

# High-Throughput Estimation of Crop Traits

*A review of ground and aerial phenotyping platforms*

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Crop yields need to be improved in a sustainable manner to meet the expected worldwide increase in population over the coming decades as well as the effects of anticipated climate change. Recently, genomics-assisted breeding has become a popular approach to food security; in this regard, the crop breeding community must better link the relationships between the phenotype and the genotype. While high-throughput genotyping is feasible at a low cost, high-throughput crop phenotyping methods and data analytical capacities need to be improved.

High-throughput phenotyping offers a powerful way to assess particular phenotypes in large-scale experiments, using high-tech sensors, advanced robotics, and image-processing systems to monitor and quantify plants in breeding nurseries and field experiments at multiple scales. In addition, new bioinformatics platforms are able to embrace large-scale, multidimensional phenotypic datasets. Through the combined analysis of phenotyping and genotyping data, environmental responses and gene functions can now be dissected at unprecedented resolution. This will

aid in finding solutions to currently limited and incremental improvements in crop yields.

## BACKGROUND

Worldwide demand for food will increase through 2050 and beyond due to the increasing global human population. This represents a huge challenge to crop researchers and agricultural policymakers because current yield gain rates will not be sufficient for the demands of population growth, while climate change will make the difficulty even greater. Today's DNA sequencing, marker-assisted breeding, transgenic technology, genome-wide association study (GWAS) approaches, and quantitative trait loci (QTL) identification have been applied, to a limited extent, to improve crop yields [1]–[4].

While it is now relatively easy to select for monogenic traits, current genome sequence datasets have not been sufficiently mined for more genetically complex (multigenic) performance characteristics, at least in part because of the lack of crop phenotypic information collected from real-world field situations. Furthermore, traditional crop growth analysis often involves destructive sampling that is time-consuming and prone to measurement error. At

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present, the lack of traits amenable to high-throughput analysis restricts our ability to more thoroughly explore the quantitative genetic basis of complex characteristics correlated to crop's growth status, yield, and adaptation to environmental stress conditions. Thus, high-throughput phenotyping methods need to evolve to match the advances achieved in genotyping technologies.

The relatively recent availability of high-throughput crop phenotyping methods has influenced developments in crop selection and improvement [5], [6], and previous reviews have presented new opportunities for using ground and aerial phenotyping platforms that carry different sensors [7], [8]. Compared with other recent publications, this article comprehensively introduces recent applications of high-throughput ground and aerial phenotyping platforms for the measurement of crop phenotyping traits in the field. It demonstrates how high-throughput crop phenotyping can benefit breeders and agronomists and help narrow the genome–phenome gap. In particular, it discusses the disadvantages and advantages of different sensors and their applications and analyzes how to quickly and effectively estimate crop traits using ground and aerial phenotyping platforms.

## CROP PHENOTYPING

Phenotyping has been an important part of crop and elite-variety selection since humans found the best traits of plant species for domestication [9]. The term *phenotyping* does not have a completely clear-cut definition [10]. *Phenotyping* has been described as the use of protocols and methodologies to obtain a particular trait linked to plant functions and structures, with characteristics ranging from cells to whole plant levels [11]. An individual genotype represents all genetic characteristics, whereas a phenotype may include selected measured or all conceivable characteristics [12]. A complicated interaction between environmental conditions and genotypes leads to the phenotypic performance of crops [13]. One genotype can express various phenotypes due to plasticity in response to the environmental conditions in which plants grow. Thus, crop performance as an expression of genetic background requires an exploration of the relationship between phenotypes and genetics [14].

A common approach in crop breeding is to select the best genotype based on a phenotypic expression under various environmental conditions [15], [16]. Some traits (Table 1) considered in the field are green-area indexes (GAIs) [17]–[27], chlorophyll content [17], [18], [25], [27]–[33], nitrogen content [8], [17], [27], [29], [34]–[38], plant density at emergence [39]–[45], ear density [46]–[52], grain number and size [53]–[56], fraction of absorbed photosynthetically active radiation (FAPAR) [57]–[61], staygreen/senescence [62]–[66], crop dynamics monitoring [19], [67]–[73], phenology (e.g., at anthesis) [74]–[90], canopy coverage [62], [91]–[97], plant diseases and pests [8], [98]–[106], canopy height [107]–[112], canopy temperature [113]–[123], leaf rolling [124]–[126], leaf angle [112], [127]–[130], leaf wilting

[131]–[133], lodging [134]–[145], chlorophyll fluorescence [6], [146]–[153], photosynthetic status [152], [154]–[160], biomass [23], [24], [107], [161]–[171], water content [114], [150], [172]–[179], grain quality [180]–[188], water use efficiency [168], [189]–[196], canopy structure [112], [197]–[204], weed infestation [41], [205]–[210], light use efficiency [211]–[218], nitrogen use efficiency [219]–[226], nitrogen nutrition index [227]–[233], and yield [170], [234]–[245].

Crop phenotyping aims to accurately and precisely obtain traits linked to crop growth status, yield, and resilience to environmental stress, at various scales ranging from cell to canopy [11]. To achieve this goal, scientists and engineers from crop breeding, electronic engineering, molecular biology, computer image processing, mathematics and statistics, and agronomy cooperate to develop high-throughput crop phenotyping platforms that are operational in the field. These crop phenotyping platforms use combinations of unmanned aerial vehicles (UAVs), robotics, remotely controlled systems, and image-processing and analysis technology to monitor crops' growth status and performance. Current field crop phenotyping platforms will continue to be developed for the simultaneous evaluation of multiple crop phenotyping traits for numerous plant species at various scales.

## SENSOR DEVELOPMENT FOR FIELD PHENOTYPING TRAITS

Enablers of the monitoring of crop phenotyping traits in the field include rapidly developing sensor technologies, including red–green–blue (RGB), multispectral, hyperspectral, and thermal cameras; photosynthesis and fluorescence sensors; stereo cameras; and lidar devices. These sensors are usually divided into leaf-level, near-canopy, and airborne sensors, based on their application scale for crop phenotyping traits. In general, RGB, multispectral, and hyperspectral technologies and thermal cameras are airborne sensors, while photosynthesis and fluorescence sensors are leaf-level instruments and stereo cameras and lidar are near-canopy devices.

## LEAF-LEVEL SENSORS

### PHOTOSYNTHESIS SENSORS

Photosynthesis sensors are passive instruments used in the visible spectral region to measure the response of a crop photosynthesis status to different stress treatments [154]. This technology has been widely used in physiological and ecological studies [152], [159]. Photosynthesis sensors can be used to study the response of crop photosynthesis under different environmental conditions to better understand crop photosynthetic adaptation mechanisms [154]. As an example, the LI-COR 6400 is an open system, which means that transpiration and photosynthesis estimations are based on the differences in water (H<sub>2</sub>O) and carbon dioxide (CO<sub>2</sub>) in an air stream flowing through the leaf cuvette [155]. The net photosynthesis is computed using (1) [246]

**TABLE 1. SENSORS FOR ESTIMATING CROP PHENOTYPING TRAITS UNDER FIELD CONDITIONS, WITH THE LEVEL OF POTENTIAL APPLICATION.**

PHENOTYPING TRAITS	SENSOR							POTENTIAL APPLICATION LEVEL										REFERENCE
	RGB	MULTI/HYPERSPECTRAL	THERMAL	PHOTOSYNTHESIS	FLUORESCENCE	STEREO	LIDAR	1	2	3	4	5	6	7	8	9	10	
GAI																		[17]–[27]
Chlorophyll content																		[17], [18], [25], [27]–[33]
Nitrogen content																		[8], [17], [27], [29], [34]–[38]
Plant density at emergence																		[39]–[45]
Ear density																		[46]–[52]
Grain number and size																		[53]–[56]
FAPAR																		[57]–[61]
Staygreen/senescence																		[62]–[66]
Crop dynamic monitoring																		[19], [67]–[73]
Phenology (e.g., anthesis)																		[74]–[90]
Canopy coverage																		[62], [91]–[97]
Plant diseases and pests																		[8], [98]–[106]
Canopy height																		[107]–[112]
Canopy temperature																		[113]–[123]
Leaf rolling																		[124]–[126]
Leaf angle																		[112], [127]–[130]
Leaf wilting																		[131]–[133]
Lodging																		[134]–[145]
Chlorophyll fluorescence																		[6], [146]–[153]
Photosynthetic status																		[152], [154]–[160]
Biomass																		[23], [24], [107], [161]–[171]
Water content																		[114], [150], [172]–[179]
Grain quality																		[180]–[188]
Water use efficiency																		[168], [189]–[196]
Canopy structure																		[112], [197]–[204]
Weed infestation																		[41], [205]–[210]
Light use efficiency																		[211]–[218]
Nitrogen use efficiency																		[219]–[226]
Nitrogen nutrition index																		[227]–[233]
Yield																		[170], [234]–[245]

$$sa = u_i c_i - u_o c_o, \quad (1)$$

where  $s$  is the leaf area ( $\text{m}^2$ ),  $a$  is the assimilation rate ( $\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ),  $c_o$  and  $c_i$  are the outgoing and incoming mole fractions of the  $\text{CO}_2$  ( $\text{mol CO}_2 \text{ m}^{-2} \text{ air}^{-1}$ ), and  $u_o$  and  $u_i$  are the incoming and outgoing flow rates ( $\text{mol s}^{-1}$ ). The transpiration is obtained using (2) [246]

$$sE = u_o w_o - u_i w_i sE = u, \quad (2)$$

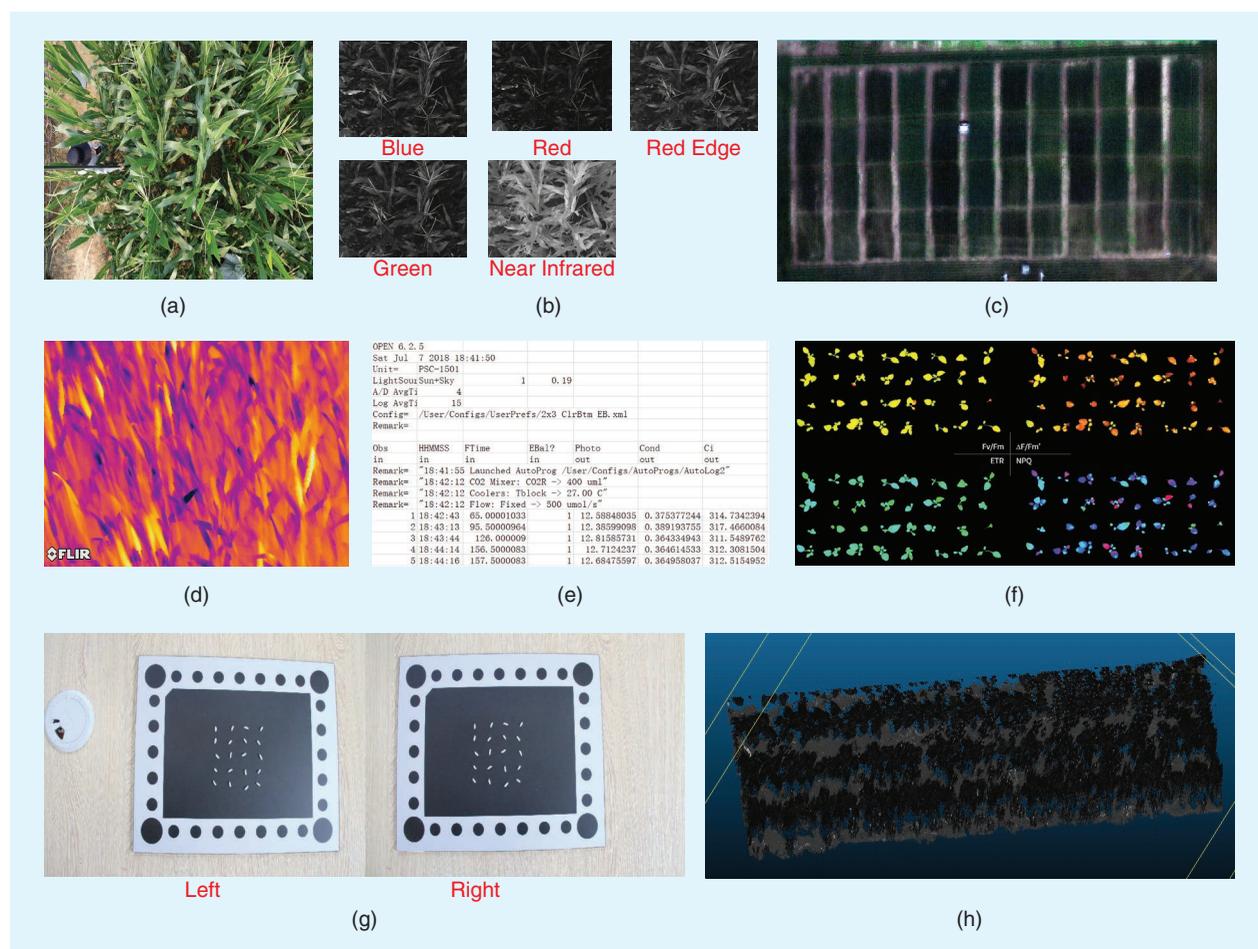
where  $E$  is the transpiration rate ( $\text{mol m}^{-2} \text{ s}^{-1}$ ) and  $w_o$  and  $w_i$  are the outgoing and incoming water mole fractions ( $\text{mol H}_2\text{O mol air}^{-1}$ ).

The LI-COR 6400 contains a computing and storage section; an infrared  $\text{CO}_2$  analyzer; and temperature, humidity, and light sensors. The moisture and  $\text{CO}_2$  of the LI-COR 6400 instrument should be cleared and checked to obtain a value equal to zero to calibrate the sensor before measuring crop leaves [157]. Figure 1(e) shows an example of cotton measured from an LI-COR 6400 instrument. The exported calculated parameters include the photosynthetic rate, conductance to  $\text{H}_2\text{O}$ , intercellular  $\text{CO}_2$  concentration, transpiration rate, and so forth [158]. These parameters can

be used to carry out studies of crop growth analyses, gas exchanges, and stable isotope examinations [159]. At present, photosynthesis sensors do not provide any images and cannot be combined with ground and aerial imaging platforms [156]. The specific model, sensor parameters, details, estimated phenotyping traits, imaging environment, and limitations of the photosynthesis sensor are summarized in Table 2. New technologies will be required for rapid, image-based photosynthesis sensors to better explore the response of crop photosynthesis to environmental changes in the future.

## FLUORESCENCE SENSORS

Fluorescence sensors are passive or active in the ultraviolet, visible, and near-infrared spectral regions, which are sensitive to fluorescence signals [153]. They are commonly used to detect the resilience of the crop metabolic status in stressed environments [147]. The fluorescence sensor provides an image for exploring the spatial patterns of the leaf photosynthetic status [151] and for the early detection of crop stress symptoms, as influenced by diseases and pests [98]. Fluorescence is light that is emitted in the ultraviolet, visible, and near-infrared spectral wavelengths



**FIGURE 1.** Example images from different sensors, including (a) an RGB camera, (b) a multispectral camera, (c) a hyperspectral camera, (d) a thermal camera, (e) a photosynthesis sensor, (f) a fluorescence sensor, (g) a stereo camera [254], and (h) lidar.

**TABLE 2. COMPARISON AND LIMITATIONS OF DIFFERENT SENSORS FOR FIELD CROP PHENOTYPING TRAITS.**

SENSOR TYPE	MODEL	SENSOR PARAMETERS	DETAILS	PHENOTYPING TRAITS	IMAGING ENVIRONMENT	IMAGING TECHNIQUES	LIMITATIONS
Photosynthesis sensor	LI-COR 6400	<ul style="list-style-type: none"> <li>✓ Sensor type: absolute, open-path, nondispersive infrared gas analyzer</li> <li>✓ Bandwidth: 10 Hz</li> <li>✓ Range: 0–3,100 <math>\mu\text{mol mol}^{-1}</math></li> </ul>	Passive sensor at the visible spectral region	Plant diseases and pests, lodging, chlorophyll fluorescence, photosynthetic status, biomass, water use efficiency, light use efficiency, nitrogen use efficiency, nitrogen nutrition index, yield	Controlled environment; field	No	<ul style="list-style-type: none"> <li>✓ Sensors are costly.</li> <li>✓ Measurements are time consuming.</li> <li>✓ Only measured point data are obtained.</li> <li>✓ It is not easy to measure at the canopy scale.</li> <li>✓ No image.</li> <li>✓ It is not possible to mount on field crop phenotyping platforms.</li> </ul>
Fluorescence sensor	PlantExplorer	<ul style="list-style-type: none"> <li>✓ Sensor type: charge-coupled device</li> <li>✓ Image resolution: 1.3 megapixels</li> <li>✓ Spectral range: 350–1,000 nm</li> <li>✓ Frame rate: 20 images per second</li> </ul>	Passive or active sensor at the ultraviolet, visible, and near-infrared spectral regions	Plant diseases and pests, lodging, chlorophyll fluorescence, photosynthetic status, biomass, grain quality, light use efficiency, nitrogen use efficiency, nitrogen nutrition index, yield	Controlled environment; field	Fluorescence imaging	<ul style="list-style-type: none"> <li>✓ Difficult to measure at the canopy scale due to low signal-to-noise ratio.</li> <li>✓ Difficult to implement on field crop phenotyping platforms.</li> <li>✓ It will be strongly influenced by background noise.</li> </ul>
Stereo camera	Stereolabs ZED	<ul style="list-style-type: none"> <li>✓ Sensor resolution: dual 4-megapixel sensors with 2-<math>\mu\text{m}</math> pixels</li> <li>✓ Sensor format: native 16:9 format for a greater horizontal field of view</li> <li>✓ Sensor size: 1/3-in backside illumination sensors with high low-light sensitivity</li> </ul>	Camera with two or more lenses, with a separate image sensor for each lens	Ear density, grain number and size, FAPAR, crop dynamic monitoring, phenology, canopy coverage, canopy height, leaf rolling, leaf angle, lodging, biomass, canopy structure, weed infestation, yield	Controlled environment; field	3D imaging	<ul style="list-style-type: none"> <li>✓ Experimental conditions influence its performance.</li> <li>✓ Resolution of images is lower.</li> <li>✓ Field application is limited.</li> <li>✓ Sensor needs calibration before use.</li> </ul>
Lidar	Sick LMS400-2000	<ul style="list-style-type: none"> <li>✓ Light source: visible red light (650 nm)</li> <li>✓ Laser class: 2</li> <li>✓ Aperture angle: horizontal 70°</li> <li>✓ Scanning frequency: 230–500 Hz</li> <li>✓ Angular resolution: 0.1–1</li> <li>✓ Working range: 0.7–3 m</li> </ul>	Active sensor that detects the distance to a target from pulses of laser light	GAI, ear density, grain number and size, FAPAR, crop dynamic monitoring, phenology, canopy coverage, canopy height, leaf rolling, leaf angle, lodging, biomass, canopy structure, weed infestation, yield	Controlled environment; field	Laser imaging	<ul style="list-style-type: none"> <li>✓ Sensitive to small differences in path length.</li> <li>✓ High cost of the sensor.</li> <li>✓ Sensor should be integrated with GPS for georeferencing information.</li> <li>✓ Data processing is time consuming.</li> </ul>

*Continued*

**TABLE 2. COMPARISON AND LIMITATIONS OF DIFFERENT SENSORS FOR FIELD CROP PHENOTYPING TRAITS.**

SENSOR TYPE	MODEL	SENSOR PARAMETERS	DETAILS	PHENOTYPING TRAITS	IMAGING ENVIRONMENT	IMAGING TECHNIQUES	LIMITATIONS
Digital camera (RGB)	Sony Alpha 6000	<ul style="list-style-type: none"> <li>✓ Sensor: Advanced Photo System Type C (23.5 × 15.6 mm)</li> <li>✓ Aspect ratio: 3:2</li> <li>✓ Image processing: Boinz X</li> <li>✓ Image resolution: 6,000 × 4,000</li> <li>✓ Focal length lens: 60 mm</li> </ul>	<ul style="list-style-type: none"> <li>✓ Gray value [digital number (DN)]</li> <li>✓ Color image</li> <li>✓ Texture information</li> </ul>	<ul style="list-style-type: none"> <li>• GAI, chlorophyll content, nitrogen content, plant density, ear density, grain number and size, FAPAR, staygreen/senescence, crop dynamic monitoring, phenology, canopy coverage, plant diseases and pests, canopy height, leaf rolling, leaf angle, leaf wilting, lodging, biomass, canopy structure, weed infestation, nitrogen use efficiency, nitrogen nutrition index, yield</li> </ul>	Controlled environment; field	Visible light imaging	<ul style="list-style-type: none"> <li>✓ Limited to visual three spectral bands.</li> <li>✓ Only relative value is obtained.</li> <li>✓ Images do not carry out calibration.</li> <li>✓ Effect of illumination condition on image acquisition quality limits image processing.</li> </ul>
Multispectral camera	MicaSense RedEdge-M	<ul style="list-style-type: none"> <li>✓ Bands: blue (475 ± 20 nm), green (560 ± 20 nm), red (668 ± 10 nm), near infrared (840 ± 40 nm), red edge (717 ± 10 nm)</li> <li>✓ Aspect ratio: 4:3</li> <li>✓ Image resolution: 1,280 × 960</li> <li>✓ Focal length lens: 5.4 mm</li> </ul>	<p>Several spectral bands of each pixel at the visible and infrared spectral regions</p>	<ul style="list-style-type: none"> <li>• GAI, chlorophyll content, nitrogen content, plant density, FAPAR, staygreen/senescence, crop dynamic monitoring, phenology, canopy coverage, plant diseases and pests, canopy height, leaf rolling, leaf wilting, lodging, biomass, water content, grain quality, water use efficiency, canopy structure, nitrogen use efficiency, nitrogen nutrition index, yield</li> </ul>	Controlled environment; field	Visible and near-infrared imaging	<ul style="list-style-type: none"> <li>✓ Limited to several spectral bands.</li> <li>✓ Spectral data should be frequently calibrated using referenced objects.</li> <li>✓ Effects of camera geometrics, illumination condition, and sun angle on the data signal.</li> </ul>
Hyperspectral camera	Specim FX10	<ul style="list-style-type: none"> <li>✓ Spectral range: 400–1,000 nm</li> <li>✓ Spectral bands: 224</li> <li>✓ Spectral full width at half maximum: 5.5 nm</li> <li>✓ Spatial sampling: 1,024 pixels</li> <li>✓ Frame rate: 330 frames per second, full frame</li> <li>✓ Aperture: F/1.7</li> <li>✓ Camera signal-to-noise ratio: 600:1</li> </ul>	<p>Discrete or continuous spectral bands of each pixel at the visible and infrared spectral regions</p>	<ul style="list-style-type: none"> <li>• GAI, chlorophyll content, nitrogen content, FAPAR, staygreen/senescence, crop dynamic monitoring, phenology, canopy coverage, plant diseases and pests, leaf rolling, leaf wilting, lodging, biomass, water content, grain quality, water use efficiency, canopy structure, nitrogen use efficiency, nitrogen nutrition index, yield</li> </ul>	Controlled environment; field	Hyperspectral imaging	<ul style="list-style-type: none"> <li>✓ Image dataset processing is challenging.</li> <li>✓ Price of the sensor is expensive.</li> <li>✓ Spectral data should be frequently calibrated using referenced objects.</li> <li>✓ Effects of camera geometrics, illumination condition, and sun angle on the data signal.</li> </ul>
Thermal camera	Flir Vue Pro 336	<ul style="list-style-type: none"> <li>✓ Spectral range: 7.5–13.5 μm</li> <li>✓ Focal length lens: 6.8 mm</li> <li>✓ Image resolution: 336 × 256</li> <li>✓ Thermal imager: uncooled vanadium oxide microbolometer</li> </ul>	<p>Temperature of each pixel at the thermal spectral infrared regions</p>	<ul style="list-style-type: none"> <li>• GAI, canopy coverage, plant diseases and pests, canopy temperature, leaf rolling, leaf wilting, lodging, water content, water use efficiency, yield</li> </ul>	Controlled environment; field	Thermal imaging	<ul style="list-style-type: none"> <li>✓ Experimental conditions influence performance.</li> <li>✓ Difficult to detect very small temperature differences.</li> <li>✓ Price of the high-resolution sensors is expensive.</li> <li>✓ Small-volume and low-weight sensors suffer during temperature shifts.</li> <li>✓ Sensor needs to be timely calibrated.</li> </ul>

during radiation absorption [148]. Irradiating chloroplasts with actinic or blue light will produce some reemission of absorbed light by chlorophyll [149]. Compared with incident radiation, the fraction of reemission light is variable and relies on the crop's ability to convert harvested light to metabolic activity [247]. The reemitted light is termed *fluorescence*, and it is a good indicator of a crop's capacity to assimilate actinic light [247].

Furthermore, the combination of actinic light and brief, saturating blue pulses can be used to measure the efficiency of photoassimilation, nonphotochemical quenching, and other physiological crop parameters [247]. Fluorescence sensors use a charge-coupled-device camera that is very sensitive to fluorescence signals, where fluorescence signals occur by illuminating crops with ultraviolet or visible light using pulsed lasers, pulsed flashlight lamps, or light-emitting diodes [150]. The image pixel value of the fluorescence indicators is presented by a false color code ranging from zero to one [248]. Ultraviolet illumination produces two kinds of fluorescence, that is, blue-to-green and red-to-far-red regions, which is the principle of multi-color fluorescence images [248]. This method can capture the simultaneous fluorescence emission from four spectral bands [blue (440 nm, F440), green (520 nm, F520), red (690 nm, F690), and far-red (740 nm, F740)] by excitation with a single wavelength [249]. The fluorescence sensor does not need to be calibrated before measuring crops, but it is better used under uniform illumination conditions to reduce the effect of the light source on the measurement results [250].

Figure 1(f) presents an example of soybeans provided through PlantExplorer, and the corresponding exporting fluorescence parameters include the initial fluorescence ( $F_0$ ), maximum fluorescence ( $F_m$ ), photosynthetic system 2 original light-energy conversion efficiency ( $F_v/F_m$ ), photosynthetic quantum yield, photochemical-quenching coefficient, nonphotochemical-quenching coefficient, apparent electron transfer rate, and so forth [251]. These parameters can be used to carry out studies of crop stress analyses, photosynthetic functions, and chloroplast content estimations [250].

Current fluorescence sensors are focused mainly at the leaf scale. Chlorophyll fluorescence at the canopy scale is restricted by sensor and background noise, decreasing the signal-to-noise ratio [150]. Table 2 contains more information about photosynthesis sensor models, parameters, phenotype estimation traits, imaging environments, imaging techniques, and limitations. Therefore, fluorescence sensors are generally not available for integration with ground and aerial phenotyping platforms. However, chlorophyll fluorescence sensing on ground phenotyping platforms was used to assess the nitrogen status and biomass of wheat, barley, and colza [146], [147], [252]. There is a need for further development of fluorescence sensors and protocols for fluorescence image-based, high-throughput crop phenotyping to enhance field applications in the future.

## NEAR-CANOPY SENSORS

### STEREO CAMERAS

A stereo camera has two or more lenses, with separate imaging sensors for each [167]. Structures are observed in 3D because of the parallax effect of human eyes [253]. Due to the distance between a person's eyes, the two images formed at the fundi are basically similar but slightly different, and there is a certain disparity [130]. The brain then computes a 3D image. Stereo cameras with charge-coupled device chips take advantage of the parallax characteristics of both eyes [46]. In general, the stereo camera is very similar to the traditional digital camera but with two or more lenses. Some scientists use two identical, unmodified cameras to obtain 3D images [130], [254].

To maintain the accuracy and stability of a stereo camera, the device should be calibrated during experiments [254]. The calibration of a stereo camera is performed as follows:

- 1) The instrument needs to be tuned to obtain internal and external parameters and the homographic matrix.
- 2) The calibration results are used to correct the original image so that the two adjusted pictures are located on the same plane and parallel to each other.
- 3) The two corrected images must be matched according to the same pixel.

Figure 1(g) provides example rice seed images from a pair of cameras (Canon EOS 7D) [254]. The exported data of a stereo camera contain RGB images and a 3D structure [130], [254] that can be used to obtain crop growth and development information [130].

The model, parameters, estimated phenotypic traits, imaging environments, imaging techniques, and limitations of the stereo camera are presented in Table 2. The advantage of the stereo camera is that it provides 3D structures at a relatively low cost, but the spatial resolution is limited and sensitive to variable outdoor illumination conditions [247]. These restrictions have become major challenges for the stereo camera's application to crop phenotyping in the field. Currently, the stereo camera is used mostly to obtain the 3D structure of single plants; with it, estimating canopy-level traits in the field remains difficult [130]. Improvements in the stereo camera's resolution and sensitivity to illumination will be needed in the future.

### LIDAR

Lidar has an active sensor that determines the distance to an object using pulses of laser light in the 600–1,000-nm region [129]. Lidar is an Earth-observation technology that can directly obtain the 3D coordinates of object surface points through data such as positions, distances, and angles, and it realizes the extraction of the surface information and the reconstruction of the 3D scene [112]. Lidar includes a single-beam narrowband laser and a receiving system. The laser generates and emits a light pulse, which hits the target object and reflects back to a receiver [199].

The receiver accurately measures the propagation time of the light pulse from emission to reflection [112]. Because the pulse travels at the speed of light, the receiver always obtains a reflected pulse before the next one is sent. Given that the speed of light is known, the time of flight can be converted into a measurement of distance [200].

The laser itself has a very accurate ranging ability, and its ranging accuracy can reach the millimeter level. In general, an airborne lidar system integrates three technologies: laser scanning, GPS/differential GPS, and an inertial navigation system. This combination can locate with high accuracy a laser beam spot hitting an object [255]. The principle of measuring the ground from a flying carrier concerns the “two-way” time measurement method [200]. That is to say, we need to record only the time between a laser signal’s emission and its return. Combined with other data, we can accurately calculate the  $x$ -,  $y$ -, and  $z$ -coordinates of the ground spot. Lidar does not need to be calibrated, but known referenced objects are necessary to accurately obtain crop structure information.

Figure 1(h) shows an example image of sunflowers that was acquired with a Sick LMS400. Lidar can provide a 3D point cloud to describe the crop canopy structure [129]. The point cloud can be used to monitor dynamic crop changes during the growing season [197], [200]. Table 2 describes a typical lidar model and its parameters, phenotype estimating traits, imaging environment, imaging techniques, and limitations. Compared with other sensors, lidar is less influenced by environmental conditions, and it has a relatively high estimation accuracy for several crop phenotyping traits because it is an active sensor [198]. However, the price of lidar is generally higher than that of passive sensors. Lidar is sensitive to small differences in the path length, although the data processing is more complex (Table 2). Lidar has been integrated with ground and aerial phenotyping platforms to estimate crop phenotyping traits [112], [197], [200], [220], [255]. However, in the future, the density of the achievable 3D point cloud needs to be increased to better describe 3D crop structures.

## AIRBORNE SENSORS

### RGB CAMERAS

RGB cameras record red, green, and blue spectral bands in the visible spectral regions [39]. An RGB camera produces digital images and can mimic human visible perception to obtain information for estimating crop phenotyping traits in plant breeding [74]. The most common implementation of RGB cameras is based on silicon sensors with charged-coupled devices or CMOS arrays that are very sensitive to visible bands of light and show images in 2D; this is the simplest imaging technique for crop phenotyping traits [247]. RGB camera images are typically shown in spatial matrices of intensity values (from zero to 255) corresponding to photon fluxes in blue, green, and red spectral bands of the visible region [46]. To obtain better high-quality RGB

images, a color calibration plate is commonly used to adjust images using algorithms to reduce the effect of different illumination conditions on the photo quality [256], [257].

Figure 1(a) gives an example image of maize taken with a Sony Alpha 7 camera. The RGB camera can export images that include gray value, color, and texture information [39]. The applications of RGB cameras for crop phenotyping traits are shown in Tables 1 and 2, including GAI, plant density, ear density, FAPAR, canopy coverage, canopy height, leaf rolling, leaf angle, lodging, and so on, during the whole crop growing season. In addition, a typical camera model and its parameters, phenotype estimating traits, imaging environment, imaging techniques, and limitations are presented in Table 2. RGB cameras have been widely applied to evaluate crop phenotyping traits in the field to provide measurements at an affordable price [46], [47], [162], [164], [258].

Compared with other sensors, the advantage of the RGB camera relates to its affordable price and high-spatial-resolution imagery [237]. However, it remains a huge challenge to get good image segmentation results when adjacent plant leaves overlap [39]. In addition, the gray values and color information are limited to three visual spectral bands. New technologies are required to solve these shortcomings and improve the application of the RGB camera for crop phenotyping trait estimation [7]. Currently, a modified RGB camera [the so-called color-infrared (CIR) camera] with near-infrared, red, and green bands may be applied to obtain spectral vegetation images when it is radiometrically calibrated [259], [260]. The pseudonormalized-difference vegetation index (pseudo-NDVI) from a CIR camera has been applied for crop phenotyping [7], [258], [261], [262]. CIR cameras can be used to obtain high-spatial-resolution imagery with a limited number of wide spectral bands at a relatively low cost. However, CIR cameras may be unstable, and they have a limited application, as they are not designed for precise radiometric measurements [263].

### MULTISPECTRAL AND HYPERSPECTRAL CAMERAS

Multispectral and hyperspectral cameras depend on the interaction between solar radiation and crops [71]. The reflectance of single leaves or canopies is low in the visible spectral ranges (400–700 nm) because solar radiation is absorbed by leaf pigments (such as chlorophyll), with a peak of reflectance in the green spectral region of roughly 550 nm [45]. The reflectance (so-called red-edge region) is sharply increased with the transition from the visible to near-infrared spectral region [32]. A large fraction of the incident solar radiation is reflected by leaves because of scattering within the leaf structure and mesophyll in the near infrared (700–1,200 nm) region [24]. Furthermore, near-infrared radiation is transmitted from the upper to the lower leaves of the canopy, which can reflect solar radiation back to the upper section of the leaf cover [168]. Therefore, the leaf thickness and growth status as well as the canopy architecture primarily influence the reflectance pattern in

this spectral region [27]. The reflectance is gradually decreased with increasing wavelengths of up to 2,500 nm because the absorption of solar radiation is increased by the water in crop leaves [59].

Multispectral cameras commonly include several spectral bands in the visible and infrared spectral regions [34]. Spectral bands are generally sensitive to leaf pigments and the leaf and canopy structure [45], [67]. Compared with their multispectral counterparts, hyperspectral cameras have a higher spectral resolution, with continuous or discrete spectral bands in the visible and infrared spectral regions [17], [264]. Hyperspectral bands are more sensitive to small changes in leaf pigments, such as carotenoids, chlorophyll a and b, and xanthophylls as well as leaf and canopy structures [265]. Multispectral or hyperspectral bands are used to obtain VIs through band calculations. More advanced methods based on radiative transfer model inversion techniques can also be employed [266].

To reduce the effects of illumination conditions on multispectral and hyperspectral cameras, radiometric image calibration should be carried out at each band [266]. First, several calibration targets ( $1 \times 1$  m) that have different reflectance values are placed in the UAV flight area. Second, the digital number (DN) of each target is collected from UAV multispectral and hyperspectral images, and the corresponding actual reflectance of each target is also measured using, for example, an ASD spectroradiometer [166]. Third, the regression relationship between the reflectance and the DN value is established at each band image through empirical linear or nonlinear methods, and the DN value in the images is converted to the normalized reflectance using established regression relationship equations [71].

Figure 1(b) and (c) gives examples of maize and wheat images acquired with MicaSense RedEdge-M and Cubert UHD185 cameras, respectively. Multispectral and hyperspectral cameras can export images that include DN values and texture, reflectance, and color information [45]. Crop phenotyping trait estimations from multispectral and hyperspectral cameras are presented in Tables 1 and 2, including GAI, plant density, ear density, FAPAR, canopy coverage, staygreen/senescence, crop dynamics monitoring, phenology, plant diseases and pests, and so forth, across the whole crop growing season. The models, parameters, phenotype estimating traits, imaging environments, imaging techniques, and limitations of multispectral and hyperspectral cameras are detailed in Table 2.

The advantage of multispectral and hyperspectral cameras is that they have spectral information (except texture and color information) that could be better used to estimate crop phenotyping traits through fusion algorithms (this is an image process of integrating related information from two or more images into a single image and could be applied to acquire more useful information from the input images) [59], [115]. The application of multispectral and hyperspectral cameras in the field of crop phenotyping traits has attracted the attention of breeding programs

[22], [103], [104], [267]. The main limitations concern the very large volume of data associated with spectral imagery and the generally more complex operation of hyperspectral cameras compared to multispectral and RGB cameras. These shortcomings impede their application and adaptation [36], [266].

In summary, multispectral cameras are mainly used for VI-based traits, due to the limited number of available bands, while hyperspectral cameras enable the calculation of more advanced crop phenotyping traits, such as photosynthetic status and fluorescence [265], [266]. With the rapid development of multispectral, hyperspectral, and computer image-processing technologies, cheaper multispectral and hyperspectral cameras will be developed and combined with high-performance graphics and computer-clustering technologies.

### THERMAL CAMERAS

Thermal cameras are used to measure infrared radiation in the thermal spectral infrared regions as a water stress indicator [115]. Thermal cameras operate in the spectral range from 3 to 14  $\mu\text{m}$ , and their most-used spectral wavelengths are 3–5  $\mu\text{m}$  and 7–14  $\mu\text{m}$ , respectively [71], [266], [268]. The infrared radiation atmospheric transmission is very close to its maximum value in these two spectral ranges [121]. The thermal sensitivity of the spectral range from 3 to 5  $\mu\text{m}$  is associated with higher energy levels than the spectral range from 7 to 14  $\mu\text{m}$ , but using wavelengths of 7–14  $\mu\text{m}$  has advantages for certain applications [269]. The wavelength ranges between 8 and 14  $\mu\text{m}$  can reduce errors from the atmospheric absorption of infrared radiation for objects through longer atmospheric paths [269].

Thermal cameras should be calibrated to maintain the measurement accuracy and stability of their images. First, a thermal camera is preheated for roughly 20 min in the field before a UAV flight to reduce the temperature drift, and the automated nonuniformity correction is enabled during the data measurement [270]. Second, according to a thermal camera's preheating guidelines, one JPEG image of a blackbody radiometric background scene is taken, which is immediately obtained from one 14-b TIFF-format image of the same scene [235]. Third, the blackbody radiometric JPEG is loaded into FLIR Tools software, which provides options for imaging target-related parameters and environmental conditions, to execute a radiometric conversion [271]. At the same time, the air humidity and temperature are added from a weather station, along with the target distance (flight height) and object emissivity to obtain temperature values that are output in a text file [272]. The bias between the blackbody and the image temperature is determined. Finally, each pixel's radiometric value in the 14-b TIFF image is correlated to the corresponding value in the text file, with a linear model being used as a radiometric conversion regression equation, and the radiometric value in the images is transformed into the temperature using the established regression relationship equation [235].

Figure 1(d) provides an example image of wheat obtained using a FLIR SC620 camera. Thermal cameras export images that include the temperature, radiometric values, and texture information [271]. The application of a thermal camera to crop phenotyping trait estimation is illustrated in Tables 1 and 2. Thermal camera models, parameters, phenotype estimating traits, imaging environments, imaging techniques, and limitations are shown in Table 2. In recent years, thermal cameras with a high thermal sensitivity have been widely applied to estimate crop growth statuses under water stress conditions [116], [120], [266], [273]. Spatial patterns obtained from thermal images may be used to carry out image segmentation to differentiate stressed and unstressed vegetation by using image-processing algorithms [273]. The cost of thermal cameras is generally higher than for multispectral cameras, and lightweight models are typically thermally unstable due to changing ambient conditions [268]. Thermal cameras can be utilized with ground and aerial phenotyping platforms to carry out the analysis of crop phenotyping traits.

Potential crop phenotyping trait applications have been scored based on the difficulty of the data acquisition and the available optical sensors (Table 1). The results show that the highest and lowest potential crop phenotyping application levels are canopy coverage and plant diseases and pests, respectively. The high potential-application level of crop phenotyping traits includes canopy coverage, GAI, plant density, staygreen/senescence, canopy height, ear density, weed infestation, biomass, lodging, yield, water content, chlorophyll content, nitrogen content, FAPAR, monitoring of crop dynamics, phenology (e.g., anthesis), canopy temperature, leaf rolling, leaf angle, leaf wilting, nitrogen use efficiency, nitrogen nutrition index, grain number and size, canopy structure, chlorophyll fluorescence, photosynthetic status, grain quality, light use efficiency, water use efficiency, and plant diseases and pests. These potential application levels of crop phenotyping traits can provide a guideline for prioritizing trait selection using high-throughput methods at the field scale.

Figure 2 demonstrates potential applications of key crop phenotyping traits with different sensors at the critical growth stages in a maize breeding program. The figure presents the phenological stages of maize, including emergence (VE, 1), the first leaf collar (V1, 2), the third leaf collar (V3, 3), the sixth leaf collar (V6, 4), the ninth leaf collar (V9, 5), tasseling (VT, 6), silking (R1, 7), and maturity (R6, 8). An RGB camera is used to estimate the plant density in VE (1), the tassel density in VT (6), and the yield component in R6 (8) through color and texture information and related image-processing algorithms [39], [55], [74], respectively. The dynamic monitoring period of RGB cameras extends from the V1 (2) to the R6 (8) growth stages. Crop growth, diseases, stress, senescence, structure, phenology, and so on can be dynamically monitored by RGB cameras.

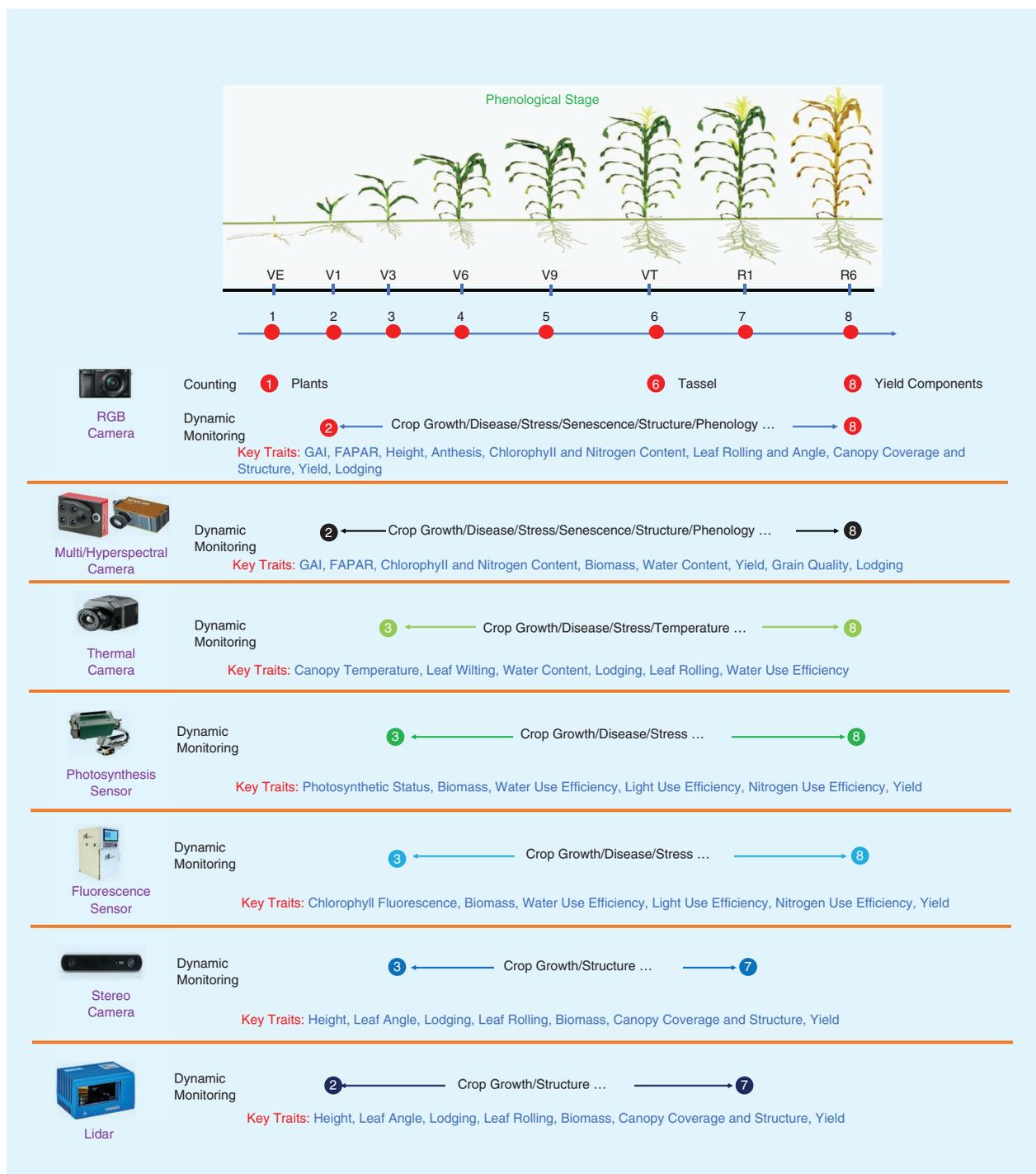
The key dynamic monitoring traits obtained by an RGB camera include GAI, FAPAR, height, anthesis, chlorophyll

and nitrogen content, leaf rolling and angle, canopy coverage and structure, yield, and lodging. The dynamic monitoring stage of a multi/hyperspectral camera is consistent with that of an RGB camera, but key dynamic monitoring traits differ between the devices. For example, multi/hyperspectral cameras can be used to detect crop water content [116], [266] and grain quality [64], [274] through spectral characteristics and machine learning methods, with the exception of the same key dynamic monitoring traits from an RGB camera. The dynamic monitoring stage (3–8) of a thermal camera is less than that (2–8) of RGB and multi/hyperspectral cameras because thermal cameras' image resolution is not very high and plants are small at the V1 (2) stage [131]. Thermal cameras are mainly used to dynamically monitor crop growth, diseases, stress, temperature, and so on. The spectral region of a thermal camera [116], [131], [132], [271] enables the estimation of canopy temperature, leaf wilting, water content, lodging, leaf rolling, and water use efficiency.

There is a good consistency between the dynamic monitoring stage (3–8) and key dynamic monitoring traits of photosynthesis sensors and the dynamic monitoring stage (3–8) and key dynamic monitoring traits of fluorescence sensors. However, a photosynthesis sensor is specifically used to assess the photosynthetic status [159], and a fluorescence sensor is employed for evaluating the chlorophyll fluorescence [152]. The dynamic monitoring stage of a stereo camera is from V3 (3) to R1 (7). Due to their advantage of providing 3D structure images [130], stereo cameras can be used to monitor crop growth and structure, and their corresponding key dynamic monitoring traits involve height, leaf angle, lodging, leaf rolling, biomass, canopy coverage and structure, and yield. The dynamic monitoring stage (2–7) of lidar is longer than that of a stereo camera because lidar provides refined point cloud data that are used to describe small plants without the effect of illumination conditions [112]. The key dynamic monitoring traits of lidar agree with those of stereo cameras.

Figure 2 conveys a simple example of how to use different optical sensors to estimate key crop phenotyping traits across the growth stages in a maize breeding program; additional significant crop phenotyping traits are included in Table 2. Furthermore, different sensors can be comprehensively combined to improve the estimation accuracy of some vital phenotyping traits (such as lodging [134], yield [235], biomass [107], and so forth). More details are presented in the "Application of Ground and Aerial Phenotyping Platforms" section. In addition, a SPAD-502 chlorophyll meter [233] (a leaf-level sensor), an ultrasonic sensor [111], and the GreenSeeker system (a near-canopy sensor) [66] may be used, relatively inexpensively, to obtain the chlorophyll content, nitrogen content, and plant height, respectively.

Currently, it is a challenge to sufficiently improve the estimation accuracy of crop phenotyping traits with different optical sensors to satisfy the needs of crop



**FIGURE 2.** Examples of potential applications of field crop phenotyping with different sensors at the key growth stages in maize breeding programs. VE: emergence 1; V1: first leaf collar, 2; V3: third leaf collar, 3; V6: sixth leaf collar, 4; V9: ninth leaf collar, 5; VT: tasseling, 6; R1: silking, 7; R6: maturity, 8.

breeding programs [7]. There is a need for new ways to combine image information from different optical sensors (sensor fusion) to increase the estimation accuracy of crop phenotyping traits in the future. Image-processing scientists will increasingly focus in this direction to enhance the functionality of sensors in crop phenotyping studies [115].

## DEVELOPMENT OF GROUND AND AERIAL PHENOTYPING PLATFORMS AND APPLICATIONS

In this article, field phenotyping platforms are divided into two types based on ground and aerial levels. Currently, ground and aerial phenotyping platforms can carry different sensors to obtain crop phenotyping traits in the field. The ground and aerial phenotyping platforms are shown

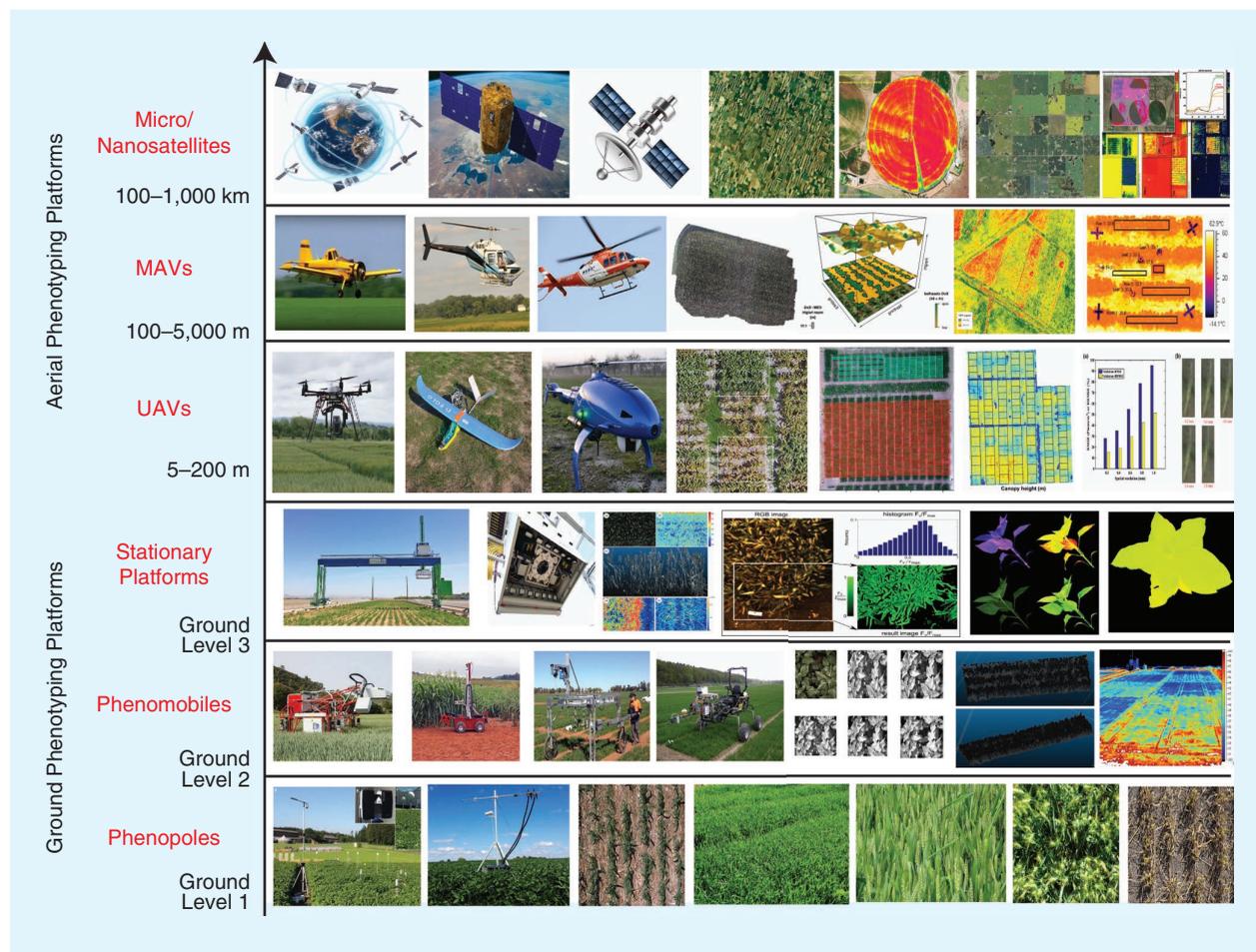
in detail in the next section and the “Aerial Phenotyping Platforms” section, respectively.

### GROUND PHENOTYPING PLATFORMS

Ground phenotyping platforms may be classified as phenopoles, phenomobiles, and stationary platforms (Figure 3). With phenopoles, poles (typically made from aluminum, steel, or plastic fibers) are used to directly mount sensors to obtain crop phenotyping trait data. Phenopoles include fixed and mobile versions. Fixed phenopoles are very similar to a field weather station: a low-cost RGB camera sensor is installed on a pole to obtain images that are updated to a server, for example, every 30 or 60 min based on sunlight illumination conditions [5]. The PhenoCam is an example of a fixed phenopole. The advantage of fixed phenopoles relates to the ability to obtain RGB images to monitor the dynamic changes to crops’ canopy coverage, FAPAR, and GAI. The PhenoCam Network [275] uses digital cameras to track the seasonal dynamics of vegetation across a range of ecosystem types. Phenopoles are limited in that they focus only on a subsection of an experimental plot, and the price of a phenotyping study is expensive when cameras are installed across a large experimental site [59].

Mobile phenopoles are manually carried. Their camera is commonly controlled by a cellphone through Wi-Fi data transformation. Mobile phenopoles offer the advantage of obtaining higher-resolution images because they are near the ground level (1–3 m). Their limitation is that they require a lot of manpower to gather images for all the plots at a phenotyping experiment site. Fixed and mobile phenopoles are mainly used to obtain information pertaining to the plant density, canopy cover, GAI, flowering stage, VI, phenology, ear density, and so forth.

Phenomobiles include automatic, human, and tractor-propelled platforms. The automatic phenomobile commonly includes sensors, an integrated GPS system, a navigation control system, a data acquisition system, and a power supply [276]. The sensors and the integrated GPS system provide compatibility and efficient data acquisition, and they ensure that sensor outputs are recorded with navigation coordinates. The navigation control system is used to design a reasonable walking route through the field, while the data acquisition system maintains the consistency of the flashlight and monitors the obtained information to protect the data quality and minimize the effects of the background illumination. The power supply system



**FIGURE 3.** Ground phenotyping platforms (including stationary platforms, phenomobiles, and phenopoles) and aerial phenotyping platforms [including UAVs, manned aerial vehicles (MAVs), and micro/nanosatellites] [39], [40], [111], [200], [259], [260], [276], [280].

guarantees continuous electricity through a regulator. The cooperation between each system can safeguard the normal operation of the automatic phenomobile.

Currently, the automatic phenomobile can autonomously obtain RGB, multi/hyperspectral, and thermal images and point clouds using different optical sensors for each plot in a field. These images and point clouds can be employed to estimate the canopy cover, GAI, canopy height and temperature, chlorophyll and nitrogen content, water content, yield, grain quality, relative values of the in-season biomass, and so forth. The dynamic changes of crop phenotyping traits can be monitored through the automatic phenomobile without much human intervention. The advantage of the automatic phenomobile lies in the ability to simultaneously collect different types of images; however, such platforms are expensive. Current automatic phenomobile examples include Ladybird [277] at the University of Sydney, Australia, and Phenomobile [276] at the Institut National de la Recherche Agronomique, Paris, France.

Human phenomobiles are moved by manpower through a field. Examples include Phenomobile Lite [200], which contains data acquisition systems controlled by a person with a laptop. The system can incorporate RGB, multi/hyperspectral, and thermal sensors to obtain the related crop phenotyping traits. The price of human phenomobiles is low, but the data acquisition quality and efficiency of the sensors cannot be guaranteed.

Tractor phenomobiles consist of integrated sensor and GPS systems, a data acquisition system, and a power supply [278]. The basic integrated-sensor-and-GPS system combines non-image-based sensors (such as GreenSeeker and CropCircle, which are hyperspectral), the the outputs of which are recorded with GPS coordinates; similarly, the data acquisition system maintains the brightness of the flashlight and the consistency of the sensor data to ensure the quality the gathered information. The tractor provides the power supply. The PhenoTrac4 could be used to obtain VIs, nitrogen uptake, biomass, and water status. Its price is lower than other phenomobiles because its sensors are non-image-based [278]. Phenomobiles are relatively flexible in terms of sensors.

Finally, the sensors of stationary platforms include RGB cameras, multi/hyperspectral cameras, and thermal cameras and laser sensors. Stationary platforms are simultaneous and fully automated, fixed-site phenotyping equipment [279]. They can carry sensors for the noninvasive estimation of crop growth, physiology, morphology, and health. Their advantage is that they can simultaneously monitor different crop traits, although only in a defined area of, for example, roughly 1 hectare, when they are mounted on trackways [279].

### **AERIAL PHENOTYPING PLATFORMS**

Aerial phenotyping platforms may be classified as MAVs, UAVs, and micro/nanosatellites (Figure 3). Recent advances in technology have promoted the development of UAVs,

which have been transformed into crop phenotyping platforms that provide high-spatial-resolution images for crop phenotyping trait estimation in the field [259], [260]. They include fixed-wing and multirotor configurations. Fixed-wing UAVs have the advantages of fast flight, high flight efficiency, long endurance time, large payload capacity, and a high flight altitude. However, they have certain requirements for taking off and landing, they cannot hover, and they will cause blurred images because of their high-speed shooting [281]. Multirotor UAVs have a simple structure, the ability to hover, and modest requirements for taking off and landing, but they possess a slow flight speed, short endurance time, a low flight altitude, and small payload volume [281].

Sensor images from UAVs are processed using Agisoft Photoscan Professional (Version 1.2.2, Agisoft, Saint Petersburg, Russia). The overlap between images should be larger than 60% for the software to compute the position of the camera corresponding to every acquired image [39]. The software can automatically recognize targets applied as ground control points (GCPs) for the absolute geopositioning of the images, and it can precisely locate the objects' center on the images [39]. The specific processing is conducted as follows:

- 1) check the camera calibration and optimizing the parameters
- 2) align the photos
- 3) build dense point cloud
- 4) construct the mesh
- 5) create the texture
- 6) build digital elevation models (DEMs)
- 7) develop the orthomosaic
- 8) export the orthomosaic and the DEMs.

The lidar point cloud from UAVs can be processed by CloudCompare or by writing code as needed [112], [199]. Currently, UAVs can carry different sensors (RGB cameras, multi/hyperspectral cameras, thermal cameras, and lidar) to estimate different crop traits at varying spatial scales. Images from UAVs can be employed to estimate grain yield [99], canopy temperature [266] and NDVI [259], [282], plant height [108], [282], [283], biomass [162], GAI [109], lodging [134], plant density [39], fluorescence [266], and nitrogen status [36], mostly through proxies for plant traits. The advantage of UAVs concerns the relatively high-resolution images that are obtainable in a relatively short time; however, it is difficult to cover very large areas due to the vehicles' limited range and speed. The onboard sensors of MAVs are comparable with those of UAVs. MAVs can cover larger regions in a relatively short time, albeit with a lower image resolution, and they have a greater payload capability. Images from MAVs are mainly used to estimate the GAI, biomass, chlorophyll content, nitrogen content, plant height, and biomass [8].

Finally, micro/nanosatellites can provide data in several spectral bands (which are sensitive to the crop structure, leaf pigment, and water content), and they enable meter-level-resolution images for estimating crop phenotyping traits at

a very large scale [24], [79], [87], [88], [144], [193], [236], [239]. Micro/nanosatellites include optical and synthetic aperture radar (SAR) satellites. Optical satellites include the *Worldview* series, *QuickBird*, *Ikonos*, *Geoeye-1*, *Satellite for Earth Observation* series, *RapidEye*, *Landsat* series, *Gaofen* series, and so on. SAR satellites include the *Environmental Satellite (ENVISAT)*, *Radar Satellite (RADARSAT)*, *TerraSAR-X*, *Sentinel-1*, *Advanced Land Observation Satellite-Phased Array Type L-band Synthetic Aperture Radar*, and so forth. Optical satellite images are processed (including radiometric calibration, atmospheric correction, and geometric correction) using ENVI 4.7 software [284].

The fast line-of-sight atmospheric analysis of the spectral-hypercubes module is employed to retrieve the land surface reflectance. The geometric correction of each image is based on the measured GPS GCPs. SAR satellite images are processed using polarimetric SAR (PolSAR) Pro v5 [24], including the following steps:

- 1) The DN is converted into the backscattering coefficients using look-up tables in the product file (radiometric calibration) [285].
- 2) A  $5 \times 5$  boxcar filter is applied for screening and for the multilook to suppress speckle [286].
- 3) The filtered SAR images are applied to obtain a scattering matrix (S2) and then converted to a symmetrized  $3 \times 3$  covariance matrix (C3) [287].
- 4) The SAR images are orthorectified using DEM simulation and registration [288].
- 5) Finally, the SAR images are rectified using GPS GCPs.

Optical satellites use the relative reflectance at certain spectral wavelengths to estimate crop phenotyping traits based on VIs, machine learning methods, weather data, environmental factors, and crop varieties [79], [193], [236], [289]. However, optical satellites have saturation problems for VIs and a reduction in their estimation accuracy at high plant densities [24]. Compared with optical satellites, SAR satellites have some advantages for estimating crop phenotyping traits at high plant densities because SAR sensors use longer wavelengths, can penetrate crop canopies, and are not influenced by the presence of clouds and haze [88], [242]. For example, the backscatter and polarimetric decompositions of PolSAR are very sensitive to crop morphological structure changes [87]. SAR satellites have also been used to monitor crop phenology, yield, lodging, and so on through machine learning methods and polarimetric parameters [87], [143], [242]. More detail about the application of aerial phenotyping platforms is provided in the "Application of Ground and Aerial Phenotyping Platforms" section.

With the rapid development of micro/nanosatellite technology, higher-resolution images (<1 m) may be expected in the future. The advantages of micro/nanosatellites include international standard protocols for image processing at a relatively low cost. However, the image quality of micro/nanosatellites may be influenced by weather conditions, and the current resolution of micro/nanosatellites limits

their application in crop trial monitoring [59]. A future possible application of images from micro/nanosatellites could lie in evaluating and verifying variety releases across wide geographic areas.

In summary, higher-resolution images are obtained by ground phenotyping platforms that have a relatively low efficiency in terms of coverage. Compared with ground phenotyping platforms, aerial phenotyping platforms can provide images with relatively high efficiency and cover larger areas. It is currently challenging to manage and process the images from ground phenotyping platforms; additionally, there are no international standards for doing so. Although image analysis protocols are available for UAVs and MAVs, large differences exist among laboratories. Scientists need to coordinate to develop internationally uniform protocols for image management and processing for ground and aerial phenotyping platforms, similar to the standard conventions for micro/nanosatellites.

### APPLICATION OF GROUND AND AERIAL PHENOTYPING PLATFORMS

This section introduces applications of ground and aerial phenotyping platforms for estimating crop phenotyping traits under different environmental conditions. Applications include early season crop mapping, crop growth condition monitoring (nitrogen stress, water stress, disease, phenological parameters, lodging, and others), and crop yield estimation.

#### EARLY SEASON CROP MAPPING

Early and timely knowledge of crop types and conditions is extremely valuable information about regional production, yield estimation, and food security [290], [291]. Passive optical satellite sensors have been widely used for early season crop mapping because they improve the spectral, spatial, and temporal resolution of images and increase the availability of satellite data [292], [293]. In multitemporal remote sensing data, different classification algorithms have been used to process and analyze the time series of VIs to characterize growth conditions and then classify crop types during the early season [294]–[299]. Until now, several remote sensing classification algorithms have been successfully applied, such as support vector machines, random forest (RF), decision trees, and neural networks [300], [301]. Skakun et al. [302] used moderate-resolution imaging spectroradiometer (MODIS) NDVI data, a Gaussian mixture model, and growing-degree-days information to detect early season winter crops in large regions; the results showed a good consistency between official statistics and estimates ( $R^2 = 0.85$ ).

In addition, SAR sensors can be used to classify crop types because they are not affected by clouds and atmospheric conditions [24]. Some scientists have investigated the potential of SAR backscatter data for crop mapping using images obtained by *Sentinel-1*, *RADARSAT-1* and 2, *ENVISAT* advanced SAR, and phased-array-type L-band SAR [303]–[308]. In recent years, some studies have carried out

early season crop (wheat, cotton, spring maize, sugarcane, and rice) type classification based on the combination of optical satellite and SAR data [302], [307]–[309]. Hao et al. [307] used the improved artificial immune network and *Sentinel* data to carry out early season crop mapping in Hengshui, China, and the result indicated that the overall accuracy for summer crops and winter wheat was 98.55 and 99%, respectively. Jiang et al. [308] combined machine learning algorithms and *Sentinel-1A/2* time series data to map sugarcane during the early season in Zhanjiang City, China, and the outcome showed that the value of the kappa coefficient was 0.902 and that the estimation accuracy of the sugarcane mapping area was approximately 86.3%.

Compared with satellite platforms, UAVs can provide ultrahigh spatial resolution for classifying crops during the early season. UAVs equipped with RGB and multispectral cameras have been employed to classify weeds and crops [41], [311]. Zheng et al. [311] used RGB, NIR-G-B, and multispectral images from UAVs to detect rice plants during the early season, and the results indicated that a spectral-features decision tree had a high classification accuracy during the early growth stages. With the improvement of the spatial and temporal resolution of images from satellites and UAVs, deep-learning algorithms, e.g., convolutional neural networks (CNNs) [312] and recurrent neural networks [313], will ideally be employed to map early season crop types, with their advantages for image processing and analysis.

## CROP GROWTH CONDITION MONITORING

### NITROGEN STRESS

Nitrogen deficiency in crops results in a decreased chlorophyll content, a slower growth rate, and less photosynthesis as well as more sensitivity to diseases and pests [314]. These changes in the chlorophyll content, growth rate, photosynthesis, and disease and pest occurrence can be monitored using near-infrared and visible spectral reflectance [8], [29], [34]–[36]. These factors have been estimated using UAVs to monitor different nitrogen levels. The results demonstrated that multispectral and hyperspectral images from UAVs can estimate the biomass, nitrogen content, and grain yield with multiple nitrogen treatments [34]–[36]. In addition, a study by Camino et al. [35] showed that solar-induced chlorophyll fluorescence derived from hyperspectral images is a good indicator of photosynthesis under multiple nitrogen treatments in crop phenotyping experiments. Similarly, multispectral and hyperspectral sensors are integrated with phenomobiles to estimate the nitrogen content under different nitrogen treatments [67], [278]. Additionally, fluorescence sensors combined with UAVs and phenomobiles have a great potential for the early monitoring of crop traits under nitrogen stress in the field. Developing specific, lightweight spectral fluorescence sensors in combination with UAVs and phenomobiles for the early detection of nitrogen stress is a great opportunity.

### WATER STRESS

Stomata are typically closed under water-stressed conditions, which reduces crop growth and photosynthesis and may aggravate crop heat stress because of decreased transpirational cooling [8]. The canopy temperature is a good indicator of the response of crops to water stress [315], and scientists have reported that under water stress the measurement can be used to identify drought-adapted cultivars [114], [123], [136], [268], [316]–[318]. Thermal images need to be normalized to the ambient temperature and relative humidity to control the effect of environmental conditions on measurements. In a study, Jones et al. [268] indicated that the relative value among genotypes was more important than the absolute value for crop phenotyping evaluations. In addition, the background temperature (soil and dead leaves) of thermal images ideally should be eliminated from the signal of the green leaves using related image-segmentation algorithms [121].

Using thermal cameras with UAVs and phenomobile platforms enables the collection of more crop temperature images in less time compared to ground-based approaches [259], and it may assist in selecting water-stress-resistant genotypes. The limitation of the crop growth status (such as the biomass) owing to water stress could be estimated using near-infrared and visible optical sensors [319]. Therefore, hyperspectral and multispectral cameras can be applied in combination with thermal cameras to select water-stress-resistant genotypes [66], [123]. Images from thermal, multispectral, and hyperspectral cameras collected through the whole crop growing season can offer important quantitative and qualitative data sets that could be correlated with other ground-truthing data sets. These methods will promote the uptake of high-throughput, nondestructive sensors for estimating crop phenotyping traits in the field and may finally take the place of traditional data collection methods.

### DISEASES

Crop yield losses are persistent problems in agriculture due to the widespread occurrence of pathogens, such as fungi, viruses, nematodes, and bacteria. Advanced disease-monitoring technologies are required to minimize crop yield losses [98]. Remote sensing technologies have been applied to estimate diseases [320] and monitor new outbreaks worldwide [265]. However, the application of UAV-based optical sensors for detecting crop diseases and the tolerance of different crop varieties to maladies is less well developed [8], [99]. Nebiker et al. [99] used a threshold of NDVI values from multispectral images to identify blight infestation in potatoes. Crop diseases are mainly detected using near-ground optical sensors (such as hyperspectral, RGB, and thermal cameras) [100], [103], [104]. Recent research on detecting Huanglongbing (HLB) disease-infected citrus trees demonstrated that a good classification accuracy of HLB disease was obtained using the combination of visible, near infrared, and thermal spectral bands [100].

The results obtained by Jansen et al. [321] showed that a *Cercospora* leaf-spot index obtained from spectral indices was correlated with the *Cercospora* disease severity in sugar beet varieties. Furthermore, the classification accuracy of tomato diseases was better using a superresolution method (which produced a spatial resolution-enhanced image using low-resolution images) compared to using the conventional image-scaling methods with RGB images [103]. Behmann et al. [104] employed a miniaturized, handheld hyperspectral camera to identify powdery mildew at the canopy scale in barley. In short, these results indicated that the occurrence of disease and the resistance of crop varieties to infection can be effectively estimated using near-ground and UAV-based sensors. The fluorescence sensor was more sensitive to crop diseases than other instruments. It has the potential to be used in combination with hyperspectral, multispectral, RGB, and thermal cameras for the early detection of crop disease severity. Such sensors have a great potential to be harnessed to estimate the crop disease severity in future crop breeding programs. These near-ground and UAV-based sensors will detect the resistance of crop varieties to disease more effectively when combined with newly developed image classification methods.

#### PHENOLOGICAL PARAMETERS

Crop phenology provides important information for simulating and monitoring crop growth and development [322], estimating crop yields [323], optimizing production management and decision-making [324], and analyzing crops' response to climate change [289]. Remote sensing-based observations with short revisit times and large spatial coverage have been employed to successfully retrieve crop phenological parameters using VI time-series data at regional to global scales [76], [77]. The NDVI and enhanced VI have been commonly employed to estimate crop phenology. The most-studied phenological parameters contain the start of season (SOS), peak of season, and end of season (EOS) [78]. Currently, various methods have been used to extract these phenological parameters using VI time series data, including dynamic threshold methods [79], fixed threshold methods [79], maximum-slope methods [80], function-fitting methods [81], moving average methods [82], and the valley-detection method [83].

The dynamic threshold method is the most commonly employed for estimating the crop SOS and EOS because it needs fewer parameters, has an easier application, and produces high-accuracy results. Sakamoto [84] developed the shaped model-fitting method with MODIS wide-dynamic-range VI time-series data to evaluate the timing of phenological parameters for 36 crop development stages of major U.S. food products. However, Verger et al. [85] found that leaf area index (LAI) is more robust and sensitive than VIs in considering vegetation from different satellite data. Furthermore, Luo et al. [86] used global land surface satellite LAI products and optimal filter-based phenology detection methods to obtain the crop SOS and EOS and produced a

1-km spatial resolution phenological survey of three staple crops in China from 2000 to 2015.

PolSAR is very sensitive to crop morphological structure changes [87]. *RADARSAT-2* and *TerraSAR-X* provide opportunities to explore the potential of these data for crop SOS and EOS estimation. Lopez-Sanchez et al. [88] used dual-polarized *TerraSAR-X* data and simple decision tree algorithms to evaluate rice phenological parameters. Furthermore, they harnessed the three polarimetric parameters (entropy, anisotropy, and alpha angle) from polarimetric *RADARSAT-2* data and hierarchical decision trees to estimate rice phenological parameters [89]. Wang et al. [87] applied the multitemporal *RADARSAT-2* dataset, SAR polarimetric decompositions, and an RF algorithm to monitor canola, maize, soybean, and wheat phenology based on crop phenological parameters. With the fast development of UAVs and sensors, near-real-time crop phenology retrieval is feasible. Yang et al. [90] proposed a near-real-time deep-learning method for monitoring rice phenological parameters and estimated rice phenology from UAV RGB images with an accuracy rate of 83.9%. In the future, the PheNoCams network will be combined with satellite images to better estimate different crops' phenological parameters. UAV images and advanced machine learning algorithms will improve crop phenological parameter retrieval in near real time and thus enable optimized crop management.

#### LODGING

Crop lodging is the permanent deviation of crop plants from the upright position [325]. Crop lodging not only influences harvest operations but also results in yield losses and can reduce quality [326]. Therefore, accurate and rapid evaluation of crop lodging is important for research and screening, optimizing field crop management, obtaining better production, and estimating crop yield losses. The reflectance and backscatter at different wavelengths are affected by changes in the crop structure [327], biochemical properties [328], and morphology [329] in the lodging condition.

Many researchers have used crop phenotyping platforms to estimate crop lodging [134]–[143]. An early study by Fitch et al. [330] applied the linear polarization of backscatter from wheat to evaluate its potential in detecting wheat lodging using ground-based platforms. Ogden et al. [135] used motor-driven cameras to analyze textural information in images to estimate the extent of rice lodging. Liu et al. [331] demonstrated that optical hyperspectral data can be used to detect rice lodging. Recent advances in the development of UAVs and sensors have been used to detect crop lodging areas and evaluate lodging severity [134], [136]–[138]. Chapman et al. [136] reported that wheat lodging areas are successfully detected using thermal images from UAV-mounted thermal cameras. The combination of spectral and texture features and the digital surface model further increased the estimation accuracy of rice lodging classification using single-feature probability

algorithms. In a recent study, Liu et al. [134] employed the fusion of RGB and thermal image information to improve the estimation accuracy of lodging classification in different rice species.

Satellite-based platforms can monitor crop lodging due to the availability of their images. Yang et al. [143] presented the potential of radar-based satellite images to detect wheat lodging at the farm scale, due to the sensitivity of SAR to crop structural changes. Some studies have employed *RA-DARSAT-2* quad-polarimetric images to estimate lodging in wheat and canola [142], [144]. In the future, the different phenotyping platforms and new machine learning algorithms will be better integrated to address the reasons for crop lodging and provide improved estimates of lodging at regional to global scales.

#### OTHER APPLICATIONS

The different sensors from ground and aerial phenotyping platforms can be used to estimate many other important crop morphology phenotyping traits (GAI, canopy coverage, plant vigor, canopy height, leaf rolling, leaf angle, FAPAR, leaf staygreen/senescence, phenology, crop dynamics monitoring, biomass, and canopy structure), resource use phenotyping traits (nitrogen use efficiency, light use efficiency, and water use efficiency), yield component phenotyping traits (plant density, ear density, grain number and size, and grain quality), physiological phenotyping traits (chlorophyll content, water content, chlorophyll fluorescence, photosynthetic status, and nitrogen nutrition index), and stress phenotyping traits (plant diseases and pests and weed infestation) in crop breeding programs. In addition, the effects of soil components and compaction, field management practices, and soil spatial variability on crop phenotyping traits should be measured in parallel to include the effect of these environmental factors on the estimation accuracy of crop phenotyping traits.

#### CROP YIELD ESTIMATION

Crop yield is widely used to measure cropland productivity. Estimating how much crop yield is obtained prior to harvesting is essential for field crop management, food trade balance, food security evaluation, and policy-making in the world [241], [332]. Furthermore, the rapid and efficient estimation of crop yield with high accuracy will allow identification of high-yielding genotypes in large germplasm panels [333]. Many studies have been successfully employed to estimate crop yield using different crop phenotyping platforms [170], [234]–[241], such as ground-based, UAV, and satellite-based platforms. For ground-based platforms, Jin et al. [170] assimilated the biomass and canopy cover derived from field hyperspectral VIs into the AquaCrop model based on the particle swarm optimization algorithm to improve the estimation accuracy of the maize yield with  $R^2$  and root-mean-square error (RMSE) values of 0.78 and 1.44 tons/hectare, respectively. Li et al. [240] combined field spectral VIs and meteorological data through hierarchical

linear modeling (HLM) to estimate wheat yield, and their results showed that the approximated power of yield estimation derived through HLM was higher than that obtained using ordinary least-squares regression, with  $R^2$  and RMSE values of 0.75 and 1.10 tons/hectare, respectively.

For UAV platforms, Maimaitijiang et al. [235] applied multimodal data fusion and deep-learning methods to estimate the soybean yield from UAV RGB, multispectral, and thermal images, and the results demonstrated that the highest estimation accuracy was obtained by a deep neural network with an  $R^2$  and relative RMSE of 0.720 and 15.9%, respectively. Another study by Yang et al. [238] used deep CNNs to estimate rice yield at the ripening stage from UAV, RGB, and multispectral imaging data and demonstrated that CNNs perform better than VI-based models for rice yield estimation ( $R^2 = 0.59$  and RMSE = 0.66 tons/hectare).

Zhang et al. [237] extracted excess green color features from UAV RGB images and built an estimation model for maize yield, and the corresponding mean absolute percentage error ranged from 6.2 to 15.1%. For satellite-based platforms, Sakamoto [234] incorporated MODIS VIs and environmental variables to estimate the yields for maize and soybeans using a RF regression algorithm, and their results indicated that the RF method obtained a better yield estimation accuracy (maize RMSE = 0.539 ton/hectare and soybean RMSE = 0.206 ton/hectare) at the state level. Similarly, Schwalbert et al. [236] integrated long short-term memory (LSTM) neural networks, weather data, and machine learning for improving crop yield prediction in southern Brazil and showed that LSTM neural networks performed better than other algorithms. In addition, Dong et al. [145] estimated wheat yield using a light use efficiency model and wheat variety data, and the results indicated that the proposed method enhanced the model simulation performance and achieved an 82% estimation accuracy of the interannual yield variation.

In short, the preceding studies demonstrated that the combination of different crop phenotyping platform images, meteorological data, crop models, variety data, environmental variables, and machine learning methods could be used to improve current estimation accuracy of crop yield. In particular, deep-learning algorithms, such as deep CNNs and LSTM neural networks, can significantly improve the estimation accuracy of crop yield. It can be assumed that researchers will have to pay more attention to the integration of multisource data with deep-learning methods to improve crop yield estimation in the future.

#### FUTURE PERSPECTIVES

While high-throughput crop phenotyping technologies have made some progress, further improvements are needed due to the requirements for more accurate estimations of most crop phenotyping traits. Advanced sensors, together with cutting-edge ground and aerial phenotyping platforms, have led to a major requirement for advances in

image processing. Recently, new machine learning algorithms have been used to identify useful crop traits using image information. For example, deep-learning algorithms have shown advantages for trait detection and segmentation [46], [103]. Crop phenotyping sensors are commonly influenced by field environmental conditions; therefore, developments of more stable and refined crop phenotyping sensors are necessary for field crop phenotyping research.

Currently, many crop phenotyping sensors and platforms are too expensive for numerous crop breeding programs; however, the rapid development of miniaturized and mobile technologies has provided some affordable and powerful sensors for crop phenotyping with high-resolution images. As sensors have become lighter and smaller, they have been integrated with the different ground and aerial phenotyping platforms, facilitating crop phenotyping research [59], [115], [281]. RGB, multispectral, hyperspectral, and thermal cameras; photosynthesis and fluorescence sensors; stereo cameras; and lidar have been used to evaluate crop phenotyping traits. How to combine the outputs of these sensors to increase the estimation accuracy of crop phenotyping traits is still a challenge for crop phenotyping traits [7], [59]. Micro/nanosatellites with relatively high-resolution images also provide a good resource for crop phenotyping traits in large-scale validation, due, in part, to the international standard protocols for image processing.

The combination of available sensors and platforms provides ground-based crop phenotyping platform systems with the ability to simultaneously obtain, process, and store data in an affordable and efficient manner [59]. However, compatibility issues between software and hardware still exist. In addition, the processing of images from ground-based crop phenotyping platform systems does not have internationally uniform standards, so the use and sharing of image data sets will be limited [14]. Multidisciplinary teams are needed to build efficient integrated crop phenotyping management systems. Such systems need user-friendly data analysis and management interfaces that are integrated with data selection and processing and decision-making functions.

Furthermore, corresponding field soil properties and weather information should be included with image data set analyses and management systems to increase the estimation accuracy and stability of crop phenotyping traits [59]. Finally and importantly, the rapid development of high-throughput crop phenotyping technology will improve future study of precision agriculture. How to effectively combine these crop phenotyping traits and agronomic indicators to enhance crop field management at different growth stages, and thus implement precision agriculture, will be a long-term goal [243].

In summary, crop phenotyping platforms should be adopted by crop breeders as a powerful tool for genetic improvement. It is against this background that affordable and efficient crop phenotyping systems will become the routine choice for crop breeding programs. A crop functional

structure model has been used to simulate crop 3D growth structural changes during the whole crop growth season. This can be integrated with crop phenotyping platforms to improve the estimation accuracy of crop phenotypic traits using data assimilation methods, and it can be used to design crop ideotypes for the future.

However, in many cases, this kind of information would be more detailed than most breeders need. Their main requirements would be reliable estimates of the heading date and plant height, spectral indices that detect biotic and abiotic stress, and approximations of the yield and yield components, including spike density and biomass [310]. These traits can generally be remotely projected with sufficient heritability and combined into selection indices to help choose material that is either for advancement to the next breeding generation or for inclusion in multilocation trials. In addition, QTL and GWAS approaches are not always successful for analyzing relationships between genetics and phenotype and then selecting key gene loci [5]. Therefore, the QTL and GWAS approaches will need to be improved by combing other statistical methods (such as meta-analysis, meta-GWAS methods, and so forth) to identify more stable and effective key gene loci in the future.

## CONCLUSION

High-throughput crop phenotyping trait selection is important for improving crop yield and stress resistance under different biotic/abiotic environmental conditions in plant breeding programs. Currently, the ability to provide high-throughput identification of crop phenotyping traits limits our capacity to determine quantitative genetic traits linked with yields, crop growth statuses, and adaptation to environmental stress. The rapid development of sensors, ground and aerial phenotyping platforms, and image-processing technologies is providing effective tools for high-throughput crop phenotyping traits in field.

This article first introduced the origin and definition of crop phenotyping. Second, it described the development of current sensors (RGB, multispectral, hyperspectral, and thermal cameras; photosynthesis and fluorescence sensors; stereo cameras; and lidar) for crop phenotyping traits in the field, including an analysis of the advantages and limitations of different sensors and their levels of potential application. Third, it highlighted the development of ground (stationary platforms, phenomobiles, and phenopoles) and aerial (UAVs, MAVs, and micro/nanosatellites) phenotyping platforms and their applications for crop phenotyping under nitrogen, water, and disease stresses in the field. Finally, new opportunities and directions of crop phenotyping technological developments for the future were presented. In the future, high-throughput crop phenotyping technology will increase the estimation accuracy of measuring standard crop traits and further accelerate the efficiency of new trait identification in crop breeding programs, based on newly developed sensors, phenotyping platforms, and image processing and data management methods.

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