of the light by converting it into electrical signals.

The type of detector within an imaging system depends primarily on the required wavelengths. In VIS-NIR systems, charge-coupled devices (CCDs) or complementary metal oxide semi-conductor (CMOS) sensors are normally used as detectors. At longer wavelengths, typically beyond 900 nm, indium gallium arsenide (InGaAs) sensors are used due to their increased sensitivity (Huang, Liu, & Ngadi, 2014). The InGaAs detectors are more expensive than CCD or CMOS detectors used for detecting VIS-NIR wavelengths. As such, the higher costs associated with these cameras can limit their widespread application and availability in comparison to cameras operating across VIS-NIR wavelengths.

Hyperspectral imaging systems typically use one of four acquisition methods: point scanning (whiskbroom imaging); line scanning (pushbroom imaging); area scanning (wavelength scanning); or snapshot (nonscanning) (Figure 3). Point-scanning scans a single position along two spatial dimensions by moving either the sample or the detector and uses a spectrophotometer to collect the data pixel-by-pixel (Qin, 2010). Instead of scanning a single point at a time, the line-scanning method measures a line of spatial and spectral information simultaneously for each detector in the line. The dataset is collected as the line scans in the direction of motion. The area-scanning method uses a filter wheel or tunable spectral filter in front of a camera to measure a 2D grayscale image for each wavelength. Finally, the snapshot method simultaneously collects both the spatial and spectral information during a single detector integration period without scanning. Snapshot cameras can generate images with high resolutions and, unlike the scanning methods, do not suffer from motion artifacts (Hagen & Kudnov, 2013).
The data collected from hyperspectral imaging systems is in the form of a three-dimensional spatial map of spectral variation, commonly referred to as a “data cube” (Figure 4). The first two dimensions reflect the spatial information and the third provides the spectral properties for each pixel. An entire reflectance spectrum is measured for each pixel, making each pixel spectrally and spatially unique.

The raw spectra acquired by hyperspectral cameras not only record the target plants but are also influenced by properties of the background illumination and the instrument itself. Spectral calibration is required to account for these additional, unwanted effects and to obtain only the spectral information of the target plants. Calibration is performed by normalizing the spectrum against black and white references. The black reference measurement is acquired by completely blocking out the instrument’s light source, thereby measuring any inherent properties of the instrument. The white reference is measured by scanning a Lambertian surface with 99% reflectance. The calibrated reflectance of the target is then calculated by:
where \( R_c \) is the calibrated reflectance, \( DN_{\text{raw}} \) is the raw digital number of the target measured by the hyperspectral imaging system, \( DN_{\text{dark}} \) is the dark reference measurement, and \( DN_{\text{white}} \) is the white reflectance measurement. This calibration must also be performed with nonimaging instruments. However, spectral acquisition software can often perform these calculations automatically with prompting from the operator.

### 3.3 Scales and platforms

Hyperspectral instruments can be mounted on a variety of platforms to acquire spectral information at different spatial and spectral resolutions. Such platforms include spaceborne, airborne, ground-based, and controlled environment high-throughput platforms. The spatial resolution of imaging systems decreases as the distance between the platform and target increases. However, the area of coverage also increases with distance. Therefore, there is a trade-off between coverage extent and resolution when selecting an appropriate platform from which to perform hyperspectral measurements.

Spaceborne platforms involve mounting a hyperspectral sensor on a satellite that is then placed in either low, medium, or geostationary orbit. Satellite platforms are stable platforms, offer repeated measurements, and can cover extensive areas simultaneously. However, compared with other platforms, most satellites suffer from low spatial resolution and may be limited by cloud cover and by the fixed timing of acquisitions.

Aerial platforms are common for hyperspectral instruments because they are flexible in that the height, flight direction, timing, and frequency of flights can be varied. Aerial platforms not only include aircraft (Haboudane et al., 2004), but can also include balloons or helicopters (Chen & Vierling, 2006; Dehaan, Weston, & Rumbachs, 2012). Aerial acquisitions are performed at altitudes of less than 1 km to 10’s of kilometers depending on the capabilities of the platform and sensor. Because they have a smaller spatial coverage than satellites, aerial platforms are normally used for local or limited regions of interest. Aerial acquisitions are more flexible with their timing, but they can still suffer from cloud cover and payload capabilities and can be expensive.

Unmanned aerial vehicles (UAVs) are aboveground vehicles that do not require an onboard pilot for their control (Lodhi, Chakravarty, & Mitra, 2018). They are a well-suited option for small-scale and research applications due to their ability to collect high-resolution data that is available almost instantly. However, most UAVs have a limited payload and a short flight endurance, limiting their implementation in large-scale studies. There are some exceptions to this, such as the Alta UAV (The Freefly Systems), which is capable of a 35-min flight time with a payload of 18 kg, and the Vulcan UAV Airlift (Mitcheldean), which has payload capacities of up to 25 kg.

For ground-based hyperspectral sensing, data is collected at ground level from either a moving or stationary platform. Ground-based platforms can include stepladders, cherry-pickers, or ground-based vehicles (Delalieux et al., 2007; Fletcher, Everitt, & Yang, 2011; Mistele & Schmidhalter, 2010). Ground-based vehicles are less affected by cloud cover and atmospheric conditions, not limited by payload, and are flexible in their deployment and timing. Like UAVs, they are most suited for small-scale or research applications because of their small coverage area.

Hyperspectral cameras have recently been installed in controlled environment high-throughput phenotyping systems such as those at the Australian Plant Phenomics Facility at The University of Adelaide (www.plantphenomics.org.au), the Controlled Environment Phenotyping Facility at Purdue University (https://ag.purdue.edu/cepf), and the Greenhouse Innovation Complex at the University of Nebraska-Lincoln (https://ard.unl.edu/phenotyping/nebraska-innovation-campus-greenhouse). High-throughput phenotyping systems are fully automated platforms specifically designed to nondestructively collect plant growth data. Most high-throughput phenotyping facilities consist of individual pots on conveyor belts that transport the pots to imaging and watering stations. However, some high-throughput phenotyping systems, such as that at LFL Weihenstephan (www.lfl.bayern.de) or the Pflanzenkulturhalle system at IPK (www.ipk-gatersleben.de/phänotypisierung/organismus), are capable of imaging much larger soil bins containing numerous plants. The LemnaTec Field Scanner at the University of Arizona Maricopa Agricultural Center and USDA Arid Land Research Station, containing a 30-Mg gantry traveling along a 200-m steel raling system, has been developed and employed for high-throughput phenotyping of crops in the field (https://www.terraref.org). Many of these research facilities use hyperspectral cameras for model calibration and proof of concept studies to estimate nutrient content or to detect early stress symptoms.

### 4 APPROACHES TO HYPERSPECTRAL ANALYSIS

Collecting hyperspectral data over hundreds of contiguous narrow bands results in data with high redundancy due to the high correlation of adjacent wavebands (Bajwa & Kulkarni, 2011; Jiang, Tang, Wang, & Wang, 2002; Stellacci et al., 2012). This complexity creates great challenges in data handling and analysis (Sahoo et al., 2015; Stellacci et al., 2012). Therefore, advanced analysis techniques are required...
FIGURE 5  Levels and approaches to hyperspectral analysis. The complexity increases with each level. The analysis selected will depend upon the desired outcome from the data. Adapted from Resmini, 2005

to extract relevant information from hyperspectral data. This can be achieved through appropriate preprocessing, data reduction, and the selection of a suitable analysis approach. Depending on the desired outcome, the analysis of hyperspectral data can be considered as either detection, classification, discrimination, identification, characterization, or quantification (Figure 5). In regard to estimating wheat variables, the most common approach is quantification, the determination of the absolute abundance of a particular constituent (e.g., N content).

4.1 | Preprocessing

Hyperspectral data can be strongly influenced by scattering effects inadvertently added during the measurement process, which will strongly influence the sample spectra (Huang, Romero-Torres, & Moshgbar, 2010; Rinnan, van den Berg, & Engelsen, 2009). Scatter effects add variability unrelated to the property of interest and are caused by physical phenomena such as cell, leaf or canopy structure, size, or shape rather than chemical properties (Al Makdessi et al., 2017; Asner, 1998; Huang et al., 2010; Ollinger, 2011). The effects of scatter can largely be reduced by the application of suitable preprocessing techniques (Rinnan et al., 2009), optimizing the subsequent data analysis.

Various pretreatment techniques are used to reduce spectral scatter and improve the signal/noise ratio of measurements depending on the type of scatter present, whether additive or multiplicative. The different correction techniques can be grouped into two main categories: scatter-correction methods or derivatives. The multiplicative scatter correction (MSC) (Martens, Jensen, & Geladi, 1983) is one of the most commonly applied corrections because of its ability to estimate and remove both additive and multiplicative effects. The multiplicative scatter correction is achieved by regressing a measured spectrum against a reference spectrum, commonly the average of all samples (Martens & Naes, 1989; Santos Panero, Santos Panero, Santos Panero, & Bezerra da Silva, 2013). Other common scatter correction methods include extended multiplicative scatter correction (EMSC) (Martens & Stark, 1991), detrending, and standard normal variate (SNV) correction (Barnes, Dhanoa, & Lister, 1989).

Derivative methods are also common and proven pretreatment methods for enhancing resolution and correcting baseline shifts in hyperspectral data. Derivative methods calculate the first or higher-order derivatives of reflectance with respect to wavelength. Two main techniques are used to calculate derivatives: Savitzky-Golay (SG) derivatives (Savitzky & Golay, 1964) or Norris-Williams (NW) derivatives (Norris & Williams, 1984). Derivatives are advantageous for the fact that they can resolve and enhance spectral features and correct for baseline shifts. However, a number of data points at the beginning and end of the spectra are lost depending on the selected parameters.
Bad pixels such as dead pixels or spikes, those pixels from which no valuable information can be extracted, must also be identified and removed from hyperspectral images. Dead pixels are created when the detector does not sense light levels correctly and leads to permanent black pixels (Mishra et al., 2017), whereas a spike is a false intensity peak of a particular pixel (Dorrepaal, Malegori, & Gowen, 2016). Bad pixels are caused by defective detector elements, including defects on electronics or errors in reading and accessing the data generated by the sensors (Celestre, Rosenberger, & Notni, 2016). Approximately 1% of pixels in NIR detectors are considered as “bad pixels” (i.e., either dead or a spike) and should be removed or appropriately adjusted (Dorrepaal et al., 2016; Firtha et al., 2008). To adjust dead pixels and spikes, their erroneous values are often substituted with an interpolated value through signal smoothing algorithms (Mishra, Karami, Nordon, Rutledge, & Roger, 2019; Vidal & Amigo, 2012).

For hyperspectral imaging data, preprocessing is often followed by image segmentation. Segmentation separates the image into regions based on either spectral or spatial similarities and is used to extract certain sections of the image related to the target property. The most common segmentation task for images of single plants is the removal of background pixels from vegetation pixels of interest. The easiest way to perform such segmentation is to compute a vegetation index of the image, which highlights areas of photosynthetic vegetation. A certain threshold can then be determined which identifies all the vegetation within the image while excluding all other materials.

4.2 | Wavelength selection

The main advantage of hyperspectral data is the ability to nondestructively estimate properties of a target by measuring a large amount of spectral information. However, the substantial amount of data collected with each measurement poses considerable challenges when trying to extract useful information. Many of the narrow bands contain similar or redundant information and removing such bands is required to extract only the wavelengths providing unique and useful information.

The selection of optimal wavelengths from hyperspectral data has been performed in numerous studies. Kawamura, Watanabe, Sakanoue, and Inoue (2008) found that significantly higher accuracies and lower errors were obtained using partial least squares models with selected wavelengths when predicting pasture biomass and quality. Using second derivative spectra, Becker, Lusch, and Qi (2005) identified eight significant wavelengths that were useful in differentiating coastal wetland species and Wang, Huang, Zhou, and Wang (2008) identified 15 wavelengths for the estimation of leaf area in paddy rice. Stepwise multiple linear regression (MLR) is one of the most common methods used for wavelength selection for estimation of plant properties (Das et al., 2017; Feng et al., 2018; Kokaly & Clark, 1999). Other approaches to identify optimal wavebands include partial least squares regression (PLSR), principal component analysis (PCA), stepwise discriminant analysis (SDA), lambda-by-lambda plots, uniform feature design and wavelet transforms (Andersen & Bro, 2010; Cheng et al., 2004; Stellacci et al., 2016; Stellacci, Castignano, Troccoli, Basso, & Buttafuoco, 2016; Thenkabail, Lyon, & Huete, 2011; Wilson, Zhang, & Kovacs, 2014).

There are many approaches for selecting optimal wavelengths but there is no “best” selection method due to the unique interaction between each method and the experimental data characteristics (Mehmood, Liland, Snipen, & Saebo, 2012; Thenkabail, Encola, Ashton, & Van Der Meer, 2004). Wavelength selection is complicated by the fact that the optimal wavelengths will vary between different crops, applications, or even different growth stages (Wang et al., 2008). Identifying which bands give the maximum information should therefore be carried out for specific crops and growth stages.

4.3 | Vegetation indices

Vegetation indices (VIs), also called spectral indices, are simple relationships between specific wavelengths, which are used to quantify various plant traits. Vegetation indices are often used due to their simplicity, ability to synthesize multiple bands, and wide applicability (Liang et al., 2018; Roberts, Roth, Wetherley, Meerdink, & Perroy, 2018; Xue & Su, 2017). Many indices have been developed for use with multispectral reflectance data captured in relatively few bands of wide bandwidths. These include the normalized difference vegetation index (NDVI) (Rouse, Haas, Scheel, & Deer, 1974) and simple ratio (SR) (Jordan, 1969). However, some of these broadband indices are prone to signal saturation at higher vegetation levels and are unable to explain a large proportion of variability in modeling parameters (Thenkabail et al., 2011).

Vegetation indices have also been developed specifically using hyperspectral narrowband data, such as the yellowness index (YI) (Adams, Philpot, & Norvell, 1999), water index (Peñuelas, Pinol, Ogaya, & Lilella, 1997), and the modified chlorophyll absorption reflectance index (MCARI) (Daughtry, Walthall, Kim, Brown de Colstoun, & McMurtrey, 2000). Narrowband indices have also been developed to specifically assess wheat properties. For example, Mahajan et al. (2014) proposed an index to predict N, P, S, and K. Pimstein et al. (2011) proposed a narrowband index for monitoring K and P and Zhou et al. (2019) proposed a novel carotenoid/chlorophyll ratio index to monitor wheat physiological and phenological status.
4.4 | Classification

Classification techniques separate data into two or more different groups or categories based on their spectral information (Figure 5). Classification can be considered as either supervised or unsupervised. Unsupervised techniques require no prior knowledge of the data, only input data is required without corresponding output variables. On the other hand, supervised classification requires output variables to be defined. The algorithm then learns the mapping function from the input data to the corresponding output variables. Common supervised classification techniques for hyperspectral data include linear discriminant analysis (LDA) (Arafat, Aboelghar, & Ahmed, 2013; Carvalho, van der Putten, & Hol, 2016), support vector machines (SVM) (Axelsson et al., 2013; Xiaming et al., 2015), partial least squares-discriminant analysis (PLS-DA) (Liu et al., 2014; Matzrafi et al., 2017), and artificial neural networks (ANNs) (Goel et al., 2003; Yi, Huang, Wang, & Liu, 2007). Unsupervised techniques are less common than supervised techniques but include k-means clustering (Behmann et al., 2014; Bergsträßer et al., 2015) and PCA (Kalacска, Bohman, Sanchez-Azofeifa, Castro-Esau, & Caelli, 2007; Liu et al., 2014). Classification techniques have been used in wheat studies to discriminate between crop and weeds (De Castro, Jurado-Expósito, Gómez-Casero, & López-Granados, 2012; Shapira, Herrmann, Karniel, & Bonfil, 2013), to discriminate between different wheat varieties or grades (Miralbès, 2008; Wang & Paliwal, 2006), classify plants as stressed vs. non-stressed (Liang et al., 2016; Moghimi, Yang, Miller, Kianian, & Marchetto, 2018), and to distinguish pest infected plants from healthy plants (Yuan et al., 2014; Zhang et al., 2012).

4.5 | Multivariate regression

Rather than assigning samples into distinct groups, regression analysis is a quantification method that relates the spectral response to a particular parameter (Figure 5). Regression methods decompose the high-dimensional data into valuable information and build either a linear or nonlinear relationship to the target parameter (Figure 6). The choice of which regression algorithm to use for hyperspectral data analysis depends on the nature of the sample, the benefits and limitations of the algorithm, the size of the dataset, and the required accuracy of prediction (Pan, Sun, Cheng, & Pu, 2016). The most common regression techniques applied to hyperspectral data include MLR, principal component regression (PCR), and PLSR.

Multiple linear regression has been used previously in wheat applications to quantify leaf water status (Das et al., 2017), assess freezing injury (Feng et al., 2018), and for selecting optimal wavelength regions for the assessment of growth, water relations, and ion contents of wheat irrigated with saline water (El-Hendawy et al., 2019a). Principal component regression has been used to estimate biomass (Yue, Feng, Yang, & Li, 2018), N stress response (Stellacci et al., 2012), and the protein and hardness of wheat kernels (Mahesh, Jayas, Paliwal, & White, 2015). Partial least squares regression has been used to estimate canopy N (Hansen & Schjoerring, 2003; Li, Mistele, Hu, Chen, & Schmidhalter, 2014c), water-soluble carbohydrates (Dreccer et al., 2014), chlorophyll content (Inoue et al., 2016), and photosynthetic traits (Silva-Perez et al., 2018). Other common multivariate algorithms for monitoring wheat properties also include SVM, random forest (RF), multivariate adaptive regression splines (MARS), and ANNs (Yao et al., 2015; Das et al., 2017).

As evident in Table 1, some methods can be used for either regression or classification tasks with small modifications. Other algorithms cannot be used interchangeably between regression and classification (e.g., linear regression or k-means clustering). Both regression and classification use past data to predict values of or make decisions regarding new data. However, in regression, the output variable is numerical (i.e., continuous) whereas classifications provide categorical (i.e., discrete) values. Nevertheless, there is some overlap between the two and common steps in their implementation. For example, classification may be used to predict a continuous value, but the result will be a probability for a class label. Equivalently, regression can predict a discrete value in the form of an integer quantity. Another major difference between classification and regression is the way their performance is quantified: classification is evaluated using an accuracy assessment (% of data correctly classified) whereas regressions are evaluated with error terms such as root mean square error (RMSE), ratio of performance to deviation (RPD), or coefficient of determination ($R^2$).

4.6 | Cross validation

Cross validation is an important step in all hyperspectral model development to help evaluate the quality of the models, help select a model that will generalize well to new data, and to avoid overfitting and underfitting. Common cross validation techniques include k-fold cross validation (Verrelst et al., 2016a; Lee, Chang, Anantrio Putra, Kim, & Hwan Kim, 2018), leave one out cross validation (LOOCV) (Wang et al., 2019; Abdel-Rahman et al., 2017) and stratified cross validation (Diamantidis, Karlis, & Giakoumakis, 2000). In k-fold cross validation, the subset of the data used for training is randomly split into k groups. The model uses each of these groups for validation when it runs (k times), averaging the results across all k replicates (Maxwell, Warner, & Fang,
FIGURE 6  Generalized workflow of hyperspectral modeling. Hyperspectral information from the sample plants are fed into a mathematical model alongside the reference values obtained through laboratory analysis. In future predictions, the reference values are not required, and traits can be predicted with the spectral information alone.

2018). The LOOCV is the specific case of \( k \)-fold cross validation where the number of groups is equal to the number of data observations and is particularly useful when a small number of observations are present; however, can be computationally intensive compared with other techniques (Cheng, Garrick, & Fernando, 2017). In stratified cross validation, the groups are stratified so that the mean response value is approximately equal for all groups.

4.7 | Three-dimensional models

The complex geometry and varying orientation of plants, as well as their interaction with external illumination and conditions, severely affects the spectral information obtained from hyperspectral sensors (Pinter et al., 2003; Axelsson et al., 2013; Behmann et al., 2016; Al Makdessi et al., 2019). In fact, up to 60% of the spectral information recorded can be due to plant geometry (Mahlein et al., 2017). This variation in reflectance can help be described by radiative transfer models, which aim to understand the light interception of plant canopies by attempting to quantify the amount of absorption and scattering (Jacquemond et al., 2009). The SAIL bidirectional reflectance model, first introduced by Verhoef (1984), is the most widely used model at the canopy scale, whereas PROSPECT is the most popular leaf-level model (Jacquemond & Baret, 1990). The PROSPECT model calculates reflectance and transmittance as a function of leaf chemistry and geometry by incorporating leaf structure, equivalent water thickness, chlorophyll content, and dry matter content (Verrelst et al., 2016b). The SAIL canopy model considers leaf area index, leaf angle distribution, the ratio of diffuse and direct radiation, a soil coefficient, sensor geometry, and leaf and soil reflectance spectra (Verrelst et al., 2016b). The combination of both the PROSPECT and SAIL models, known as PROSAIL (Baret, Jacquemond, Guyot, & Leprieur, 1992), is particularly effective for describing the variation of canopy reflectance through the incorporation of both leaf- and canopy-level parameters.

Another promising approach to overcome the effects of plant geometry, orientation, and illumination on the measured spectral signal of single plants is to create 3D hyperspectral models. This additional 3D spatial component provides further information on plant structure that cannot otherwise be visualized with 2D imaging alone. The simultaneous classification of spectral data with the structural segmentation of
**Table 1** Advantages and disadvantages of common approaches to hyperspectral analysis for both regression and classification problems

| Method          | Approach         | Type          | Advantages                                                                 | Disadvantages                                                                 |
|-----------------|------------------|---------------|----------------------------------------------------------------------------|------------------------------------------------------------------------------|
| k-means clustering | Classification | Unsupervised  | Simple to implement; computationally fast; results in tight clusters; flexible | Sensitive to outliers; manual selection of k; initial seeds and order of data will affect results; spherical clusters |
| LDA             | Classification   | Supervised    | Simple model; stable with highly separated classes and small datasets       | Linear models; assumes multivariate normal distribution                       |
| PCA             | Classification   | Unsupervised  | Deals with multicollinearity and large datasets; low noise sensitivity; adaptive; fast | Difficulty in interpreting resultant model outputs; linear assumption         |
| PLS-DA          | Classification   | Supervised    | Handles highly collinear and noisy data; easily interpretable output statistics | Requires strong cross-validation; prone to overfitting; performs poorly for small datasets |
| MLR             | Regression       | Supervised    | Relatively small dataset required; theory easy to understand; interpretable model outputs | Linear models only; sensitive to outliers; assumption of data independence; assumes normal distribution |
| PCR             | Regression       | Supervised    | Performs well with highly correlated or collinear data; intuitive           | Does not consider the response variable when selecting PCs (unsupervised)       |
| PLSR            | Regression       | Supervised    | Considers response variable (supervised); performs well with highly correlated or collinear data | Greater difficulty in interpreting resultant loadings                        |
| ANN             | Regression or classification | Supervised | Performs well with complex, nonlinear data; requires less formal statistics training; detects all possible interactions between predictor variables | Requires a large number of samples in dataset; “black box” nature; prone to overfitting |
| MARS            | Regression or classification | Supervised | Flexible; suitable for large datasets; computationally fast; no data assumptions; robust to outliers | Appropriate cross validation of model required; does not handle missing values; prone to overfitting |
| RF              | Regression or classification | Supervised | Handles nonlinear or complicated data; handles missing or noisy data; robust to overfitting and outliers | Complex visualization of output trees; algorithm parameters must be set appropriately |
| SVM             | Regression or classification | Supervised | Effective for nonlinear data; small training data; effective in high dimensional space | Algorithm parameters must be set appropriately; does not perform well when data is noisy, missing or has overlapping classes |

*Note.* ANN, artificial neural network; LDA, linear discriminant analysis; MARS, multivariate adaptive regression splines; MLR, multiple linear regression; PCA, principal component analysis; PCR, principal component regression; PLS-DA, partial least squares discriminant analysis; PLSR, partial least squares regression; RF, random forest; SVM, support vector machine.

3D data can enhance object identification, data extraction, and parameter estimation (Brell, Segl, Guanter, & Bookhagen, 2019).

The generation of 3D hyperspectral models is still in early stages and models are not fully developed yet. Only a few 3D hyperspectral systems have been implemented and still rely heavily on advanced instrument design and complex preprocessing. According to Mahlein et al. (2017), there are four stages to the successful generation of hyperspectral 3D plant models: acquiring the hyperspectral images, calibrating the hyperspectral camera against a reference, measuring the 3D geometry of each corresponding plant, and combining the spectral and spatial information. Three-dimensional hyperspectral models have a strong potential to be incorporated in future studies, particularly in high-throughput phenotyping systems for large-scale genomic studies. Such phenotyping systems already exist for capturing 2D hyperspectral images (e.g., Busemeyer et al., 2013; Virlet et al., 2016) and 3D RGB models (Ward et al., 2019) but a future step would be in combining these techniques to generate 3D hyperspectral models in a high throughput manner.
### 5. HYPERSONTAL APPLICATIONS FOR YIELD-LIMITING FACTORS IN WHEAT

Agricultural, horticultural, viticulture, and forestry industries have all applied hyperspectral methods to quantify various vegetation parameters. Vegetation studies utilizing hyperspectral data are too numerous to review as a whole, in regard to both the type of vegetation and the targeted parameters. As such, wheat was selected as an example crop in which to review the use of hyperspectral techniques for specifically quantifying yield-limiting factors. Wheat is the major winter crop grown in Australia with approximately 12.2 million ha of land dedicated to wheat in the 2016–2017 season and a production of 31.8 million Mg (ABARES, 2017). Worldwide, wheat is the staple food for more than 35% of the human population, providing 19% of our total available calories (FAOSTAT, 2016). Obtaining the highest possible yields for major crops on existing agricultural land is crucial for achieving global food security with a 60% increase in wheat production required by 2050 (FAOSTAT, 2013).

Wheat yields can be improved by addressing one or more limiting factors such as resource use efficiency, including N, or water use efficiency (Sadras & Angus, 2006; Mueller et al., 2012; Hochman & Horan, 2018) via improved management practices or breeding. The need for a nondestructive and rapid method for assessing these yield-limiting factors has seen the increasing implementation of hyperspectral methods. If identified early in the season, management practices can be applied to maximize potential yields and varieties with increased resource use efficiency can be targeted by long-term breeding programs.

#### 5.1 | Nitrogen content

The majority of previous research applying hyperspectral methods to wheat has focused on quantifying N levels. In fact, a meta-analysis of 157 papers involving the hyperspectral assessment of crop parameters revealed that wheat was the most common crop species studied (34% of all studies) whereas N was the target parameter in 50% of all publications (Bruning, unpublished data, 2016). This strong emphasis on wheat N levels is likely due to the significance of N to overall crop performance and the strong desire to find a nondestructive and reproducible alternative to traditional N measurements.

Early studies by Stone et al. (1996) found good correlations between the N concentration in wheat plants and normalized difference indices based on the 671- and 780-nm wavelengths. Mistlele and Schmidhalter (2008) measured the reflectance of a wheat canopy and found that the REIP was highly correlated to crop N status (coefficient of determination, $R^2 = .95$).

Also taking measurements at the canopy scale, Rodriguez, Fitzgerald, Belford, and Christensen (2006) evaluated several reflectance indices as an indication of N stress and found that the canopy chlorophyll content index (CCCI) could explain 68% of the variation in N stress regardless of the level of water stress or canopy density. Yao, Zhu, Tian, Feng, and Cao (2010) and Yao et al. (2013) further explored and compared hyperspectral indices for leaf N accumulation and uptake in wheat and determined that the majority of the sensitive spectral bands were located within the visible and NIR regions. In particular, the best spectral index for estimating leaf N accumulation was the normalized difference spectral index (NDSI, R860, R720), whereas N uptake could be estimated with the highest precision using similar wavelengths (NDVI, 807, 736). Guo et al. (2017) developed a red-edge index, which was able to predict leaf N uptake in winter wheat with $R^2 = .83$ and RMSE = 1.56 g m$^{-2}$. Hansen and Schjoerring (2003) used PLSR and all of the two-band NDVI combinations between 438 and 884 nm to measure N status in winter wheat. In their study, PLSR showed significant improvements over the various NDVI indices for prediction, suggesting PLSR may prove to be a better method for predicting N than VIs.

More recently, there has been a shift toward using PLSR or other multivariate techniques to quantify wheat N levels. Vigneau et al. (2011) used a hyperspectral camera mounted on a tractor gantry to estimate leaf N concentration from reflectance (400–1000 nm) spectra in wheat plants. Using PLSR models and a pooled dataset of greenhouse and field data, they predicted N content with $R^2 = .875$ and standard error of prediction (SEP) = 0.496%. Yao et al. (2015) assessed six retrieval algorithms for estimating leaf N content from canopy reflectance of winter wheat and found that the SVMs using the first derivative canopy spectra produced the highest association and accuracy (validation coefficient of determination, $R^2$-val = .78; root mean square error of prediction, RMSEP = 0.3%). At the leaf scale, Ecarnot, Compan, and Roumet (2013) assessed leaf N content of fresh and dried plants of durum wheat. Their PLSR model was able to accurately predict N for both fresh and dry leaves regardless of the phenologic stage (standard error of cross-validation, SECV = 0.215–0.320% and coefficient of determination in calibration ($R^2$-cal) ranging from .932 to .958).

#### 5.2 | Nutrient levels

In addition to N, hyperspectral sensing has been used to extract information on other wheat nutrients such as P, K, S, Mg, and Ca. Ayala-Silva and Beyl (2005) used hyperspectral methods to determine whether deficiency in P, K, Ca, and Mg, as well as N, altered the spectral reflectance properties of wheat leaves. They found that all nutrient deficiencies increased the reflectance in the visible range.
while causing a shift in the position of the red edge. Xu et al. (2015) used hyperspectral data to estimate the C/N ratio in a wheat canopy and achieved an $R^2 = .74$ with RMSE = 0.99. Mahajan et al. (2014) obtained leaf and canopy reflectance of wheat and proposed new indices for the prediction of N, P, S, and K. Importantly, they found that although indices gave a high and significant precision of nutrient content, models were improved when biomass-based nutrient contents were used instead of concentrations. This supports the earlier work of Pimstein et al. (2011) who found a significant improvement in the prediction of K and P using hyperspectral reflectance when total nutrient content was considered rather than relative content.

Models developed to predict micronutrients (B, Cu, Fe, Mn, and Zn) generally have lower accuracies than those for macronutrients (N, Ca, K, Mg, P, and S). Certainly, Abdel-Rahman et al. (2017) achieved lower accuracies for PLSR models for the prediction of micronutrients than for macronutrients in Swiss chard. Similarly, Pandey et al. (2017) were unable to quantify micronutrients as well as the macronutrient levels in maize and soybean using PLSR. On the other hand, N predictions are strong because models are usually based on foliar chlorophyll, an accurate, reliable, indirect estimate of plant N (Filella, Serrano, Serra, & Peñuelas, 1995). One reason that micronutrient models have poorer performance is likely their lower tissue concentrations, more subtle spectral expressions and smaller signal/noise ratio (Van Maerschalkenweerd & Husted, 2015). Micronutrient predictions can be improved if spectra are collected on dried, ground plant material rather than fresh or in situ leaves (Huang, Han, Yang, & Liu, 2009). Drying and grinding removes the spectral expression of water and homogenizes the sample. However, this introduces a sample preparation step, impeding the rapid nature of hyperspectral methods.

### 5.3 Water content

Tucker (1980) and Gausman (1985) were among the first to examine the influence of water content on visible and NIR reflectance and found that NIR wavebands were strongly affected by plant water content. Since then, several water indices have been developed to estimate water content parameters in a variety of crop types including the water band index (WBI) (Peñuelas, Filella, Biel, Serrano, & Savé, 1993), the normalized difference water index (NDWI) (Gao, 1996) and the normalized difference matter index (NDMI) (Wang, Qu, Hao, & Hunt, 2011).

Several studies have used hyperspectral systems to specifically predict various water parameters in wheat, utilizing both imaging and nonimaging techniques. Lelong, Pinet, and Poilvé (1998) collected airborne hyperspectral images to detect water deficiency in a wheat crop using only 12 ground-truth measurements. Liu et al. (2004) collected canopy spectra to estimate wheat plant water content using red edge parameters and found strong correlation between red edge width and plant water content ($R^2 = 0.91$ and RMSE = 0.99. Das et al. (2017) collected hyperspectral signatures of wheat plants to estimate the relative water content as an indicator of water-deficiency and developed a PLSR model with $R^2_{val} = .91$ and RMSE = 0.9%. Chattaraj et al. (2013) derived VIs from hyperspectral data to estimate crop coefficients to aid irrigation scheduling and found that the soil adjusted vegetation index (SAVI) was superior to NDVI for retrieval of LAI. Compared to NDVI, SAVI did not saturate until much higher biomasses because it accounts for partially vegetated canopies by including a soil brightness factor in its calculation, allowing dynamic soil–vegetation behavior to be modeled throughout the growing season (Huete, 1988).

### 5.4 Salinity

Variations in plant spectra have been related to differences in biophysical and biochemical characteristics caused by osmotic and ionic changes due to salinity. Sytar et al. (2017) reviewed imaging systems, particularly hyperspectral techniques, for detecting changes in plants caused by salt stress at the plant and canopy level. For wheat studies, Hackl, Mistele, Hu, and Schmidhalter (2013) analyzed the potential to detect salt stress-related traits of spring wheat cultivars grown in pots or in a close-to-field container platform. They found that simple indices derived from visible and NIR wavelengths showed significant correlation with various agronomic parameters of wheat plants grown under salinity stress. Moghimi et al. (2018) developed a novel processing pipeline to quantitatively rank salt stress tolerance in different wheat lines by collecting and analyzing hyperspectral images in the 400–900 nm range. They were able to detect tolerant wheat lines as early as 1 d after salt treatment. El-Hendawy et al. (2019a) spectrally assessed the growth, water relations, and ion contents of wheat under simulated saline field conditions. Through the acquisition of canopy hyperspectral signatures, they were able to establish that shoot dry weight, water relations, and ion content were effective as screening criteria for evaluating the salt tolerance of wheat cultivars.

### 5.5 Other properties

Other yield-limiting factors in wheat have also been assessed with hyperspectral data including various abiotic and biotic stresses. These include the presence of disease (Bauriegel & Herppich, 2014; Whetten, Hassall, Waine, & Mouazen, 2018), insect infection (Nansen, Macedo, Swanson, & Weaver, 2009), freezing stress (Feng et al., 2018), fungal
Hyperspectral methods offer high-throughput approaches to quantify or detect traits of hundreds or thousands of different genotypes. This is particularly relevant for plant breeding programs, which still rely heavily on destructive, costly, wet chemistry analysis to determine nutritional value or performance of different genotypes. These disadvantages of laboratory analyses prevent their widespread application in breeding, particularly in early stages where a large number of genotypes are being screened (Osborne, 2006). Hyperspectral methods can predict plant traits in these early generations that would otherwise be unfeasible with traditional techniques (Wiegmann et al., 2019). The early application of NIR spectroscopy methods in breeding trials focused on predicted end-use quality traits (Williams, Preston, Norris, & Starkey, 1984; Rubenthaler & Bruinsma, 1978; Delwiche, 1995; Dowell, Maghirang, & Baenziger, 2009). More recently, hyperspectral methods have been used at the crop canopy level to estimate yield (Montesinos-López et al., 2017a), perform disease assessment (Yu et al., 2018), and quantitatively rank the salt tolerance of wheat breeding lines (Moghimi et al., 2018). Hyperspectral imaging systems have also been mounted to ground-based platforms to acquire field data to help accelerate the breeding process. (Busemeyer et al., 2013; Virlet, Sabermanesh, Sadeghi-Tehrani, & Hawkesford, 2017; Deery, Jimenez-Berni, Jones, Sirault, & Furbank, 2014). However, the cost of systems, weather and environmental variability, the heritability of data, and the complexities of data processing and analysis currently limit the widespread application of hyperspectral techniques in breeding programs.

Another emerging field is in the integration of hyperspectral data into genomic selection models. Aerial hyperspectral data has been used alongside genomic data to model the genetic main effects and genotype × environment interactions within a wheat breeding program (Krause et al., 2019). They found that models incorporating hyperspectral data performed similarly to or better than marker and pedigree-based genomic selection models. Similarly, Montesinos-López et al. (2017b) incorporated hyperspectral imaging data with pedigree and genomic information to assess hyperspectral × environment interactions. They were able to identify important hyperspectral wavelength × environment interactions and demonstrated that combining hyperspectral data with genomic selection shows strong promise for candidate selection.

6 | LIMITATIONS OF PREVIOUS HYPERSONSPECTRAL STUDIES

While hyperspectral methods can provide near-instant results without destructive sampling, not all methods are adequately validated. A stronger emphasis is required on verifying methods, assessing data ranges (i.e., nutrient concentration ranges) and the extent and transferability of models. One critical aspect of previous hyperspectral work that is rarely tested is the specificity and transferability of the calibrations and methods developed. Most techniques are developed for a specific crop, species, or environment and their ability to be applied to other circumstances is often not assessed. Transferability across environments is crucial for any practical field use since field conditions vary significantly (e.g., soil type, fertilization, nutrient supply and levels, etc.). The growth conditions and physiological stage of crops, which have a high influence on the nutrient concentration and distribution of crops, also need to be considered and assessed in future model development.

For many plant parameters, particularly nutrient prediction, models developed from hyperspectral data are usually indirect. This means that nutrients are not directly measured but are based on some secondary physiological effect of the nutrient status. Other abiotic or biotic factors, such as water stress or disease presence, may also produce the same physiological effect as nutrient status. Therefore, models must be thoroughly investigated to ensure that they are directly and exclusively measuring information on nutrient levels. Van Maarschalkerweerd and Husted (2015) reviewed 13 previous articles using VIS-NIR calibrations to determine a broad range of essential nutrients in plants and found that not one of the reviewed articles gave an indication of the exact compounds being detected. Greater focus is needed on the specific compounds or processes being detected to ensure secondary deficiencies to that in question are not influencing the measurements.

The coefficient of determination is the most widely used statistic to assess the performance of hyperspectral models.
However, it can be affected by several factors, some of which are more closely associated with the original dataset than the model predictions. The range of values within the calibration dataset has a large influence on the $R^2$ of developed models; correlations will be stronger if there is more variability in the data. The $R^2$ value is also affected by the arrangement of values within the range (Cornell & Berger, 1987). Even though other statistics such as RPD and RMSE are often used alongside $R^2$, the valid concentration ranges must be investigated when developing hyperspectral models for estimating plant properties (Van Maarschalkerweerd & Husted, 2015).

### 7 TECHNICAL CHALLENGES OF HYPERSPECTRAL DATA

Hyperspectral techniques have many advantages over traditional measurement methods including being nondestructive, requiring no sample preparation, having rapid measurement times, and, for hyperspectral imaging systems, their ability to give a visual representation of plant biochemistry. However, hyperspectral imaging remains costly compared with other imaging techniques such as RGB cameras. Therefore, hyperspectral techniques are often used as an initial, exploratory phase to identify significant wavelengths. These wavelengths can then be used to design a multispectral system containing fewer bands but still able to capture the relevant information at a lower cost.

Despite its many advantages, hyperspectral images contain substantial data, much of which is redundant, posing considerable computational challenges. Further to the additional computing costs, the time taken to capture a hyperspectral image is significant compared with other imaging techniques. However, the exact time taken depends on the scanning speed of the particular instrument and the required resolution. Furthermore, hyperspectral techniques suffer from the fact that extracted data contains information from numerous, overlapping bands, which can cause the problem of multicollinearity. Multicollinearity can only be reduced with appropriate preprocessing and subsequent analysis techniques but can never be completely removed (ElMasry & Sun, 2010).

A component of the signal measured by hyperspectral instruments may relate to internal light scattering, which is inadvertently added during acquisition. The measured reflectance is strongly affected by the distance between the plant and the camera and light source and is also complicated by the structure and surface of leaves, the 3D geometry of plants and self-shading, all of which influence the reflectance of the underlying biochemical properties (Al Makdessi et al., 2019; Axelsson et al., 2013; Pinter et al., 2003). These effects must be overcome so that the reflectance is characteristic of the underlying biochemical properties rather than these illu-

### 8 FUTURE APPLICATIONS AND DIRECTIONS

Several challenges must be overcome to advance hyperspectral techniques in plant science beyond current applications. Of priority, bidirectional reflectance properties of canopies must be better understood and accounted for. Even basic relationships between LAI and NDVI change significantly with solar illumination angles, sensor viewing direction, or plant row orientation (Pinter et al., 2003). An improved understanding of bidirectional effects will make the measured spectral characteristics less dependent on these factors.

Hyperspectral techniques to estimate particular nutrient levels in plants are challenged by issues relating to secondary nutrient levels and transferability across environments. The ratios of different nutrient levels may be more crucial to plant health than the concentration of a single specific nutrient. Therefore, a hyperspectral technique that can simultaneously determine the concentration of multiple nutrients within a plant may provide a better indication of plant health. Additionally, being able to estimate multiple nutrients with a single analysis will save time and money. Therefore, future hyperspectral techniques should aim to develop methods or sensors that target multiple nutrients and mixed-stresses.

Future hyperspectral applications in plant science will see a strong need for mobile sensor platforms. This will require technology development that is able to capture hyperspectral imagery from hand-held or ground-based platforms. In years to come, platforms housing hyperspectral sensors will also need to be able to apply treatments (e.g., nutrients or pesticides) to defined areas or even to single plants depending on the instantaneous analysis of the hyperspectral data. Automatic or autonomous robots and swarm technology have the greatest potential to achieve this goal.
9 | CONCLUSION

Nondestructively quantifying plant properties gives us an improved understanding of overall plant health during the season and utilizing this knowledge can provide an opportunity for improving final grain yields. Hyperspectral methods can provide significant advancement in quantifying, assessing, and monitoring biochemical and biophysical attributes of crops, which are otherwise impossible or unfeasible with traditional methods. They have the vast and superior advantage over other optical techniques since they can provide layered trait information within the same spectral region. However, suitable preprocessing and analytical methods are required in order to derive information related to foliar properties. Limitations preventing the widespread application of hyperspectral techniques include the high cost involved, the amount of computational power required for analysis and storage, and the added spectral influence from the varying illumination. These technical issues must be addressed before hyperspectral imaging becomes a common and reliable approach for assessing yield-limiting factors in wheat. One way to achieve this may be through the selection of key parts of the spectral information (i.e., significant wavelengths with strong correlation to plant properties of interest) to develop affordable, easily implemented sensors for use in the field.

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CONFLICT OF INTEREST

The authors declare there are no conflicts of interest.

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