Agri Articles
(e-Magazine for Agricultural Articles)
Volume: 05, Issue: 06 (NOV-DEC, 2025)
Available online at http://www.agriarticles.com

Role of High Throughput Phenotyping in Crop Improvement

*R. Muthuvijayaragavan[1] and E. Murugan[2]
[1]Senior Research Fellow, Department of Plant Biotechnology, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India - 641003
[2]Professor (Plant Breeding and Genetics) and Head, Agricultural Research Station, TNAU, Paramakudi, Tamil Nadu, India- 623707
*Corresponding Author's email: muthu.ragavan@gmail.com

The world population is expected to reach approximately 9 to 10 billion by 2050; therefore, a gain of around 25–70% above present-day production levels will be required to meet these burgeoning population demands (Hunter et al. 2017). Further, various biotic and abiotic factors cause adverse environmental conditions or stress for crop plants, resulting in a significant reduction in their yields. This significant reduction in crop yield due to stress can jeopardize global food security (Strange and Scott 2005). Enhancement of crop yields is an ever-changing global challenge for plant breeders, entomologists, pathologists, physiologists and farmers. Hence, an in-depth understanding of plant stress is pivotal for improving yield protection for sustainable production systems (Pessarakli 2019). Plant scientists rely on crop phenotyping for precise and reliable trait collection and utilization of genetic resources and tools to accomplish their research goals.

Phenotyping

Plant phenotyping is defined as the comprehensive assessment of complex traits of plants such as development, growth, resistance, tolerance, physiology, architecture, yield, ecology, and the elementary measurement of individual quantitative parameters that form the foundation for complex trait assessment (Li *et al.* 2014). Breeding programs generally aim to phenotype large populations for numerous traits throughout the crop cycle (Sandhu *et al.* 2021). This phenotyping challenge is further aggravated by the need to sample at multiple environment with replicated trials. Traditional phenotyping is very costly, laborious, destructive, and could decrease the significance or preciseness of the results. The development of automated, high throughput phenotyping (HTP) systems merged with artificial intelligence has largely overcome the problems linked with the contemporary state-of-the-art crop stress phenotyping. HTP has offered great potential for non-destructive and effective field-based plant phenotyping. Manual, semi-autonomous or autonomous platforms furnished with single or multiple sensors record temporal and spatial data, resulting in large amounts of data for storage and analysis (Kaur *et al.* 2021; Sandhu *et al.* 2021b). For the analysis and interpretation of these massive datasets, machine learning (ML) and its subtypes, i.e. deep learning (DL) approaches, are utilized (Sandhu *et al.* 2021a).

High-throughput phenotyping

High-throughput phenotyping (HTP) in plant breeding uses automated systems with sensors and imaging to rapidly and accurately collect trait data from large plant populations, replacing slow, manual methods. This allows for faster selection of improved crop varieties with traits like yield, stress tolerance, and disease resistance by enabling breeders to evaluate more plants with greater precision.

Principle

- **Automated platforms**: HTP utilizes automated platforms, such as those on drones, that can move across fields.
- **Advanced sensors:** These platforms are equipped with a variety of sensors, including RGB cameras, hyperspectral and multispectral sensors, and near-infrared sensors.
- **Data collection:** These systems can capture detailed data on plant characteristics, including canopy height, biomass, and coverage, often from a non-destructive viewpoint.
- **Data analysis**: The collected data is processed using advanced software, sometimes incorporating AI and machine learning, to analyze traits like stress response, disease resistance, and yield.

Key benefits for plant breeding

- **Accelerated progress:** HTP significantly speeds up the breeding cycle by allowing breeders to evaluate more genotypes than possible with traditional methods.
- **Increased accuracy:** By providing more precise and objective data, HTP enhances the accuracy of selecting the best performing plants.
- **Evaluation of difficult traits:** It enables the measurement of traits that are hard to quantify manually, such as subtle variations in plant morphology or internal quality attributes.
- **Sustainability:** HTP contributes to the development of more resilient crops that require fewer resources like water and fertilizer.
- **Better data management:** The resulting digital data can be easily stored, shared, and re-analyzed, improving the reproducibility of research findings and facilitating collaboration.

Phenotyping Platforms

The area of plant stress phenotyping is steadily progressing, with destructive, low throughput phenotyping protocols/methods being substituted by non-invasive high-throughput methods (Barbedo 2019). Expeditious developments in non-invasive affordable sensors and imaging techniques and tools over the decades have transformed plant phenomics. Moreover, these developments have brought harmony between the sensors, imaging techniques and analytical tools. This consonance has led to the development of one-piece compact imaging platforms for HTP studies. Several HTP platforms exist and are presently employed to phenotype different biotic and abiotic stress-associated traits in various crops (Table 1).

Table 1. Details of some selected imaging platforms used for trait phenotyping for biotic and abiotic stress and other morphological traits in crops (Gill et al., 2022)

Platform	Traits recorded	Crop	References
A. Biotic and abiotic stresses			
PHENOPSIS	Plant responses to water stress	Arabidopsis (Arabidopsis thaliana)	Granier *et al.* (2006)
PHENODYN	Soil water status (drought scenarios), leaf elongation rate, and micrometeorological variable	Rice (Oryza sativa) and maize (Zea mays)	Sadok *et al.* (2007)
Field monitoring support system	Occurrence of the rice bug in the field	Rice (Oryza sativa)	Fukatsu *et al.* ((2012)
LemnaTec 3D scanalyzer system	Salinity tolerance traits	Rice (Oryza sativa)	Hairmansis *et al.* (2014)
RADIX	Root and shoot related traits under control and as well as stress conditions	Maize (Zea mays)	Le Marié *et al.* (2016)
PhenoImage	Plant responses to water stress	Wheat (Triticum aestivum), sorghum (Sorghum bicolor)	Zhu *et al.* (2021)

B. Morphological and physiological traits (recorded under unstressed conditions)			
High-throughput rice phenotyping facility (HRPF)	Agronomic traits	Rice (Oryza sativa)	Yang *et al.* (2014)
Zeppelin NT aircraft	Leaf area index, leaf biomass, early vigour, plant height	Maize (Zea mays)	Liebisch *et al.* (2015)
Phenocart	Morphological traits	Wheat (Triticum aestivum)	Crain *et al.* (2016)
Phenobot 1.0	Biomass-related traits	Sorghum (Sorghum bicolor)	Salas Fernandez *et al.* (2017)
PhenoRoots	Root related traits	Cotton (Gossypium hirsutum L.)	Martins *et al.* (2020)

Data has also been recorded in an automated and high throughput manner for root and shoot related traits, leaf traits, plant height, plant biomass, early vigor, radiation use efficiency, photosynthesis in different plant species such as rice, wheat, maize, sorghum (Sorghum bicolor L.), cotton (*Gossypium hirsutum* L.), Arabidopsis, Brachypodium (*Brachypodium distachyon* L.), rapeseed (*Brassica napus* L.), and barley (*Hordeum vulgare* L.) among others using different phenotyping platforms such as RootReader3D (Clark *et al.* 2011), GROWSCREEN-Rhizo (Nagel *et al.*, 2012), Zeppelin NT aircraft (Liebisch *et al.* 2015), Phenocart (Crain *et al.* 2016), Phenovator (Flood *et al.* 2016), PHENOARCH (Brichet *et al.* 2017), Field Scanalyzer (Virlet *et al.* 2016), CropQuant (Zhou *et al.* 2017), and MVS-Pheno (Wu *et al.* 2020). These platforms have the potential to be utilized for HTP of traits associated with stress tolerance/resistance in different crops.

Imaging Techniques

Phenomics has been extensively used to monitor diseases, pest infestations, drought stress, nutrient status, growth, presence of weeds and yield under stresses and normal conditions in different crop species (Barbedo 2019). Technological advancement has made novel imaging techniques available for use in HTP. Imaging techniques range from handheld mobile phones to highly flexible drone imaging using unmanned aerial vehicles (UAV). UAVs offer a platform that rapidly records data using different imaging sensors over large areas and potentially gives images with high spatial resolution. UAV can be used to cover plots or multiple fields in one flight, but their limited battery capacity reduces their utility for very large-scale HTP. Numerous studies have been conducted where drone imaging is used to assess biotic and abiotic stresses in plants. Some of these are provided in Table 2.

Table 2. Different imaging techniques used for plant stress phenotyping. (Gill *et al.*, 2022)

S.No.	Imaging techniques	Remarks	Disadvantage	References
1.	Satellite Imagery	Easily covers very large areas The data can easily help predict droughts and epiphytotics as very large areas can be covered at the same time	High cost associated with constructing and launching satellites.	Wheat yield (Fieuzal *et al.* 2020), cotton yield (He and Mostovoy 2019)
2.	Mobile Cameras/Imaging	Convenient, portable Rapid and no operational cost	Taking pictures of each plant in the field is not practical	Iron deficiency chlorosis severity in soybean(Naik *et al.* 2017), salinity stress tolerance (Awlia *et al.* 2016)

3.	UAV Imaging	UAVs (Unmanned Aerial Vehicles) can cover large areas, Very economical, High resolution data and Easy to operate with low learning curve	They cannot cover as much spatial area as satellites because of their limited battery capacity and flight height.	Grain yield wheat (Hassan *et al.* 2019), Plant Nitrogen content (Camino *et al.* 2018), wheat biomass (Yue *et al.* 2017), maize yield (Maresma *et al.* 2016),
4.	Imaging using robots	Most advanced technique and Highly efficient as it provides human-like manual phenotyping results	Still an evolving technique Much work required for workflow and data management	Plant architecture (Qiu *et al.* 2019), Heat stress and stripe rust resistance wheat (Zhang *et al.* 2019),

Spectral Indices for Plant Stress Phenotyping

Images captured by the aforementioned techniques need to be decoded, and spectral indices (SIs) are used to assess the information in these images (Hunt *et al.* 2013). SIs involve conducting various sets of operations on different spectral layers of an image. These sets of operations include some mathematical calculations and combination of spectral reflectance from two or more wavelengths. The result of this mathematical combination generates a number that denotes the relative abundance of the feature of interest (Jackson and Huete 1991).

Conclusion

Plant stress phenotyping is an important parameter for predicting crop losses caused by various biotic and abiotic stresses. It can be used to identify superior disease resistant and stress-tolerant genotypes as well as to assess disease management decisions. The phenotypic parameters include not only morphological data, but also a large number of physiological and biochemical data, as well as deeper mechanistic data, allowing scientists to identify and predict heritable traits through controlled phenotypic and genotypic studies. Current methods for stress severity phenotyping are used at various scales, such as the number of plants affected or exact counts of lesion numbers, or estimates of the severity or surface area affected by a particular biotic/abiotic stress at the canopy of single plant and field levels. More research is needed in the future to improve UAV-based sensing for plant phenotyping. High-performance and low cost UAVs should be introduced in future studies. For long-term and large-field plant phenotyping, high-performance UAVs with high flight stability, precision, long flight duration, and heavy payload are required. Unlike ground-based phenotyping, UAV-based phenotyping is afflicted by a serious issue: the safety of the UAV and its sensors. With the need to double food production to feed the projected population of 10 billion by 2050, high throughput plant phenotyping technology are the other new breeding innovations which are essential tools to ensure food security in future.

References

1. Awlia M, Nigro A, Fajkus J. (2016) High-throughput non-destructive phenotyping of traits that contribute to salinity tolerance in Arabidopsis thaliana. Front Plant Sci 7:1414.
2. Barbedo JGA (2019) A review on the use of unmanned aerial vehicles and imaging sensors for monitoring and assessing plant stresses. Drones 3:40.
3. Brichet N, Fournier C, Turc O. (2017) A robot-assisted imaging pipeline for tracking the growths of maize ear and silks in a high-throughput phenotyping platform. Plant Methods 13:96.
4. Brichet N, Fournier C, Turc O. (2017) A robot-assisted imaging pipeline for tracking the growths of maize ear and silks in a high-throughput phenotyping platform. Plant Methods 13:96.

5. Camino C, González-Dugo V, Hernández P et al (2018) Improved nitrogen retrievals with airborne-derived fluorescence and plant traits quantified from VNIR-SWIR hyperspectral imagery in the context of precision agriculture. Int J Appl Earth Obs Geoinf 70:105–117.
6. Clark RT, MacCurdy RB, Jung JK. (2011) Three-dimensional root phenotyping with a novel imaging and software platform. Plant Physiol 156:455–465.
7. Crain JL, Wei Y, Barker J. (2016) Development and deployment of a portable field phenotyping platform. Crop Sci 56:965–975.
8. Fieuzal R, Bustillo V, Collado D, Dedieu G (2020) Combined use of multi-temporal landsat-8 and sentinel-2 images for wheat yield estimates at the intra-plot spatial scale. Agronomy 10:327.
9. Flood PJ, Kruijer W, Schnabel SK. (2016) Phenomics for photosynthesis, growth and reflectance in Arabidopsis thaliana reveals circadian and long-term fluctuations in heritability. Plant Methods 12:14.
10. Fukatsu T, Watanabe T, Hu H .(2012) Field monitoring support system for the occurrence of Leptocorisa chinensis Dallas (Hemiptera: Alydidae) using synthetic attractants, field servers, and image analysis. Comput Electron Agric 80:8–16.
11. Gill T, Gill SK, Saini DK, Chopra Y, de Koff JP, Sandhu KS.(2022) A Comprehensive Review of High Throughput Phenotyping and Machine Learning for Plant Stress Phenotyping. Phenomics. 4;2(3):156-183.
12. Granier C, Aguirrezabal L, Chenu K. (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.
13. Hassan MA, Yang M, Rasheed A. (2019) A rapid monitoring of NDVI across the wheat growth cycle for grain yield prediction using a multi-spectral UAV platform. Plant Sci 282:95–103.
14. Hairmansis A, Berger B, Tester M, Roy SJ (2014) Image-based phenotyping for non-destructive screening of different salinity tolerance traits in rice. Rice (NY) 7:16.
15. He L, Mostovoy G (2019) Cotton yield estimate using sentinel-2 data and an ecosystem model over the southern US. Remote Sens (basel) 11:2000.
16. Hunt ER, Doraiswamy PC, McMurtrey JE. (2013) A visible band index for remote sensing leaf chlorophyll content at the canopy scale. Int J Appl Earth Obs Geoinf 21:103–112.
17. Hunter MC, Smith RG, Schipanski ME. Agriculture in 2050: recalibrating targets for sustainable intensification. Bioscience. 2017;67:386–391.
18. Jackson RD, Huete AR (1991) Interpreting vegetation indices. Prev Vet Med 11:185–200.
19. Kaur B, Sandhu KS, Kamal R. (2021) Omics for the improvement of abiotic, biotic, and agronomic traits in major cereal crops: applications, challenges, and prospects. Plants 10:1989.
20. Kumawat, K.R. and Sharma N.K. (2018). Cisgenesis: an executable approach in crop improvement. Popular Kheti, 6(1): 110-112.
21. Le Marié C, Kirchgessner N, Flütsch P. (2016) RADIX: rhizoslide platform allowing high throughput digital image analysis of root system expansion. Plant Methods 12:40.
22. Liebisch F, Kirchgessner N, Schneider D. (2015) Remote, aerial phenotyping of maize traits with a mobile multi-sensor approach. Plant Methods 11:9.
23. Li L, Zhang Q, Huang D (2014) A review of imaging techniques for plant phenotyping. Sensors 14:20078–20111.
24. Maresma Á, Ariza M, Martínez E. (2016) Analysis of vegetation indices to determine nitrogen application and yield prediction in maize (Zea mays L.) from a standard UAV service. Remote Sens (basel) 8:973.
25. Martins SM, de Brito GG, da Gonçalves WC. (2020) PhenoRoots: an inexpensive non-invasive phenotyping system to assess the variability of the root system architecture. Sci Agric.

26. Nagel KA, Putz A, Gilmer F. (2012) GROWSCREEN-Rhizo is a novel phenotyping robot enabling simultaneous measurements of root and shoot growth for plants grown in soil-filled rhizotrons. Funct Plant Biol 39:891.
27. Naik HS, Zhang J, Lofquist A. (2017) A real-time phenotyping framework using machine learning for plant stress severity rating in soybean. Plant Methods 13:23.
28. Pessarakli M. Handbook of plant and crop stress. 4. Boca Raton, FL: CRC Press, Taylor & Francis Group; 2019.
29. Qiu Q, Sun N, Bai H. (2019) Field-based high-throughput phenotyping for maize plant using 3D LiDAR point cloud generated with a "Phenomobile." Front Plant Sci 10:554.
30. Sadok W, Naudin P, Boussuge B. (2007) Leaf growth rate per unit thermal time follows QTL-dependent daily patterns in hundreds of maize lines under naturally fluctuating conditions. Plant Cell Environ 30:135–146.
31. Salas Fernandez MG, Bao Y, Tang L, Schnable PS (2017) A highthroughput, field-based phenotyping technology for tall biomass crops. Plant Physiol 174:2008–2022.
32. Sandhu KS, Mihalyov PD, Lewien MJ, Pumphrey MO, Carter AH (2021b) Genomic selection and genome-wide association studies for grain protein content stability in a nested association mapping population of wheat. Agronomy 11(12):2528
33. Sandhu KS, Mihalyov PD, Lewien MJ. (2021). Combining genomic and phenomic information for predicting grain protein content and grain yield in spring wheat. Front Plant Sci.12:613300.
34. Sandhu KS, Lozada DN, Zhang Z. (2020) Deep learning for predicting complex traits in spring wheat breeding program. Front Plant Sci. 11:613325.
35. Strange RN, Scott PR (2005) Plant disease: a threat to global food security. Annu Rev Phytopathol 43:83–116.
36. Virlet N, Sabermanesh K, Sadeghi-Tehran P, Hawkesford MJ (2016) Field Scanalyzer: an automated robotic field phenotyping platform for detailed crop monitoring. Funct Plant Biol 44:143–153.
37. Wu S, Wen W, Wang y. (2020) MVS-pheno: a portable and lowcost phenotyping platform for maize shoots using multiview stereo 3D reconstruction. Plant Phenomics 2020:1848437.
38. Yang W, Guo Z, Huang C. (2014) Combining high-throughput phenotyping and genome-wide association studies to reveal natural genetic variation in rice. Nat Commun 5:5087.
39. Yue J, Yang G, Li C. (2017) Estimation of winter wheat aboveground biomass using unmanned aerial vehicle-based snapshot hyperspectral sensor and crop height improved models. Remote Sens (basel) 9:708.
40. Zhang C, Pumphrey MO, Zhou J. (2019) Development of an automated high- throughput phenotyping system for wheat evaluation in a controlled environment. Trans ASABE 62:61–74.
41. Zhou J, Reynolds D, Websdale D. (2017) CropQuant: an automated and scalable field phenotyping platform for crop monitoring and trait measurements to facilitate breeding and digital agriculture.
42. Zhu F, Saluja M, Dharni JS. (2021) PhenoImage : An open-source graphical user interface for plant image analysis. Plant Phenome J.