



Phenomics-Assisted Breeding: An Emerging Way for Stress Management

18

Monu Kumar, Anima Mahato, Santosh Kumar,
and Vinod Kumar Mishra

Abstract

The challenges posed by several known and unknown biotic and abiotic stresses arising due to increasing population, global warming, and other potential climatic factors have severely affected the growth and yield of many agriculturally important crops. Abiotic stresses such as drought, flood, salinity, high temperature, etc. not only influence the physiology of plants but also accompany occurrence and spread of various pathogens, insects and weeds, which may sometimes lead to a famine-like situation. In this context, understanding the crops' response towards different stress conditions and the underlying stress resistance mechanisms has become a challenging task for plant breeder in breeding stress-resistant or climate resilient varieties. With the advent of molecular technologies and functional genomics over past decade, whole genome sequence of many crops is now available and has simplified the process of cloning and characterization of key genes governing important agronomic traits along with the physiological pathways underlying them. But to appraise the full potential of a genotype under stress condition, it is important to evaluate the response in terms of phenotypic behavior and the elements coordinating such responses. So, this post-genomic era has given rise to the need of advanced phenotyping tools for efficient utilization of the vast amount of genomic data in climate resilient breeding. The

M. Kumar (✉)

Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, BHU,
Varanasi, Uttar Pradesh, India

A. Mahato

ICAR-Indian Institute of Seed Science, Mau, Uttar Pradesh, India

S. Kumar

Regional Maize Research & Seed Production Center (ICAR-IIMR), Begusarai, Bihar, India

V. K. Mishra

Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, BHU,
Varanasi, Uttar Pradesh, India

© Springer Nature Singapore Pte Ltd. 2020

A. Rakshit et al. (eds.), *New Frontiers in Stress Management for Durable
Agriculture*, https://doi.org/10.1007/978-981-15-1322-0_18

295

advanced phenotyping approaches use different imaging techniques that record interaction between plant and light which are transmitted, reflected or absorbed and provide measurements related to quantitative phenotypic traits with desired accuracy and precision. The various imaging techniques record the interaction between plants and light like photons, which are transmitted, reflected or absorbed and provide the desired level of accuracy and precision in measurements related to quantitative phenotypic traits. Visible light imaging, infrared- and thermal-based imaging, fluorescence imaging, spectroscopy imaging, and other integrated imaging techniques are currently in use for precise phenotyping of crops under varied environments. The advanced phenomics tools measure plants' response to different abiotic stresses in terms of root architecture, chlorophyll content, canopy temperature deficit and other morphological traits along with disease and insect infestation with a great precision without taking much time and simplifying the germplasm screening process to a great extent. Hence, phenomics is an indispensable tool needed to bridge the gap between phenotyping and genotyping that is highly recommended to cope up the climate resilient varieties.

Keywords

Stress · Climate resilient · Phenotyping · Phenomics · Imaging techniques

18.1 Introduction

As per the latest population prospects, the world population has been predicted to increase by 34% from 6.8 billion today to 9.1 billion in 2050 with continuation in urbanization at an accelerated pace reaching to 70% of the urban population as compared to 49% today. So, it must need to stretch the food production by 70% so as to feed this larger, additional urban and richer population (FAO 2009). It urges an undeviating expansion in crop production, despite adverse environmental conditions and a limited cultivable area to meet the global challenge to sustain the growing human population (Furbank and Tester 2011).

The challenge posed by several known and unknown biotic and abiotic stresses related to worldwide food availability has worsened in the current scenario of adverse and unexpected climatic conditions (Pereira 2016). Global warming and the potential climatic abnormalities associated with it have exposed our crops to variable number of biotic and abiotic stress combinations severely affecting their growth and yield (Mahalingam 2015; Pandey et al. 2017; Ramegowda and Senthil-Kumar 2015). Abiotic stresses such as drought, high and low temperatures, salinity, etc. resulting from global climate change have shown to affect crop production in several ways. The abiotic stresses affect crop productivity by various ways such as altering different physiological processes, spread of pathogens, insects and weeds, sometimes leading to an enhanced risk of famine (McDonald et al. 2009; Ziska et al. 2010; Peters et al. 2014; Long et al. 2015). The impact of concurrent

occurrence of abiotic and biotic stresses is not always additive and depends on the nature of interactions between various stress factors (Atkinson et al. 2013; Prasch and Sonnewald 2013; Choudhary et al. 2016; Ramu et al. 2016). Such environmental extremes cause drastic decline in crop productivity worldwide leading to an annual monetary loss of billions of dollars (Dhankher and Foyer 2018).

The agro-ecological changes occurring due to global warming influence crop physiology to a great extent and pose various threats to naturally occurring crop species (Hatfield and Prueger 2015; Espeland and Kettenring 2018; Raza et al. 2019). In this context, understanding the crops behavior towards a particular stress condition and the underlying stress resistance mechanism has emerged as a challenging task for plant breeder in breeding stress-resistant or climate resilient varieties. There is urgent need to combat these challenges by devising various innovative methodologies in order to obtain high yield and quality with limited resources. With rapid development of functional genomics and other molecular technologies over past decade, whole genome sequence of many crops is now available and dozens of key genes controlling important agronomic traits as well as physiological pathways have been cloned or characterized (Hu et al. 2006; Yang et al. 2013).

To appraise the potential of a genotype under any abiotic stress, it is required to evaluate the response in terms of phenotypic changes and the elements that coordinate a plant's response under stressed situation (Mickelbart et al. 2015). In the post-genomics era also, phenotyping is of higher concern as crop improvement techniques, like QTL analysis, genome-wide association studies (GWAS), fine mapping of genes/QTL, and genomic selection (GS) rely on the precise and accurate measure of phenotypic examination in crop plants. It has been reported the inefficient utilization of crop genetic resources due to the underdevelopment of crop phenomics techniques (Cabrera-Bosquet et al. 2012).

The adequate exploitation of available genetic information has not been possible till date due to outdated phenotypic tools. For removing this bottleneck and taking full benefit of available genomic information, high-throughput phenomics facilities should be employed to get new insight into all aspects of living plants (Poorter et al. 2012; Furbank and Tester 2011; Finkel 2009). So, the phenotypic bottleneck can now be addressed by using novel technologies such as image analysis, spectroscopy, robotics, high-performance computing, etc. for phenotypic data recording. This will definitely facilitate a more dynamic platform for field evaluation of plant performance in a much faster way (Furbank and Tester 2011).

Abiotic stress alters the basic metabolism of plants, resulting in increased production of secondary metabolites and compatible solutes, generation of reactive oxygen species (ROS) and reducing agents (Suzuki et al. 2012). Phenotypic examination of the important parameters specific to the stress using relevant and sophisticated techniques results in a precise and accurate appraisal of the phenotypic response. Various parameters have been used to measure the level of tolerance or susceptibility of a genotype under the particular degree of a stress. For example, root morphology and leaf-traits such as leaf rolling and relative water content, biomass and yield-associated traits are taken under consideration while determining tolerance to salinity and drought (Collins et al. 2008). Considering the foremost

significance of the “phenotype” in crop improvement, precise and accurate phenotyping of integral traits associated with abiotic stress is of great concern (Yang et al. 2013). To witness a dramatic advancement in crop improvement, novel phenotyping tools are required that can record the phenotypic changes precisely and accurately. In the recent past decades, stupendous progress has been made in terms of large-scale genomic technologies such as sequencing, genotyping, and next-generation sequencing with limited progress on the phenomics front. Given this, time demands the development of automated phenotyping platforms which can generate high-throughput and high-resolution precise and accurate data along with the ability to measure nonvisible phenotypic changes (Maphosa et al. 2016). Some headway has been made with establishment of high-throughput phenotyping facilities having robust software system, which encompass visible light imaging, X-ray computed tomography, and hyperspectral imaging. Many plant phenotyping centers have been established in different countries with the potentiality to automatically image thousands of plants and, notably, few QTLs have also been identified in various crops based on these modern amenities (Zhang et al. 2017). These centers are PHENOPSIS system in France (<http://www.international.inra.fr/>), High Resolution Plant Phenotyping Centre (<http://www.plantphenomics.org.au/HRPPC>) In Australia, the Institute of Biological, Environmental and Rural Sciences (IBERS) in the United Kingdom (<http://www.aber.ac.uk/en/ibers/>), and the Leibniz Institute of Plant Genetics and Crop Plant Research in Germany (<http://www.ipk-gatersleben.de>). An integrated approach to plant phenotyping will assist to better understanding of the traits being influenced by the stresses. The modern facilities offered by crop phenomics help plant breeders to adroitly identify crop genotypes with tolerance to various stresses and guide them to develop a resilient crop capable to withstand climate change.

18.2 Phenomics

Phenomics is a field of science, based on using the methods of computer image analysis and integration of biological data which combines biology and informatics to solve the problem of rapid and accurate estimation of the plant phenotype and to analyze phenotypic traits in large-scale genetic and breeding experiments in plants. Advanced phenotyping approaches use image processing with visible to near-infrared light spectrum to yield image datasets of the crop phenotype in a non-harmful manner (Rahaman et al. 2015). The advanced imaging tools for plant biology (Paprocki et al. 2012) include visible light imaging, hyperspectral imaging, infrared imaging, fluorescence imaging and X-ray computed tomography, supported with a robust software system, generate unique and multilevel phenotyping data (Sozzani et al. 2014). The various imaging techniques record the interaction between plants and light like photons, which are transmitted, reflected or absorbed and provide the desired level of accuracy and precision in measurements related to quantitative phenotypic traits. The various imaging devices currently used for high-throughput phenotyping of crop plants are as follows.

18.2.1 Visible Light (300–700 nm) Imaging

For tolerance to abiotic stress-responsive and associated traits, visual survey has been a standard practice. Visible imaging techniques based on two-dimensional (2D) digital images are being used to examine shoot-related traits like shoot biomass (Neilson et al. 2015), shoot tip extension, root and leaf morphology, panicle and seed morphology, etc. (Fahlgren et al. 2015). Visible imaging sensors like silicon sensors (CCD or CMOS arrays) are sensitive to visible spectrum (Li et al. 2014). Three-dimensional (3D) imaging as well as both integrated 2D and 3D imaging technologies are being used to generate more accuracy on complex phenotypes (Rahaman et al. 2015). Shoot dry weight of wheat seedlings for salt stress with a LemnaTec 3D Scanalyzer has been accurately measured by Golzarian et al. (2011). In case of salinity stress, salt accumulation can be correlated by measuring variation between yellow and green areas of the leaf. Image analysis has ability to record stress tolerance traits in a small as well as large populations like mapping populations or mutant populations which facilitate to undertake a genetic study to characterize genes controlling the variations among tolerance-related traits. Phenotyping has been done for various abiotic stresses in many crops using different platforms, like PHENOPSIS (Granier et al. 2006) and WIWAM (<https://www.wiwam.be/>) for drought stress in Arabidopsis; LemnaTec for drought stress in barley (Honsdorf et al. 2014) and maize (Ge et al. 2016) and for salt stress in rice (Hairmansis et al. 2014), barley (Humplik et al. 2015) and wheat (Meng et al. 2017); Plant Screen for chilling tolerance in Arabidopsis (Jansen et al. 2009) and GROWSCREEN for chilling tolerance in pea (Humplik et al. 2015).

18.2.2 Infrared- and Thermal-Based Imaging

Infrared imaging visualizes infrared radiation radiated from the object through Stefan–Boltzmann equation ($R^4/4\epsilon\sigma T^4$) which utilizes internal molecular movements emitting infrared ray for imaging (Kastberger and Stachl 2003). Infrared imaging technology having sensitive range of thermal cameras (3–14 μm) utilizes near-infrared (0.9–1.55 μm) and far-infrared (7.5–13.5 μm) ranges (Li et al. 2014). In addition, NIR imaging combined with visible imaging provides deeper visualization into plant health under various stress conditions by making available well-defined spectral features for leaf water content, pigments, and biochemicals like lignin and cellulose (Yang et al. 2013). It is also used to see the stomatal responses under salinity and drought by observing differences in canopy temperature (Rahaman et al. 2015). Nowadays, various user-friendly thermal cameras with high thermal sensitivity are available to detect plant canopy temperature with higher resolution detectors which provide images of high spatial resolution with precise measurements in large fields during varied climatic conditions on real-time basis (Li et al. 2014). Leaf water status and gas exchange can be evaluated by thermal imaging by observing leaf and canopy temperature. Canopy temperature differences amid the canopy and surrounding air can be taken as measure for drought tolerance.

Thermal infrared imaging also allows characterization of tolerance to stresses like drought and salinity on osmotic tolerance and Na⁺ exclusion basis and recording of relative chlorophyll content and leaf color (Merlot et al. 2002; Jones et al. 2009; Munns et al. 2010).

18.2.3 Fluorescence Imaging

Fluorescence is the emission of light of low wavelength after absorbance of light. Fluorescence imaging blazes light of blue wavelength (<500 nm) on the plants and in response fluorescence light is emitted at 600–750 nm in the red spectrum. The fluorescence differences are photographed and modified into color signals using software to analyze them (Weirman 2010). Chlorophyll fluorescence is generally recorded in phenomics to disclose the effect of various stresses on genes and the plant's ability to cope photosynthesis under these traumas (Weirman 2010). Fluorescence imaging (Rascher et al. 2001; Osmond et al. 2004) can also help to study stomatal movement, phloem loading and unloading, and plant metabolite content under stress. Ultraviolet light produces red to far-red region and blue to green region fluorescence (360–740 nm) which captures fluorescence emission by single excitation wavelengths (Rahaman et al. 2015). Chlorophyll fluorescence (ChlF) imaging has been used to measure growth, morphology, color, and photosynthetic performance in rice (Hairmansis et al. 2014) and *Arabidopsis thaliana* (Awlia et al. 2016) under salt stress.

18.2.4 Spectroscopy Imaging

Spectroscopy imaging is due to the interaction between solar radiation and plants via hyperspectral and multispectral cameras. Hyperspectral imaging dissects images into bands, thus generating electromagnetic spectrum in the images (Yang et al. 2013). Various spectral regions have been identified like (1) NDVI (normalized difference vegetation index) compares red and near-infrared reflectance, (2) CRI (carotenoid reflectance index) determines three wavebands in the yellow region, and (3) PRI (photochemical reflectance index) that correlates functional status of non-photochemical energy conservation (Fiorani et al. 2012). In NIR region, radiation passed from upper leaves to lower leaves is reflected back to upper part leading to resolve leaf and canopy architecture. Further, reflectance gradually decreases with an increase in wavelength and absorption due to leaf water content showing its water status. This spectral reflectance information is utilized to compute vegetation indices and enables the detection of NDVI. The vegetation indices are associated with various traits like pigment content, water status, and active biomass (Penuelas and Filella 1998; Din et al. 2017). A matrix factorization method called SiVM (simplex volume maximization) has been applied in cereal crops by Romer et al. (2012) to figure out hyperspectral data for early drought detection.

18.2.5 Integrated Imaging Techniques

Various technical progression has shifted towards live imaging of plants, e.g., functional imaging and optical 3D structural tomography. Positron emission tomography (PET) and ChlF imaging under functional imaging category evaluate photosynthetic performance by focusing on physiological changes under stress (Baker 2008). PET is a nondestructive technique which uses the positron-emitting radionuclides metabolite compounds labeled with C11, N13, or Fe52 (Kiyomiya et al. 2001). Magnetic resonance imaging (MRI) is an improved technique which creates images by integrating magnetic fields and radio waves which is used to capture root architecture in pots and internal physiological processes (Borisjuk et al. 2012) along with water diffusion and transportation via xylem and phloem in crop plants like tomatoes, tobacco, poplars, and castor beans (Windt et al. 2006). Integrated technique of both MRI and PET offers a novel image to monitor real-time changes in plant function and structure. Jahnke et al. (2009) studied photo-assimilation in sugar beet taproots and shoot-to-root carbon fluxes by coupling PET and MRI using [C11]-labeled CO₂.

Forster resonance energy transfer (FRET) is a further advanced and outstanding noninvasive or nondestructive technology for molecular phenotyping based on genetically encoded, radiometric fluorescent sensors (Jones et al. 2014). Various multiple pathways and dynamic processes of the plants can be identified through a single FRET sensor. FRET has successfully detected calcium and zinc dynamics along with subcellular spatial and temporal resolution in real time in roots during sugar transport (Jones et al. 2014). FRET by its advanced phenotyping ability can address all the basic questions related to plant growth and development. A high-resolution 3D laser scanner, PlantEye was used to phenotype wheat crop under control and salt stress in a controlled environment. It scans plants from overhead, creating a data cloud from which traits such as 3D leaf area, plant height, and leaf are computed by the system (Maphosa et al. 2016).

18.3 Application of Phenomics in Stress Management

18.3.1 Phenomics tool for abiotic Stresses

Abiotic stresses such as drought, salinity, heat, cold, water logging, etc. are major causal factor affecting agricultural productivity, thereby leading to more than 50% of worldwide yield loss of major crops every year (Verma and Singh 2016). Many of these abiotic stresses are interconnected in terms of osmotic stresses and various metabolic changes occurring within the plant. This happens mainly due to alteration in expression pattern of group of genes governing different physiological aspects of plants that finally leads to reduced growth rate and productivity (Kumar 2013). Hence, in-depth study and better understanding of complex responses of plants towards abiotic stresses will require an integrated knowledge of genomic and phenomic facilities.

Global climatic changes have resulted in an increased temperature and atmospheric carbon dioxide level accompanied with in-appropriate rainfall ultimately leading to drought. Severe drought is highly lethal and may cause premature plant death leading to entire crop failure, while intermittent drought conditions are not lethal but may cause unpredicted yield losses by affecting plant growth and development (Kumar 2013). Drought strike areas often accompany other stresses like high temperature, soil salinity, disease and pest infestation, etc., which all together bring about different morphophysiological changes in the crop. Various morphophysiological traits and the corresponding QTLs affecting yield under stress conditions were grouped as constitutive, expressed under both water stress and well-watered conditions or drought-responsive which expressed only under severe drought conditions (Blum 2006; Tuberosa 2012). The morphophysiological traits that should be targeted for developing drought tolerant varieties must include root architecture, early vigor, flowering time, stomatal conductance, canopy temperature depression (CTD), ABA concentration, osmotic adjustment, chlorophyll concentration, stay-green, delayed leaf senescence, etc.

Plants exhibit more carbohydrate allocation to the root system when grown in a nutritional or water deficit environment producing a root system with increased length and density which allows greater contact with the soil for more water and nutrient absorption (Nielsen et al. 2001; Ma et al. 2001; Lopez-Bucio et al. 2002). Phenotyping of roots under field conditions often depends on traditional methods, like root excavation techniques for determining its length and density (Araus and Cairns 2014). The difficulties associated with root phenotyping can be resolved by utilizing modern root phenomics approaches. DoVale and Fritsche-Neto (2015) grouped the modern phenotyping platforms into two groups: ex situ and in situ analysis-based phenotyping. The ex situ evaluation utilizes hydroponics, aeroponics, agar medium, etc. for easily visualizing and capturing images. Sometimes, rhizotrons and minirhizotrons are also being used to study roots while still in soil (DoVale and Fritsche-Neto 2015). Digital scanning in combination with computerized image analysis is used for rapid evaluation of root morphology viz., diameter, length, branching, topology, etc. In addition to scanners, other devices such as microscope with vertical plates, digital cameras, different hardware for acquiring automated images, etc. can also be used for capturing images of roots. The images captured with these devices can be evaluated using software, like WinRHIZO, RootTrace, RootNav, etc. (Polomsky and Kuhn 2002). Likewise, GT-Roots (Global Traits of Root System) is an integrated Java-based open-source software developed for processing of root system images of dense cereal plants captured in a high-throughput phenotyping platform (Borriane et al. 2018).

Plants undergo different physiological and anatomical changes in response to stresses which are likely to affect their ability to tolerate stresses and are needed to be effectively identified for carrying out plant breeding researches. Apart from root phenomics, field phenotyping is very crucial in breeding varieties suitable for stress prone environment. Thermal imaging in conjunction with visible and near-infrared (NIR) images enable estimation of canopy temperature and selection of specific plant parts for water stress estimation (Jones et al. 2002; Moller et al. 2007). Early

vigor is an important trait for optimizing water use efficiency that minimizes direct evaporation from soil surface by boosting an early vegetative ground cover (Condon et al. 2004; Tuberosa 2012) and accumulate more carbohydrate reserves and water for survival of plants under drought stresses (Palta et al. 1994; Rebetzke et al. 2007). This can be measured by using different sensors and cameras. These nondestructive phenotyping tools detect and quantify the spectral reflectance arising due to interaction between plant parts and electromagnetic radiation at different spectral wavelengths such as visible (VIS: 400–700 nm), near-infrared (NIR: 700–1000 nm) and short-wave infrared (SWIR: 1000–2500 nm) and can offer high throughput and reproducible screening of early vigor (Fahlgren et al. 2015; Mulla 2013). Infra-red thermometry (IRT) or thermal imaging is used for sensing stomatal conductance and is widely used for irrigation scheduling (Jones 2004; Leinonen et al. 2006). The hyperspectral imaging technique has been successfully used in various remote sensing appliances for estimating the level of soil salinity (Poss et al. 2006; Hamzeh et al. 2013; Sytar et al. 2016) and the effect of soil salinity on different crops like, cotton, corn, cogon grass, etc. (Zhang et al. 2011).

18.3.2 Phenomics tool for biotic Stresses

Plant diseases affect food safety and security by causing huge amount of economic loss in yield and quality of farm produce. The most effective and sustainable way to minimize these economic losses is development of new cultivars with high level of resistance against devastating disease. It requires extensive phenotyping of germ-plasms and breeding lines which is time consuming, less reliable and labor intensive and hinders the pace of molecular marker-based breeding. The symptoms produced due to complex plant–pathogen interactions are sometimes highly variant and not visually apparent. Visual assessment of these symptoms may give inaccurate or imprecise results that ultimately affect the entire breeding program. Most of the resistance breeding approaches are being carried out with resistance or R-genes (Mutka and Bart 2015), which have been proven to be successful in many cases, but often the resistance is lost quickly due to rapid evaluation in pathogen (Kunkeaw et al. 2010; Dangel et al. 2013). Hence, quick and accurate assessment of plant responses at an early stage is very important in investigation of plant–pathogen interactions and development of resistant cultivars through modern molecular breeding programs.

During last decade, many sensor-based phenotyping tools were developed and used for detecting various stress symptoms occurring on plants in response to disease and pest attacks (Goggin et al. 2015; Mahlein 2016; Scholes and Rolfe 2009; White et al. 2012). High-throughput phenotyping (HTP) platform utilizes digital and fluorescence cameras, near-infrared or far-infrared sensors, laser scanner, hyperspectral cameras, magnetic resonance imaging (MRI), etc. for screening of disease symptoms (Goggin et al. 2015). Microscopic digital imaging can detect growth of fungal hyphae on leaves and quantify plant immune reaction. This technique has been used by Simko et al. (2014), to detect QTLs for powdery mildew

resistance in lettuce. Chlorophyll fluorescence imaging identifies pathogenic infections that abruptly alter photosynthetic activity of plants such as southern corn rust (Duraes et al. 2002), *Cercospora* leaf spot in sugar beet (Chaerle et al. 2007), TMV infection in tobacco (Chaerle et al. 2004), etc. Multi- and hyperspectral imaging sensors can detect all kinds of electromagnetic waves including UV and IR radiations which are generally not visible by human eye (Simko et al. 2017). Laboratory-based hyperspectral imaging technique has been reliably used for detecting head blight disease in winter wheat (Bauriegel et al. 2011a, b) and *Cercospora* leaf spot disease in sugar beet (Bergstrasser et al. 2015). Multispectral imaging has been used in remote sensing devices for knowing the health status of powdery mildew and leaf rust infected wheat crop under field conditions (Franke and Menz 2007). Susceptible plant–pathogen interactions may accompany local temperature changes due to stomatal closure and limited evaporation rates (Chaerle et al. 2007); resulting hypersensitive responses can be detected using thermal imaging (infra-red thermography) before appearance of visual symptoms. Magnetic resonance imaging can produce three-dimensional (3D) images of an object by using magnetic fields and radio waves and has been used to nondestructively detect diseases caused by soilborne basidiomycetes, cyst nematodes, and other belowground symptoms (Hillnhutter et al. 2012; Simko et al. 2017).

The sensor-based imaging techniques are also useful in capturing insect injuries such as defoliation caused by caterpillar, feeding scars of thrips on leaves and stem, necrosis and chlorosis caused by aphids, etc. (Hebert et al. 2007; Goggin et al. 2015). Alteration in photosynthesis, chlorophyll content, etc. caused by herbivores are well detected by chlorophyll fluorescence imaging techniques (Nabity et al. 2009; Kerchev et al. 2012). Likewise, multi- and hyperspectral imaging techniques enable remote sensing of insect infested field (Backoulou et al. 2011), diagnosis of pest damage intensity and detect cryptic herbivores like stem borer hiding within plant tissues (Goggin et al. 2015). Besides, high-throughput imaging tools also play important role in comparing base-line performance of different genotypes, monitoring the behavior of insects as vectors, visualizing plant defense mechanism, and so on (Goggin et al. 2015).

18.4 Conclusion

High-throughput phenotyping tools measure various changes occurring within plants in response to abiotic and biotic stresses with a greater accuracy and precision. All these sensor-based phenotyping techniques in conjunction with disease or insect diagnosis assays and automatic weather monitoring facilities will not only simplify the process of screening but will also serve well in early detection of disease or insect infestation for timely application of management practices. Also, rapid advancement in next-generation sequencing and resequencing techniques, genome sequencing, development of high-throughput molecular markers like SNPs (single nucleotide polymorphisms), etc. have provided a deep insight into genetic variations associated with complex traits produced due to plants' response towards

various biotic and abiotic stresses and require precise phenotyping data for their efficient utilization in stress breeding program.

All genome-based breeding techniques such as marker-assisted selection, genome-wide association studies, gene/QTL mapping, reverse genetics approaches like TILLING, EcoTILLING, gene silencing, insertional mutagenesis, etc. generate huge amount of biological data and require precise phenotyping of thousands of plants under diverse environmental scenario for their implication in crop improvement. Automated phenotyping tools are capable of capturing information on structure, function, and phenotypic expression of large number of genotypes under varied environmental conditions; analyzing, organizing, and storing the information in different datasets; and ultimately producing models to disentangle and simulate plant's performance in a range of environmental scenario (Tardieu et al. 2017). Field phenotyping is highly crucial for screening the performance of large number of genotypes under varied climatic conditions. Potential use of these nondestructive automated phenomics tools for stress sensing will hasten the climate resilient breeding procedure by simplifying the screening and selection of tolerant genotypes. Hence, high-throughput phenomics is an indispensable tool needed to bridge the gap between phenotyping and genotyping and witness a dramatic advancement in crop improvement and cope the challenging task of breeding stress-resistant or climate resilient varieties in a shorter period of time.

References

- Araus JL, Cairns JE (2014) Field high-throughput phenotyping: the new crop breeding frontier. *Trends Plant Sci* 19:52–61
- Atkinson NJ, Lilley CJ, Urwin PE (2013) Identification of genes involved in the response to simultaneous biotic and abiotic stress. *Plant Physiol* 162:2028–2041
- Awlia M, Nigro A, Fajkus J, Schmoeckel SM, Negrao S, Santelia D, Trtlele M, Tester M, Jukowski MM, Panzarova K (2016) High-throughput non-destructive phenotyping of traits that contribute to salinity tolerance in *Arabidopsis thaliana*. *Front Plant Sci* 7:1414
- Backoulou GF, Elliott NC, Giles K, Phoofole M, Catana V, Mirik M, Michels J (2011) Spatially discriminating Russian wheat aphid induced plant stress from other wheat stressing factors. *Comput Electron Agric* 78:123–129
- Baker NR (2008) Chlorophyll fluorescence: a probe of photosynthesis in vivo. *Annu Rev Plant Biol* 59:89–113
- Bauriegel E, Giebel A, Herppich WB (2011a) Hyperspectral and chlorophyll fluorescence imaging to analyze the impact of *Fusarium culmorum* on the photosynthetic integrity of infected wheat ears. *Sensors* 11:3765–3779
- Bauriegel E, Giebel A, Geyer M, Schmidt U, Herppich W (2011b) Early detection of *Fusarium* infection in wheat using hyper-spectral imaging. *Comput Electron Agric* 75:304–312
- Bergstrasser S, Fanourakis D, Schmittgen S, Cendrero-Mateo MP, Jansen M, Scharf H, Rascher U (2015) HyperART: non-invasive quantification of leaf traits using hyperspectral absorption-reflectance transmittance imaging. *Plant Methods* 11(1):17
- Blum A (2006) Drought adaptation in cereal crops: a prologue. In: Ribaut JM (ed) *Drought adaptation in cereals*. The Haworth Press, Binghamton, pp 3–15
- Borianne P, Subsol G, Fallavier F, Dardou A, Audebert A (2018) GT-RootS: an integrated software for automated root system measurement from high-throughput phenotyping platform images. *Comput Electron Agric* 150:328–342

- Borisjuk L, Rolletschek H, Neuberger T (2012) Surveying the plant's world by magnetic resonance imaging. *Plant J* 70:129–146
- Cabrera-Bosquet L, Crossa J, von Zitzewitz J, Serret MD, Luis Araus J (2012) High-throughput phenotyping and genomic selection: the frontiers of crop breeding converge. *J Integr Plant Biol* 54:312–320
- Chaerle L, Hagenbeek D, BruyneDe E, Valcke R, Van Der Straeten D (2004) Thermal and chlorophyll-fluorescence imaging distinguish plant-pathogen interactions at an early stage. *Plant Cell Physiol* 45:887–896
- Chaerle L, Hagenbeek D, BruyneDe E, Van Der Straeten D (2007) Chlorophyll fluorescence imaging for disease-resistance screening of sugarbeet. *Plant Cell Tissue Org* 91:97–106
- Choudhary A, Pandey P, Senthil-Kumar M (2016) Tailored responses to simultaneous drought stress and pathogen infection in plants. In: Hossain MA, Wani SH, Bhattacharjee S, Burritt DJ, LSP T (eds) *Drought stress tolerance in plants*, vol 1. Springer International Publishing, Cham, pp 427–438
- Collins NC, Tardieu F, Tuberosa R (2008) Quantitative trait loci and crop performance under abiotic stress: where do we stand? *Plant Physiol* 147:469–486
- Condon AG, Richards RA, Rebetzke GJ, Farquhar GD (2004) Breeding for high water-use efficiency. *J Exp Bot* 55:2447–2460
- Dangl JL, Horvath DM, Staskawicz BJ (2013) Pivoting the plant immune system from dissection to deployment. *Science* 341:746–751
- Dhankher OP, Foyer CH (2018) Climate resilient crops for improving global food security and safety. *Plant Cell Environ* 41:877–884
- Din M, Zheng W, Rashid M, Wang S, Shi Z (2017) Evaluating hyperspectral vegetation indices for leaf area index estimation of *Oryza sativa* L. at diverse phenological stages. *Front Plant Sci* 8:820
- DoVale JC, Fritsche-Neto R (2015) Root phenomics. In: Fritsche-Neto R, Borem A (eds) *Phenomics*. Springer, Cham
- Duraes F, Gama E, Magalhaes P, Marriel I, Casela C, Oliveira A, Luchiari A, Shanahan J (2002) The usefulness of chlorophyll fluorescence in screening for disease resistance, water stress tolerance, aluminum toxicity tolerance and N use efficiency in maize. In: *Proceedings of the Eastern and Southern Africa Regional Maize Conference*, Nairobi, Kenya, pp 356–360
- Espeland EK, Kettenring KM (2018) Strategic plant choices can alleviate climate change impacts: a review. *J Environ Manag* 222:316–324
- Fahlgren N, Gehan MA, Baxter I (2015) Lights, camera, action: high-throughput plant phenotyping is ready for a close-up. *Curr Opin Plant Biol* 24:93–99
- FAO (2009) How to feed the world in 2050. http://www.fao.org/fileadmin/templates/wsfs/docs/expert_paper/How_to_Feed_the_World_in_2050.pdf
- Finkel E (2009) With 'Phenomics', plant scientists hope to shift breeding into overdrive. *Science* 325:380–381
- Fiorani F, Rascher U, Jahnke S, Schurr U (2012) Imaging plants dynamics in heterogenic environments. *Curr Opin Biotechnol* 23:227–235
- Franke J, Menz G (2007) Multi-temporal wheat disease detection by multi-spectral remote sensing. *Precis Agric* 8:161–172
- Furbank RT, Tester M (2011) Phenomics—technologies to relieve the phenotyping bottleneck. *Trends Plant Sci* 16:635–644
- Ge Y, Bai G, Stoerger V, Schnable JC (2016) Temporal dynamics of maize plant growth, water use and leaf water content using automated high throughput RGB and hyperspectral imaging. *Comput Electron Agric* 127:625–632
- Goggin FL, Lorence A, Topp CN (2015) Applying high-throughput phenotyping to plant–insect interactions: picturing more resistant crops. *Curr Opin Insect Sci* 9:69–76
- Golzarian MR, Frick RA, Rajendran K, Berger B, Roy S, Tester M, Lun DS (2011) Accurate inference of shoot biomass from high-throughput images of cereal plants. *Plant Methods* 7:2
- Granier C, Aguirrezabal L, Chenu K, Cookson SJ, Dauzat M, Hamard P, Thioux JJ, Rolland G, Bouchier-Combaud S, Lebaudy A, Muller B (2006) PHENOPSIS, an automated platform for

- reproducible phenotyping of plant responses to soil water deficit in *Arabidopsis thaliana* permitted the identification of an accession with low sensitivity to soil water deficit. *New Phytol* 169:623–635
- Hairmans A, Berger B, Tester M, Roy SJ (2014) Image-based phenotyping for non-destructive screening of different salinity tolerance traits in rice. *Rice* 7:16
- Hamzeh S, Naseri AA, AlaviPanah SK, Mojaradi B, Bartholomeus HM, Clevers JGPW, Behzad M (2013) Estimating salinity stress in sugarcane fields with space borne hyperspectral vegetation indices. *Int J Appl Earth Obs Geoinf* 21:282–290
- Hatfield JL, Prueger JH (2015) Temperature extremes: effect on plant growth and development. *Weather Clim Extrem* 10:4–10
- Hebert SL, Jia L, Goggin FL (2007) Quantitative differences in aphid virulence and foliar symptom development on tomato plants carrying the Mi resistance gene. *Environ Entomol* 36:458–467
- Hillnhutter C, Sikora RA, Oerke EC, Van-Dusschoten D (2012) Nuclear magnetic resonance: a tool for imaging belowground damage caused by *Heterodera schachtii* and *Rhizoctonia solani* on sugar beet. *J Exp Bot* 63:319–327
- Honsdorf N, March TJ, Berger B, Tester M, Pillen K (2014) High-throughput phenotyping to detect drought tolerance QTL in wild barley introgression lines. *PLoS One* 9:e97047
- Hu HH, Dai MQ, Yao JL, Xiao BZ, Li XH, Zhang QF, Xiong LZ (2006) Overexpressing a NAM ATAF, and CUC (NAC) transcription factor enhances drought resistance and salt tolerance in rice. *Proc Natl Acad Sci U S A* 103:12987–12992
- Humphik JF, Lazar D, Husickova A, Spichal L (2015) Automated phenotyping of plant shoots using imaging methods for analysis of plant stress responses—a review. *Plant Methods* 11:29
- Jahnke S, Menzel MI, Van Dusschoten D, Roeb GW, Beuhler J, Minwuyet S, Bleumler P, Temperton VM, Hombach T, Streun M, Beer S (2009) Combined MRI–PET dissects dynamic changes in plant structures and functions. *Plant J* 59:634–644
- Jansen M, Gilmer F, Biskup B, Nagel KA, Rascher U, Fischbach A, Briem S, Dreissen G, Tittmann S, Braun S, De Jaeger I (2009) Simultaneous phenotyping of leaf growth and chlorophyll fluorescence via GROWSCREEN FLUORO allows detection of stress tolerance in *Arabidopsis thaliana* and other rosette plants. *Funct Plant Biol* 36:902–914
- Jones HG (2004) Application of thermal imaging and infrared sensing in plant physiology and ecophysiology. *Adv Bot Res* 41:107–163
- Jones HG, Stoll M, Santos T, Sousa CD, Chaves MM, Grant OM (2002) Use of infrared thermography for monitoring stomatal closure in the field: application to grapevine. *J Exp Biol* 53:2249–2260
- Jones HG, Serraj R, Loveys BR, Xiong L, Wheaton A, Price AH (2009) Thermal infrared imaging of crop canopies for the remote diagnosis and quantification of plant responses to water stress in the field. *Funct Plant Biol* 36:978–989
- Jones AM, Danielson JA, Kumar MSN, Lanquar V, Grossmann G, Frommer WB (2014) Abscissic acid dynamics in roots detected with genetically encoded FRET sensors. *elife* 3:e01741
- Kastberger G, Stachl R (2003) Infrared imaging technology and biological applications. *Behav Res Methods Instrum Comput* 35:429–439
- Kerchev PI, Fentoni B, Foyer CH, Hancock RD (2012) Plant responses to insect herbivory: interactions between photosynthesis, reactive oxygen species and hormonal signaling pathways. *Plant Cell Environ* 35:441–453
- Kiyomiya S, Nakanishi H, Uchida H, Tsuji A, Nishiyama S, Futatsubashi M, Tsukada H, Ishioka NS, Watanabe S, Ito T, Mizuniwa C (2001) Real time visualization of ^{13}N -translocation in rice under different environmental conditions using positron emitting tracer imaging system. *Plant Physiol* 125:1743–1753
- Kumar M (2013) Crop plants and abiotic stresses. *J Biomol Res Ther* 3:e125. <https://doi.org/10.4172/2167-7956.1000e125>.
- Kunkeaw S, Tan S, Coaker G (2010) Molecular and evolutionary analyses of *Pseudomonas syringae* pv. *tomato* race1. *Mol Plant Microbe Interact* 23:415–424
- Leinonen I, Grant OM, Tagliavia CPP, Chaves MM, Jones HG (2006) Estimating stomatal conductance with thermal imagery. *Plant Cell Environ* 29:1508–1518

- Li L, Zhang Q, Huang D (2014) A review of imaging techniques for plant phenotyping. *Sensors* 14:20078–20111
- Long SP, Marshal-Colon A, Zhu XG (2015) Meeting the global food demand of the future by engineering crop photosynthesis and yield potential. *Cell* 161:56–66
- Lopez-Bucio JL, Hernandez-Abreu E, Sanchez-Calderon L, Nieto-Jacobo MF, Simpson J, Herrera-Estrella L (2002) Phosphate availability alters architecture and causes changes in hormone sensitivity in the *Arabidopsis* root system. *Plant Physiol* 129:244–252
- Ma Z, Bielenberger DF, Brown KM, Lynch JP (2001) Regulation of root hair density by phosphorus availability in *Arabidopsis thaliana*. *Plant Cell Environ* 24:459–467
- Mahalingam R (ed) (2015) Consideration of combined stress: a crucial paradigm for improving multiple stress tolerance in plants. *Combined stresses in plants*. Springer International Publishing, Cham, pp 1–25
- Mahlein AK (2016) Plant disease detection by imaging sensors—parallels and specific demands for precision agriculture and plant phenotyping. *Plant Dis* 100:241–251
- Maphosa L, Thoday-Kennedy E, Vakani J, Phelan A, Badenhorst P, Slater A, Spangenberg G, Kant S (2016) Phenotyping wheat under salt stress conditions using a 3D laser scanner. *Isr J Plant Sci* 1:1–8
- McDonald A, Riha S, DiTommaso A, DeGaetano A (2009) Climate change and the geography of weed damage: analysis of U.S. maize systems suggests the potential for significant range transformations. *Agric Ecosyst Environ* 130:131–140
- Meng R, Saade S, Kurtek S, Berger B, Brien C, Pillen K, Tester M, Sun Y (2017) Growth curve registration for evaluating salinity tolerance in barley. *Plant Methods* 13:18
- Merlot S, Mustilli AC, Genty B, North H, Lefebvre V, Sotta B, Vavasseur A, Giraudat J (2002) Use of infrared thermal imaging to isolate *Arabidopsis* mutants defective in stomatal regulation. *Plant J* 30:601–609
- Mickelbart MV, Hasegawa PM, Bailey-Serres J (2015) Genetic mechanisms of abiotic stress tolerance that translate to crop yield stability. *Nat Rev Genet* 16:237–251
- Moller M, Alchanatis V, Cohen Y, Meron M, Tsipris J, Naor A, Ostrovsky V, Sprintsin M, Cohen S (2007) Use of thermal and visible imagery for estimating crop water status of irrigated grapevine. *J Exp Bot* 58:827–838
- Mulla DJ (2013) Twenty-five years of remote sensing in precision agriculture: key advances and remaining knowledge gaps. *Biosyst Eng* 114:358–371
- Munns R, James RA, Sirault X (2010) New phenotyping methods for screening wheat and barley for beneficial responses to water deficit. *J Exp Bot* 61:3499–3507
- Mutka AM, Bart RS (2015) Image-based phenotyping of plant disease symptoms. *Front Plant Sci* 5:1–8
- Nabity PD, Zavala JA, DeLucia EH (2009) Indirect suppression of photosynthesis on individual leaves by arthropod herbivory. *Ann Bot* 103:655–663
- Nielsen KL, Eshel A, Lynch JP (2001) The effect of P availability on the carbon economy of contrasting common bean (*Phaseolus vulgaris* L.) genotypes. *J Exp Bot* 52:329–339
- Neilson EH, Edwards AM, Blomstedt CK, Berger B, Moller BL, Gleadow RM (2015) Utilization of a high-throughput shoot imaging system to examine the dynamic phenotypic responses of a C-4 cereal crop plant to nitrogen and water deficiency over time. *J Exp Bot* 66:1817–1832. <https://doi.org/10.1093/jxb/eru526>
- Osmond B, Ananyev G, Berry J, Langdon C, Kolber Z, Lin G, Monson R, Nichol C, Rascher U, Schurr U, Smith S (2004) Changing the way we think about global change research: scaling up in experimental ecosystem science. *Glob Chang Biol* 10:393–407
- Palta JA, Kobata T, Turner NC, Fillery IR (1994) Remobilization of carbon and nitrogen in wheat as influenced by post anthesis water deficits. *Crop Sci* 34:118–124
- Pandey P, Irulappan V, Bagavathiannan MV, Senthil-Kumar M (2017) Impact of combined abiotic and biotic stresses on plant growth and avenues for crop improvement by exploiting physiological traits. *Front Plant Sci* 8:1–15
- Paprocki A, Sirault X, Berry S, Furbank R, Fripp J (2012) A novel mesh processing based technique for 3D plant analysis. *BMC Plant Biol* 12:63–71

- Penuelas J, Filella I (1998) Visible and near-infrared reflectance techniques for diagnosing plant physiological status. *Trends Plant Sci* 3:151–156
- Pereira A (2016) Plant abiotic stress challenges from the changing environment. *Front Plant Sci* 7:1123
- Peters K, Breitsameter L, Gerowitt B (2014) Impact of climate change on weeds in agriculture: a review. *Agric Sustain Dev* 34:707–721
- Polomsky J, Kuhn N (2002) Root research methods. In: Waisel Y, Eshel A, Kafkafi U (eds) *Plantroots: the hidden half*, 3rd edn. Marcel Dekker, New York, pp 447–487
- Poorter H, Fiorani F, Stitt M, Schurr U, Finck A, Gibon Y, Usadel B, Munns R, Atkin OK, Tardieu F, Pons TL (2012) The art of growing plants for experimental purposes: a practical guide for the plant biologist. *Funct Plant Biol* 39:821–838
- Poss JA, Russell WB, Grieve CM (2006) Estimating yields of salt- and water stressed forages with remote sensing in the visible and near infrared. *J Environ Qual* 35:1060–1071
- Prasch CM, Sonnewald U (2013) Simultaneous application of heat, drought and virus to *Arabidopsis* plants reveals significant shifts in signaling networks. *Plant Physiol* 162:1849–1866
- Rahaman MM, Chen D, Gillani Z, Klukas C, Chen M (2015) Advanced phenotyping and phenotype data analysis for the study of plant growth and development. *Front Plant Sci* 6:619
- Ramegowda V, Senthil-Kumar M (2015) The interactive effects of simultaneous biotic and abiotic stresses on plants: mechanistic understanding from drought and pathogen combination. *J Plant Physiol* 176:47–54
- Ramu VS, Paramanathan A, Ramegowda V, Mohan-Raju B, Udaya-Kumar M, Senthil-Kumar M (2016) Transcriptome analysis of sunflower genotypes with contrasting oxidative stress tolerance reveals individual- and combined-biotic and abiotic stress tolerance mechanisms. *PLoS One* 11(6):e0157522
- Rascher U, Heutt MT, Siebke K, Osmond B, Beck F, Leutge U (2001) Spatio-temporal variations of metabolism in a plant circadian rhythm: the biological clock as an assembly of coupled individual oscillators. *Proc Natl Acad Sci U S A* 98:11801–11805
- Raza A, Razzaq A, Mehmood SS, Zou X, Zhang X, Lv Y, Xu J (2019) Impact of climate change on crops adaptation and strategies to tackle its outcome: a review. *Plan Theory* 8:34
- Rebetzke GJ, Ellis MH, Bonnett DG, Richards RA (2007) Molecular mapping of genes for coleoptiles growth in bread wheat (*Triticum aestivum* L.). *Theor Appl Genet* 114:1173–1183
- Romer C, Wahabzada M, Ballvora A, Pinto F, Rossini M, Panigada C, Behmann J, Leon J, Thureau C, Bauckhage C, Kersting K (2012) Early drought stress detection in cereals: simplex volume maximization for hyperspectral image analysis. *Funct Plant Biol* 39:878–890
- Scholes JD, Rolfe SA (2009) Chlorophyll fluorescence imaging as tool for understanding the impact of fungal diseases on plant performance: a phenomics perspective. *Funct Plant Biol* 36:880–892
- Simko I, Rauscher G, Sideman RG, McCreight JD, Hayes RJ (2014) Evaluation and QTL mapping of resistance to powdery mildew in lettuce. *Plant Pathol* 63:344–353
- Simko I, Jimenez-Berni JA, Sirault XRR (2017) Phenomic approaches and tools for phytopathologists. *Phytopathology* 107:6–17
- Sozzani R, Busch W, Spalding EP, Benfey PN (2014) Advanced imaging techniques for the study of plant growth and development. *Trends Plant Sci* 19:304–310
- Suzuki N, Koussevitzky S, Mittler R, Miller G (2012) ROS and redox signalling in the response of plants to abiotic stress. *Plant Cell Environ* 35:259–270. <https://doi.org/10.1111/j.1365-3040.2011.02336.x>
- Sytar O, Brestic M, Zivcak M, Olsovska K, Kovar M, Shao H, He X (2016) Applying hyperspectral imaging to explore natural plant diversity towards improving salt stress tolerance. *Sci Total Environ*. <https://doi.org/10.1016/j.scitotenv.2016.08.014> 578:90
- Tardieu F, Cabrera-Bosquet L, Pridmore T, Bennett M (2017) Plant phenomics, from sensors to knowledge. *Curr Biol* 27:770–783
- Tuberosa R (2012) Phenotyping for drought tolerance of crops in the genomics era. *Front Physiol* 3:1–25
- Verma AK, Singh D (2016) Abiotic stress and crop improvement: current scenario. *Adv Plants Agric Res* 4:345–346

- Weirman A (2010) Plant phenomics teacher resource. http://www.plantphenomics.org.au/files/teacher/FinalPhenomicsforwordwith_image.doc
- White JW, Andrade-Sanchez P, Gore MA, Bronson KF, Coffelt TA, Conley MM, Feldmann KA, French AN, Heun JT, Hunsaker DJ (2012) Field-based phenomics for plant genetics research. *Field Crops Res* 133:101–112
- Windt CW, Vergeldt FJ, De Jager PA, Van AH (2006) MRI of long distance water transport: a comparison of the phloem and xylem flow characteristics and dynamics in poplar, castor bean, tomato and tobacco. *Plant Cell Environ* 29:1715–1729
- Yang W, Duan L, Chen G, Xiong L, Liu Q (2013) Plant phenomics and high-throughput phenotyping: accelerating rice functional genomics using multidisciplinary technologies. *Curr Opin Plant Biol* 16:180–187
- Zhang TT, Zeng SL, Gao Y, Ouyang ZT, Li B, Fang CM, Zhao B (2011) Using hyperspectral vegetation indices as a proxy to monitor soil salinity. *Ecol Indic* 11:1552–1562
- Zhang X, Huang C, Wu D, Qiao F, Li W, Duan L, Wang K, Xiao Y, Chen G, Liu Q, Xiong L (2017) High-throughput phenotyping and QTL mapping reveals the genetic architecture of maize plant growth. *Plant Physiol* 173:1554–1564
- Ziska LH, Tomecek MB, Gealy DR (2010) Evaluation of competitive ability between cultivated and red weedy rice as a function of recent and projected increases in atmospheric CO₂. *Agron J* 102:118–123