



## High-throughput Phenotyping for Abiotic Stress Resilience in Cereals

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### Abstract

Around 70 percent of crop yield losses are projected because of climate induced abiotic stresses like moisture stress, soil salinity and heat stress. Critical to the stability of cropping systems in the face of climate change is the capacity to rapidly grow germplasm with tolerance to complex polygenic inherited abiotic and biotic stresses combined. Molecular breeding provides the means to speed up cereal breeding, but adequate phenotyping protocols are required to ensure that the much-anticipated benefits of novel breeding tools can be realized. Tremendous advances in phenomics have taken place in the recent past. Both forward and reverse phenomics have developed to help determine either the best genotype with the desired feature or mechanism and the genes that make the genotype the best. High-throughput phenomics studies include techniques for screening substantial germplasm sets for a specific trait using high-throughput phenotyping (HTP) technologies like advanced robots, high-tech sensors, 3D imaging, fluorescence imaging, NIR imaging, lemnatec, etc. The high velocity of plant phenotyping based on phenomics accelerates the selection phase of potential advanced germplasm resilient towards climate induced stresses.

**Key words:** Abiotic stress Resilience, Forward and reverse phenomics, High-throughput phenotyping Phenomics

## 1. Introduction

By 2050, the world's population is projected to hit 9 billion and the current production of food must be doubled to meet the needs of a rising population (Tester and Langridge, 2011, Ganie *et al.*, 2021). Currently, intermittent precipitation events, high temperatures, drought, salinity stress, etc. are more likely to occur due to global climate change, which has increased the pace of biotic and abiotic stresses (Wani *et al.*, 2018; Atif *et al.*, 2019). In turn, these biotic and abiotic stresses pose a deteriorating risk to crop yield and quality, making it difficult to resolve the global food challenge (Pereira,

2016, Wani *et al.*, 2021). Cereal crops such as Wheat are grown under rainfed conditions due to which they become more prone to abiotic stresses. Hence in-depth knowledge and mechanisms involved in drought effect on wheat metabolism is vital for evolving drought-tolerant wheat varieties (Itam *et al.*, 2020). To overcome these potential challenges of climate change, the production of elite germplasm can deal with abiotic stresses or the development of climate-smart germplasm will be a priority (Leakey *et al.*, 2009; Ziska and Bunce, 2007; Wani *et al.*, 2020a). The yield potential and stability of a genotype are



important for increasing crop production. Development of genotype having tolerance to abiotic stresses is a key factor in crop output stability (Furbank and Tester, 2011, Azhar *et al.*, 2020; Kumar *et al.*, 2021). A researcher must thoroughly analyze the response in terms of phenotypic changes and the processes that control the response of a plant under stressed conditions in order to determine the ability of a given genotype under a specific abiotic stress (Mickelbart *et al.*, 2015).

In genomics, considerable progress has been made, especially the rapid developments in next generation sequencing (NGS) technologies or molecular breeding approaches such as marker assisted breeding (MAB), marker assisted backcross (MABC) or marker assisted recurrent selection (MARS), etc., to improve crop varieties that show tolerance to stress conditions (Varshney *et al.*, 2014; Wani, 2019; Wani *et al.*, 2020b). However, there is a substantial gap between genomic knowledge and its use in practical crop improvement, and the most important reason is that accurate and high-performance phenotyping tools are not usable, leading to poor gene/QTL results for genomics-assisted breeding (Varshney *et al.*, 2012; Choudhary *et al.*, 2019; Gosal *et al.*, 2020).

The new age of phenomics has provided scientists with the tools to unlock the data coded in plant genomes (Finkel, 2009; Yang *et al.*, 2020). The high speed of plant phenotyping based on phenomics and its ability to produce full sets of field data have accelerated the selection phenomenon potential elite advanced lines that execute well during stress conditions (Montes *et al.*, 2007). An all-inclusive tactic to plant phenotyping helps to polish our awareness of environmental-influenced characteristics. The advanced tools presented by phenomics allow scientists to categorize germplasm with abiotic stress tolerance effectively.

## **2. HTP platforms used for phenotyping abiotic stress resilience related traits**

In several institutions around the world, a large range of fully automated high-throughput phenotyping systems are available based on non-destructive measurements (Furbank and Tester, 2011). By using high-tech automated sensors, imaging devices, and computing resources, HTP platforms accelerate phenotyping (Furbank, 2009; Yang *et al.*, 2020). The HTP platforms include imaging techniques viz., 3-D imaging, near infrared imaging, far infrared

imaging, fluorescence imaging, visible light scanning, hyperspectral imaging, magnetic resonance imaging and positron emission tomography, lemnaTec technology, phenonet, phenomobile, dunken technologies MS31000 imager, phenocopter, helium-filled aerostats and phenotower (Rahman *et al.*, 2015).

These systems include clear imaging features, mechanical transport for the mobility of imaging plants, efficient imaging formulas and data analytics (Gerke *et al.*, 2009). These HTP platforms have emerged as advanced plant phenotyping tools integrated with advanced software systems (Paproki *et al.*, 2012).

### **2.1 3D (Three-Dimensional) Imaging**

3D plant images are obtained by employing many cameras connected by a computer package to capture multiple images from various angles (Weirman, 2010). It is used to calculate the number of leaves, form, angle, colour, health of the leaf, tiller number, mass of the shoot, etc. For 3D imaging and mapping in plants, two methods are most widely used; foremost, laser imaging detection and ranging (LIDAR) techniques that image a act (Omasa *et al.*, 2007) and, secondly, stereo photography by means of more than two cameras (Biskup *et al.*, 2007).

For QTL mapping, Topp *et al.* (2013) used semi-automated 3D imaging and digital phenotyping to classify core regions of the root architecture managed by the rice genome. It has been used in *Arabidopsis thaliana* to calculate yield characteristics, root architecture, imbibition, germination rates, etc. under drought stress (Woo *et al.*, 2008).

### **2.2 Far-infrared Imaging**

FIR imaging is used either inside the leaves of one plant or between different plants to calculate temperature differences. FIR imaging can also be used to detect plants with cool canopies in the fields (Leinonen and Jones, 2004). FIR cameras can also calculate changes in stomatal conductance so that the photosynthesis rate can be calculated (Wang *et al.*, 2004). Differences in temperature is used to calculate photosynthetic behavior, salinity tolerance and efficient quality of water usage (Lafitte *et al.*, 2003).

With the upsurge in canopy temperature of a shrub with mounting salt levels, FIR imaging has been used to screen a number of plant varieties for salinity stress resilience in barley (Sirault *et al.*, 2009). So, the canopy of plants



that can sustain lower temperatures when imaged in the presence of salt with the FIR camera is extra salt resilient and thus grows well in high saline soils. By means of high-velocity imaging with FIR, Salt tolerance can be assessed at the stage of seedlings as well (Cook *et al.*, 2012).

### *2.3 Near- infrared imaging*

Infrared thermal imaging enables the imaging of infrared radioactivity released from the target by visualization. For imaging, this machinery uses inner molecular motions of substances emitting infrared radiation (Kastberger and Stachl, 2003). Infrared imaging is conducted at two unique wavelength ranges of light, one at a near-infrared (NIR) range of 0.9–1.55  $\mu\text{m}$  and another at a far-infrared (Far-IR) wavelength range of 7.5–13.5  $\mu\text{m}$ . NIR imaging is used to monitor leaf and soil water content and motion, while far-infrared (FIR) cameras are used to study temperature (Weirman, 2010). The NIR calculation of the soil is performed to determine the rate of absorption of water by the roots. NIR imaging is employed in seed kernels to analyze the amount of starch present in leaves in addition to oil and protein (Jones *et al.*, 2009). NIR has been used to test for drought stress germplasms such as rice and soybeans (Seelig *et al.*, 2008). NIR quantifies characteristics associated with osmotic resistance under salinity stress in wheat and barley, e.g. (Lobet *et al.*, 2011).

### *2.4 Fluorescence imaging*

Fluorescence imaging is an effective technique for plant stress determination. The photosynthetic apparatus is connected to chlorophyll fluorescence and these measurements help track and calculate how plants and leaves react to biotic and abiotic stress (Meroni *et al.*, 2009). Fluorescence can be used to research simple chlorophyll emission through pulse amplitude-modulated (PAM) fluorescence to evaluate photosystem II behavior. Fluorescence imaging has also been used to study drought responses under controlled environments (Woo *et al.*, 2008). Investigates photosynthetic performance under stressful environments e.g in wheat, tomato (Jansen *et al.*, 2009). Increase in the chlorophyll fluorescence ratios depict the stress induced decrease in chlorophyll content and are very early stress indicators (Lichtenthaler *et al.*, 1996).

### *2.5 Visible Light (Monochromatic or Color) Imaging*

Visible light imaging operates at a wavelength of between 400 and 700 nm. Digital cameras can use two-dimensional

(2D) images to examine shot biomass, yield attributing characters (Duan *et al.*, 2011), leaf structure (Bylesjo *et al.*, 2008), panicle features (Ikeda *et al.*, 2010) and root traits (Ikeda *et al.*, 2010). During green stage, rosette plants are usually photographed from the tip (Jansen *et al.*, 2009). Moller *et al.* (2007) used thermal and visible imaging to estimate the status of irrigated grapevines in crop water. Three-dimensional imaging has been developed for the rice root system along with the software framework (RootReader3D).

Using a 3D digitizer and L-system formalism, '3D virtual rice' was developed to demonstrate the differences in structure and development between cultivars and under different environmental conditions. (Watanabe *et al.*, 2005). The color data gives an estimate of the degree of senescence. Senescence during drought of older leaves suggests escape or avoidance. It is possible to distinguish stay-green genotypes that could continue photosynthesis under water stress (Foulkes *et al.*, 2007).

### *2.6 Hyperspectral Imaging*

The continuous spectrum in the entire visible and NIR region is used in hyperspectral imaging and is used for more complex studies of various forms of pigmentation, water, biochemical compositions, and Vis. The proportion of light reflected by non-transparent surfaces is spectral reflectance. To detect plants stressed by salty soil or drought, researchers use this spectral reflectance long before it can be eye-catching (Bock *et al.*, 2010). Under heat and drought stress it has been used to measure leaf and canopy temperature in rice and panicle health status in wheat (Dale *et al.*, 2013).

### *2.7 Positron emission tomography (PET)*

PET is a method which nondestructively images the dispersal of positron-emitting radionuclide-labelled compounds such as C-11, N-13 or Fe- 52 (Kiyomiya *et al.*, 2011; Tsukamoto *et al.*, 2009). It envisages the distribution and transport of metabolites labeled with elements of positron-emitting radionuclides therefore assists in the study of plant metabolism. In plants along a transport road. Buhler *et al.*, 2011 proved transport velocity and lateral loss or photo assimilate. When used together with MRI and PET, both structural and functional characteristics can be analyzed simultaneously. PET has been used to visualize shoot to root translocation in sugar beet and radish under salinity stress, root architecture under heat stress in rice



and wheat and phenology and physiology under drought stress in barley (Jahnke *et al.*, 2009).

### *2.8 Magnetic Resonance imaging (MRI)*

In order to take images, MRI includes a mixture of magnetic field and radio waves. It is widely adopted for imaging plant roots. By detecting nuclear resonance signals, MRI offers structural knowledge about inner physiological processes. The symptoms caused by cyst nematodes are visualized via MRI (Hillnhutter *et al.*, 2012). It has also been used in stress conditions to assess the water content of rice, wheat and barley. It has also been used in many crops such as poplar, castor bean, tomato and tobacco to provide practical data such as water diffusion and transportation in plants (Windt *et al.*, 2006)

### *2.9 Lemna-Tec technology*

LemnaTec, a German company, has developed a high-throughput integrated phenotyping platform that includes the pipeline of imaging, automation of image processing and data handling modules. The platform has the capacity to easily measure almost unlimited sets of parameters, allow comprehensive screening and dynamically provide statistics on different plant characteristics. To measure plant height/width, biomass and plant architecture, these chambers offer top and side imaging of both plant roots as well as canopy (Mir *et al.*, 2012).

Color, form, size and design research. Lemna Tec can significantly boost the precision and throughput of phenotyping, thereby helping to better elucidate the genetic regulation of complex characteristics of drought tolerance in plants. Bringing integrative phenotyping technology from controlled environments to the field, LemnaTec will improve the evaluation of plant responses to drought while allowing for high-throughput screening and comprehensive and accurate phenotypic data to be produced. In connection with high-throughput conveyor systems, Lemna-Tec has developed unique water management hardware and software tools to investigate how plants respond to drought stress at various time scales. It has been used in barley, tomato, corn, sorghum, Arabidopsis etc. under drought pressure and salt stress in barley, rice etc. (Araus and Cairns 2014).

### *Phenonet*

Phenonet is the CSIRO model data logger for sensors such as the FIR thermometer, chlorophyll fluorescence sensors,

soil moisture sensors, camera and weather station. It tests the temperature of the canopy and photosynthetic plant activity. It tracks changes in the climate across the field and continuous growth and growth of crops (Weirman, 2010)

### *Phenomobile*

Phenomobile is a researcher-driven crop site vehicle fitted with digital cameras to estimate leaf greenness and ground cover, a far-infrared thermometer to assess canopy temperature, a 3D stereo imaging system to determine plant biomass. It flies at speeds of 3-5 km per hour through the field site and gathers dimensions of plot immediately below it and on both sides (White *et al.*, 2012)

### *Duncan Technologies MS3100 Imager*

Duncan Technologies MS3100 Imager provides three types of optical multi-spectral, infra-red and traditional cameras. It's put on a chopper. It photographs the canopy and, by taking 3D images, decides the biomass of the plant. Without a pilot, remote helicopters are a safer substitute to pilot-based aircraft because they permit flying at lesser altitudes and cost effective in operation (White *et al.*, 2012).

### *Phenocopter*

Phenocopter is a remote-controlled, gas powered model helicopter developed by Merz and Chapman in 2011 at CSIRO. It measures plant height, canopy cover, lodging and temperature. Fluorescence emissions are studied using hyperspectral and infrared cameras as a function of water stress (Tejada *et al.*, 2009). Utilizing pilot less helicopter for thermal and narrowband multispectral remote sensing for foliage capturing has been demonstrated by (Berni *et al.*, 2009).

### *Helium-filled aerostats*

Helium-filled 2 kg payload aerostats, harbored with camera scan infrared or color images of a field 30 to 80 m above ground level (Weirman, 2010). Jensen *et al.*, (2007) identified the use of an aerostat to track the feedback of wheat to nitrogen using digital cameras, using color and near-infrared images. In a cotton irrigation analysis, Ritchie *et al.* (2008) estimated evapotranspiration using data obtained by a two-camera system from the Normalized Difference Vegetation Index (NDVI).

### *Phenotower*

The Phenotower holds infrared and reflectance sensors and is mounted on a trolley. It collects data on proportional



canopy temperature, and ground cover among diverse germplasm at once, parallel to the vegetation (White *et al.*, 2012). By collecting multi-spectral data for vegetation, nutrient and water status indices at a spatial resolution of 1 m, it characterizes water and nitrogen stress for parcels (Haberland *et al.*, 2010).

### **3. Screening for Abiotic Stress Resilience Traits**

The main abiotic stresses that cause yield loss in agricultural crops worldwide are drought, salinity and temperature extremes. Phenotyping for resistance for such stresses is also a major task due to the complexity of governing systems responsible for tolerance for abiotic stresses (Vandenbroucke and Metzloff, 2013). But with the ability of effective high-throughput phenotyping methods, it's easy to non-destructively characterize various traits for broad germplasm sets under drought and heat stress conditions and improve the genetic dissection of complex stress tolerance mechanisms by providing reliable and accurate phenotypic data (Yang *et al.*, 2013). To record phenotypic deviations involves the use of sophisticated techniques to analyze the main parameters unique to stress, ensuing an accurate assessment of the phenotypic attribute. The degree of stress and extent of resistance or vulnerability of a cultivar has been assessed using a variety of parameters (Collins *et al.*, 2008).

#### **3.1. Salinity Stress**

Phenotypically, the rejoinder of plants to elevated external points of NaCl is complex. The approaches adapted for phenotyping capture the behavior that reacts to salinity. In two different phases, a plant's responses to salinity stress occur (Munns and Tester, 2008): The initial step is the osmotic phase, that begins immediately after the proportion of salt rises to a grave level around the roots and contrarywise disturbs the plant development by reducing the growth of new leaves and hindering the initiation of young leaves. Growth rate fluctuations can be calculated by regular plant imaging and a drop in the plant development triggered by salinity stress can be recorded (Rajendran *et al.*, 2009). Ion-specific response in the second phase is also called as tissue tolerance, that begins at an advanced phase when Na<sup>+</sup> or Cl amasses to lethal stages in the plant tissue, resulting to the untimely ageing of mature leaves. By means of color data of visible light imaging, the commencement and gradation

of agedness can be measured to enable quantification of tissue tolerance in plants. A non-destructive approach has been developed by Rajendran *et al.* (2009) to phenotype crop salinity tolerance machineries by assessing osmotic tolerance and tissue tolerance with a Scanalyzer 3D using visible light imaging techniques. In contrast to osmotic-sensitive plants, osmotic-tolerant plants display meager falls in comparative growth rate, which will show a severe reduction in growth when exposed to NaCl (Rahman *et al.*, 2015). Infrared thermography is an alternative method of calculating osmotic resistance. Stomatal conductance decreases down there by rising the temperature of the leaf because of salinity. Sirault *et al.* (2009) has established a phenotyping method based on this theory to quantify osmotic tolerance-related salt tolerance components for durum wheat and barley genotypes by using infrared thermography to measure leaf temperature. Recently, thermal imaging and Scanalyzer 3D designed by LemnaTec, has been considered as the best phenotyping technique to measure the various stresses including salinity stress in different crops and cereals and pulses as well (Dissanayake *et al.*, 2020; Pineda *et al.*, 2021; Song *et al.*, 2021).

#### **3.2. Drought stress**

Drought tolerance is a dynamic characteristic that determines plant growth under moisture stress environments, which remains a difficult characteristic for breeders to manipulate (Cattivelli *et al.*, 2008, Yang *et al.*, 2020). Plants utilize numerous internal processes to deal with moisture stress, such as escape, tolerance, recovery, and avoidance (O'Toole and Chang, 1979). Germplasm having deep, coarse roots with profuse branching and penetration capacity, leaf rolling elasticity, primary stomatal closure and elevated cuticular tolerance are recorded as component characteristics of dryness prevention (Wang and Yamauchi, 2006). Leaf rolling, decrease in leaf area, condensed stem elongation rate, and lesser transpiration rate are among the early mechanisms of drought prevention (Reddy *et al.*, 2003). For leaf area index measurement using digital infrared imaging, high-throughput phenotyping techniques are available (Shibayama *et al.*, 2011, Yang *et al.*, 2020).

The starting point for the development of root system modeling for different species of crops is unravelling of genetic differences for root system architecture. Several



automated phenotyping structures are accessible to research root system architecture by means of visible, infrared or hyperspectral imaging due to advances made in the field of phenomics. Drought tolerance has been analyzed in cereal crops using advances phenomics tools including visible infrared imaging and thermal imaging (Negin and Moshelion, 2017; Badigannavar et al., 2018; Khadka et al., 2020)

Drought escape is alternative key phenomenon related to drought stress tolerance: by way of earliness in maturity and completion of growth period early to avoid late stress. Using visible imaging, flowering time can be easily tracked and genotypes may be phenotyped for their drought escape performance. The temperature of the canopy is an accurate and precise drought stress gauge. The most common technique used for visualizing temperature variations is Far-IR (also called IR thermal) imaging. Recently, thermal imaging technique was employed to measure the transpiration efficiency as a trait for drought stress tolerance in wheat (Abdelhakim et al., 2021). Infrared thermography for measuring plant's ability to tolerate stress conditions was also reported by Siddiqui et al., 2019.

### 3.3. Heat/Cold Stress

Temperature stress in plants occurs at higher places having chilling/freezing temperatures. Following heat stress, phenotypic and biochemical characteristics of cultivated plant species change, such as meagre germination ratio, less seedling development, abnormal seedling development, less seedling vigor and reduced development of radicles and plumules (Jagadish et al., 2016) and can be measured using MRI and PET. Therefore, based on the affected characteristics, the performance of adapted genotypes to heat or cold stress and the ability of crop varieties is recorded.

Structural vagaries due to heat stress comprise leaf, twig, branch, and stem scorching and sunburning, leaf ageing and fall off, shoot and root growth failure, fruit staining and permanent damage that can be quantified by visible light imaging to determine the resistant genotypes (Vasseur et al., 2014). The stay-green (SG) is a minor trait that permits crop plants to preserve their green leaves and photosynthesis ability for a lengthier time subsequent to anthesis, particularly under drought and heat stress environments. Therefore, SG plants have more grain-

filling time and afterward more yield than non-SG (Kamal et al., 2019). In another study to characterize barley germplasm for heat stress various morphological traits were taken into consideration including days to flowering, grains per spike and yield per plot with further addition of physiological characteristics such as proline content, starch content and amino acid content (Sallam et al., 2018)

The capacity of a genotype tolerate extreme temperature stress is strictly correlated with their capacity to scavenge ROS (Singh et al., 2020; Liu et al., 2021).

Among numerous abiotic stresses, low temperature stress disturbs plant growth and leads to considerable reduction in crop yields (Wani et al., 2016; Xie et al., 2020). Cold stress straightaway prevents metabolic responses and indirectly deteriorates the osmotic disparity, which decreases the elucidation of the determined inherent ability of plants, in comparison to heat stress. Low temperature is characterized as chilling stress ( $<20^{\circ}\text{C}$ ) or freezing stress ( $<0^{\circ}\text{C}$ ), together disturb plant growth via various phenomena, depending on the temperature range whereas the quantity enzymatic rejoinders and membrane transport operations is reduced by chilling stress, freezing stress leads in creation of ice quartzes and membrane injury (Wang et al., 2020). By quantifying osmotic resistance, the infra-red imaging technique determines the low temperature resistant plants. Cold tolerance is related with physiological and biochemical modifications leading to transformed gene expression, bio membrane lipid composition and buildup of small molecules and can be quantified by imaging techniques such as MRI, PET and infra-red imaging.

## 4. High Throughput Phenotyping (HTP) data analysis and management

The large quantity and array of data attained with different techniques, its handling is the major challenge in phenotyping (Krajewski et al., 2015). HTP involves highly efficient computing and IT to measure as several parameters as possible. Because the data points generated are enormous and can't be managed manually, advanced approaches of data analysis are necessary from the initiation of raw data analysis till the results are validated (Kumar et al., 2020).

In conducting quantitation and data processing, the key bottleneck is the absence of elastic algorithms. Therefore, a high-throughput image analysis framework has been built



to solve logistical and scalability problems, such as the bisque system from the Center for Bioimage Informatics, UC Santa Barbara (<http://www.bioimage.ucsb.edu/>). Billiau *et al.* (2012) have developed a “phenotyper” database on the principle of data warehouse to manage vast volumes of data from phenomics. Successful

application of HTP platforms for use in drought, salinity and heat/ cold stress tolerance in plants, particularly cereal crops need capacity infrastructure for data management and analysis. (Shakoor *et al.*, 2017; Li *et al.*, 2021). The application of HTP for abiotic stress resilience in various cereal crops has been summarized (Table 1).

**Table 1.** Summary of Applications of High-throughput Phenotyping for Abiotic Stress Resilience in Cereal Crops

Plant species	Type of stress	Instrument/HTP method	Traits measured	Number of genotypes studied	References
Rice	Salt stress	RGB imaging (multiple views using LemnaTech	Shoot biomass, shoot ion concentration and shoot senescence	2 Indica rice cultivars (IR64 and Fatmawati)	Hairmansis <i>et al.</i> , 2014
	Drought stress	RGB imaging (multiple views using LemnaTech	Canopy temperature, Crop biomass, crop greenness, green projected area, plant area/convex hull ratio	533 rice accessions	Yang <i>et al.</i> , 2015
	Salt stress	RGB imaging (multiple views using LemnaTech	Loci detection for salinity tolerance, relative growth rate, transpirational rate and transpiration use efficiency	2 rice populations Aus (257 accessions) and Indica (297 accessions)	Tamimi <i>et al.</i> , 2016
	Drought stress	RGB imaging fluorescence using LemnaTech	Projected plant area, plant color, convex hull area, plant water content, plant temperature and eccentricity	20 wild type plants, 20 mutant ( <i>OshyB</i> ) plants	Kim <i>et al.</i> , 2020
	Salt stress	Thermal imaging using FLIR-SC-620	Leaf temperature, RWC, stomatal conductance and performance index	Randomly selected rice seeds from which 26 randomly selected leaves were studied, 6 for RWC 20 for stomatal conductance	Siddiqui <i>et al.</i> , 2014
Barley	Salt stress	RGB imaging (multiple views using plant accelerator	Shoot biomass, shoot ion concentration	377 barley accessions	Saade <i>et al.</i> , 2020
	Drought stress	RGB imaging (multiple views), Hyperspectral imaging using LemnaTech	Leaf length, leaf water content, biomass accumulation	18 genotypes	Chen <i>et al.</i> 2014
	Drought stress	RGB imaging (multiple views using LemnaTech	Plant biomass, projected shoot area, relative growth rate, caliper length, height, tiller number, plant height and water use efficiency	47 barley genotypes	Honsdorf <i>et al.</i> , 2014
	Drought stress	Fluorescence imaging	Plant architecture, yield related traits, convex hull geometry and chlorophyll intensity.	95 RILs and 2 parental genotypes	Mickolajczak <i>et al.</i> , 2020
	Oxidative stress	Fluorescence imaging	Seminal root length, leaf necrosis, leaf classical viability and relative root length	11 barley genotypes	Wang <i>et al.</i> , 2019
Maize	Drought stress	Hyperspectral imaging using PHENOVISION	Root characteristics, anthesis silking interval, drought tolerance index and yield	400 genotypes	Mertens <i>et al.</i> , 2021



	Cold stress	RGB imaging (multiple views using plant CV software)	Shoot height, shoot area, leaf necrosis	40 maize genotypes	Enders <i>et al.</i> , 2019
	Drought stress	Unmanned ariel vehicle imagery	Green leaf area index (GLAI) Dynamics	400 genotypes	Blancon <i>et al.</i> , 2019
	Drought stress	Terrestrial lidar using FARO Focus <sup>3D</sup> X120 laser scanner	Plant height plant area index, projected leaf area	20 maize varieties	Su et al.,2019
	Cold stress	Hyperspectral imaging	Spectral reflectance values and effective wave lengths of plants	60 genotypes	Xie et al.,2017
	Water stress	Thermal imaging using Midas 320L infrared camera	Canopy temperature crop water stress index chlorophyll index NDVI, biomass and yield	92 genotypes	Romano <i>et al.</i> , 2011
Wheat	Salt stress	Hyperspectral imaging using LemnaTech	Na <sup>+</sup> ion uptake, K/Na <sup>+</sup> ratio, root biomass and harvest index	4 wheat genotypes	Moghimi <i>et al.</i> ,2018
	Drought stress	Thermo-imaging using self-construction semiautomated	MSALR Transcript level, ALR Enzyme activities, shoot surface area and shoot biomass	3 transgenic wheat genotypes 1779 immature embryos	Feher juhasz <i>et al.</i> , 2014
	Salt stress	Plant Eye 3D Laser scanner	Plant height, number of leaves, total leaf area and fresh biomass	80 plants	Maphosa et al., 2016
	Drought and heat stress	X-ray computed tomography	Seed shriveling, grain deformation, total seed weight/ear, seed number/ear, single seed weight, seed shape and seed surface area	315 bread wheat accessions	Schimdt <i>et al.</i> , 2020
	Drought stress	Hyperspectral sensing aerial based multispectral sensing	Water band index, plant biomass	32 wheat genotypes	Yu <i>et al.</i> ,2020
	Drought stress	Unmanned ariel vehicle imagery	Leaf chlorophyll content, leaf rolling, dry biomass, NDVI and QTL for NDVI	248 elite durum wheat accessions	Condorelli <i>et al.</i> ,2018
Sorghum	Drought stress	Unmanned ariel vehicle imagery	Stay green values, NDVI values, values, leaf senescence and canopy traits	427 hybrids	Liedtke et al., 2020
	Drought stress	Imaging box with two digital cameras using Grow SCREEN Rhizo, Win RHIZO Pro	Nodal root angle	976 BC-NAM progenies	Joshi <i>et al.</i> , 2017

## 5. Conclusion

To tackle the challenges of global climate change, development of climate-smart germplasm would be a priority to cope up with the abiotic stresses. High-throughput phenotyping for abiotic stress coupled with the advanced genotyping technologies and other phenomic technologies like transcriptomics and metabolomics have accelerated the process of developing stress resilient crops and thus cater the needs of global food challenge with respect to the changing climatic conditions and increasing population.

## Conflict of Interest

Authors declare that they have no conflict of interest.

## Ethical Compliance Statement

NA

## Author Contribution

Conceptualization: SHW, ZAD; Initial Draft: NUI, SHW, GA; Critical review and finalization: AL, AW, SHW, ZAD



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