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HIGH THROUGHPUT PHENOTYPING WITH IMAGING SENSORS

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ABSTRACT

Conventional phenotyping methods made it challenging to capture physiological and biochemical phenotypes at the level of basic plant mechanisms that reveal patterns of genetics and biology. In order to alleviate this bottleneck, since 2000, a variety of phenotyping platforms have been developed, which are now standard tools in commercial or research teams. An image-based, high-throughput phenotyping platform (HT3P) is defined as a platform that can take the image of hundreds of plants daily. "HT3P" can collect massive amounts of phenotypic data from hundreds of plants every day. This is a powerful and novel tool that monitors and quantifies crop growth and production-related phenotypic traits fast, non-destructive, and high-throughput manner. Phenotype arises from interactions between genotype and environment, and the essence is the temporal expression of the plants' gene map in distinct geographic regions. Phenotyping applies specific methods and protocols to measure morphological, physiological functional traits, structural traits, and component content traits of cells, organs, canopy, tissues, whole plants, or even populations. Genomics-assisted breeding (GAB) through genomic approaches of quantitative trait loci (QTL) mapping, genomic selection (GS), marker-assisted selection (MAS), and genome-wide association studies (GWAS), thereby assisting crop growers in adapting to changing climate conditions and market demand for yield.

INTRODUCTION

Phenotyping and genotyping are significant components of analytical breeding. Genotyping technologies have made it quicker, while phenotyping has lagged and is still limiting (Kumar, 2016). Recently, image-based sensors have emerged as a new way of accurately phenotyping a large set of genotypes in high throughput manner. Non-invasive sensors and advanced computational platforms help in extracting the plant features. The depth of component phenotypic traits and the Spatio-temporal dynamic phenotypic data generated in phenomics are enormous and unparallel to conventional phenotyping. Plant phenotyping is the comprehensive assessment to measure plant traits related to growth, yield with precision. Major plant traits are related to root morphology, biomass, yield-related characteristics, and abiotic stress response. A team of biological sciences, computer science, statistics, and engineering experts is required to phenotype precisely. Nowadays, high throughput phenotyping platforms have been deployed in control of environmental conditions. Under control conditions, the Environment effect is also nullified. So, the real genetic potential of genotype is expressed in terms of phenotypes. Automated phenotyping platforms involves

1. Non-Invasive Sensors,
2. Automated Data Processing to Obtain Phenotypic Traits of Interest,
3. Robotized Delivery of Plants to Sensors Or Vice Versa,
4. Robotized Plant Culturing, And
5. Automated Analysis of Processed Data in A Data Management Pipeline.

IMAGE-BASED PHENOTYPING

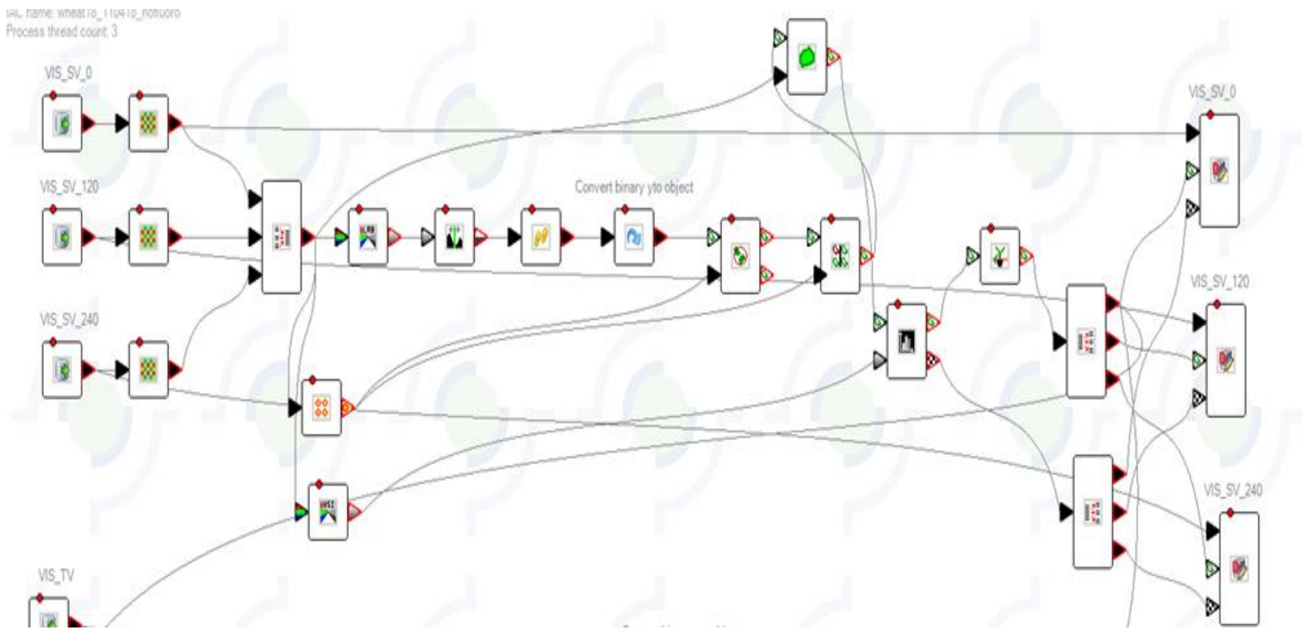
Novel imaging technologies played a crucial role in the precise measurement of plant features. Genomic data and environmental data integration need to be done to interpret experimental metadata and the measured phenotype. These imaging techniques help in measuring growth rate along with environmental interactions. Imaging platforms are not just for 'taking pictures. Imaging aims to record the data at regular intervals and interpret the data with the nature of the sensor and its correlation with plant features. Near infra-red (NIR)-to record the dynamic shift in plant water content while hyperspectral sensors like short wave infra-red (SWIR) and very near to infra-red (VNIR) was used to record biochemical parameters in high throughput manner.

PHENOTYPING OF PLANT ARCHITECTURE THROUGH RGB SENSOR

Visual digital images help mimic human perception and provide the information in digital form, which we could see, but the expression in data form becomes difficult. Visual sensitivity range starts from visible bands of light (400–750nm). In-plant phenomics facility plants are grown in greenhouses, on conveyors with radiofrequency identifiable (RFID) cars. For recording data, pots are being called in the imaging area. RGB chamber has LED tube lights, which turn on almost 10 minutes before the image recording so that the light gets saturated in the chamber. According to the plant stage, the height of the imaging platform's lifting and turning device is set up to get a total view of the pot both from side view and top view. Generally, images are taken from three angles, i.e., 0, 120, and 240 degrees along with the top view. These images are saved in database.

IMAGE ANALYSIS

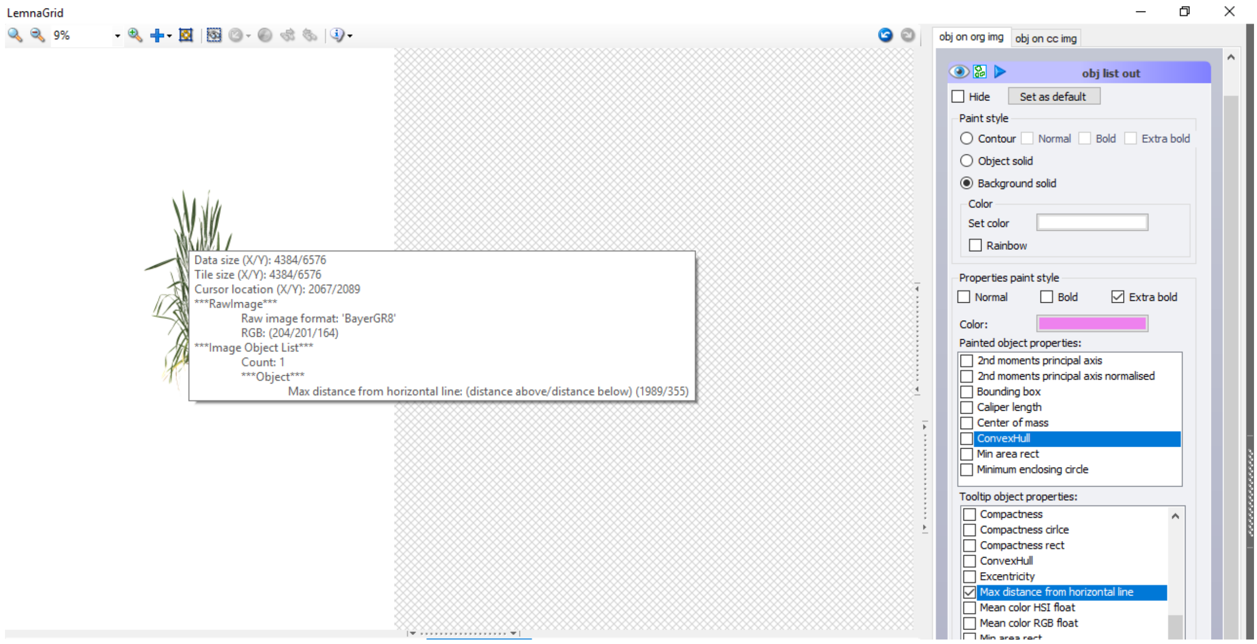
Once acquired, images are stored in the database. Afterward, Image analysis software like ENVI, Image J, Matlab, and Lemna Grid is used to extract the features of our interest. In-plant phenomics facility, we used Lemna Grid software to analyzing images. The following image pipeline is used to extract features.


Process thread count: 3
VIS_SV_0
VIS_SV_120
VIS_SV_240
VIS_TV
Convert binary yto object
VIS_SV_0
VIS_SV_120
VIS_SV_240

SIGNIFICANT COMPONENTS OF PIPELINE

1. Database Reader
2. Demosaicing Module
3. Demuxing for Batch
4. Thresholding
5. Universal Converter
6. Image composition
7. Mean color property
8. Histogram
9. Writer label

Once the image is processed, we are completed. We get information on plant traits in the total plant area, height, Convex hull, caliper length, plant height, and different color classified details. This information is used to calculate growth throughout plant development. Besides this abiotic stress, effects can be explained based on the leaf senescence by separating the yellow and green areas of the leaf.



Features available through image analysis

IMAGE-BASED PHENOTYPING FOR PLANT WATER STATUS

Near-infrared imaging is used to screen the significant genotypes for plant water status. NIR range varies from 700–1700 nm. NIR-based imaging is based on the concept that leaves reflect a large proportion of incident radiation. So based on the reflectance pattern water status of the plant can be measured. Out of the whole range of NIR, the water band lies between 900 to 1500 nm range. Leaf and canopy architecture, such as leaf thickness, are the significant determinants of the reflectance pattern. For recording data, pots are being called in the imaging area. NIR chamber has unique yellow light-based lamps, which turn on almost 10 minutes before the image recording so that the light gets saturated in the chamber. According to the plant stage, the height of the imaging platform's lifting and turning device is set up to get a total view of the pot both from side view and top view. Generally, images are taken from side view and complete view. These images are saved in the database.

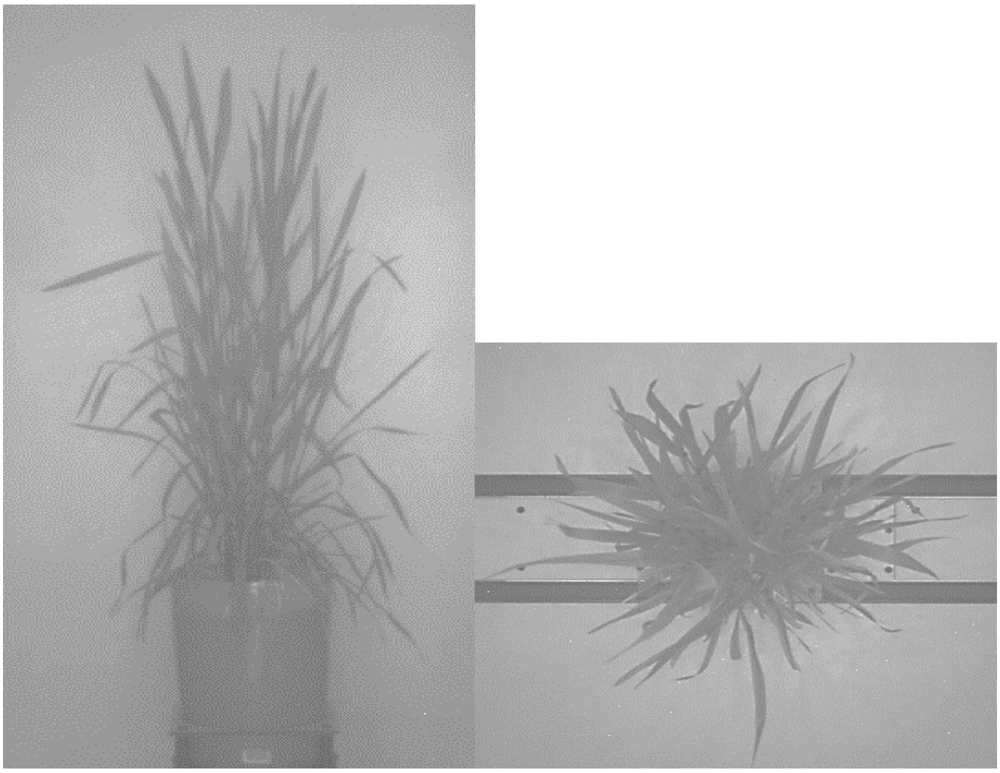
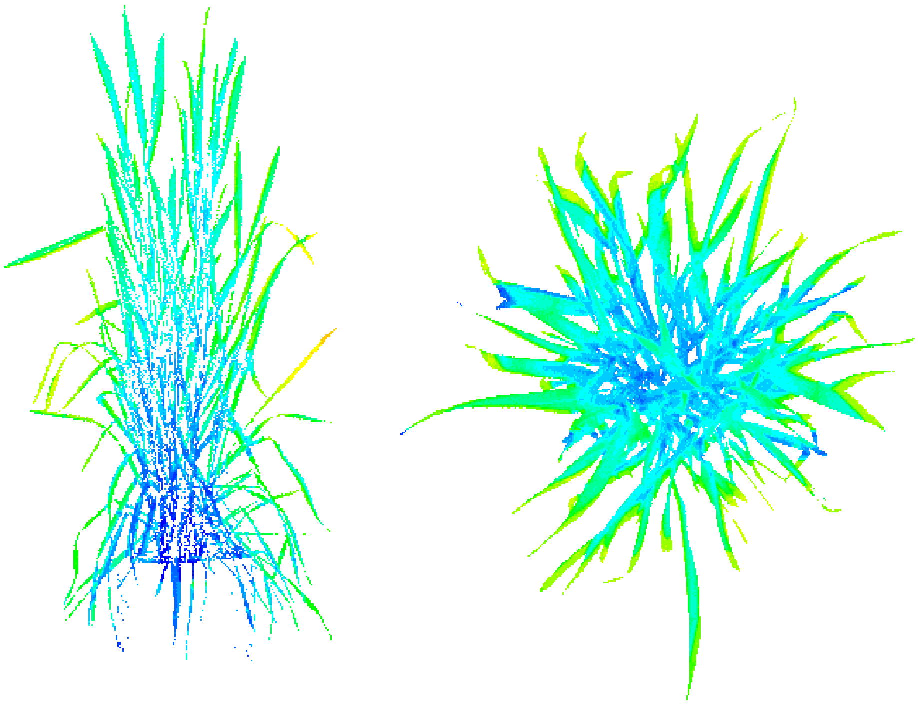


IMAGE ANALYSIS

Once acquired,images are stored in the database. Afterward, Image analysis software like ENVI, Image J, Matlab, and Lemna Grid is used to extract the features of our interest. In-plant phenomics facility, we used Lemna Grid software to analyzing images. Some of the examples of processed images are:



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Therefore, after image processing, these image-based features are converted to digital numbers. The mean grey value is used for comparing treatments.

IMAGE-BASED PHENOTYPING FOR CANOPY TEMPERATURE

IR thermography measures the temperature of the target by measuring the radiant thermal energy emitted by the target. Infrared is a type of electromagnetic radiation emitted by all objects that have a temperature to a greater or lesser degree. IR spectral region of 8 to 14 µm is typically used for thermal remote sensing. Thermal imaging allows for visualizing infra-red radiation, indicating an object as the temperature across the object's surface. Thermal imaging helps us to tap this state of the plant. It helps us find out the stomatal conductance variation among genotypes and indicates the root activity. Change in canopy temperature measures by thermal imaging help us understand the water status of the plants, i.e., both uptake and transpiration. This information is very much helpful and used in the selection criterion in breeding programs for drought resistance.

PROCESSED THERMAL IMAGE

CONCLUSION

The phenotype of a plant is the interaction between genotype and environment. The image-based phenotype is a big boon to obtain this information in high throughput manner and with precision. It records the data in no time and helps extract the data as and when we need it. So, with image-based phenotyping, we will get more and more screened material for drought tolerance.

REFERENCES

Kumar,S., Raju, D., Sahoo, R.N., and Chinnusamy, V. 2016. Phenomics: unlocking the hidden genetic variation for breaking the barriers in yield and stress tolerance. *Indian Journal of Plant Physiology* **21** (4), 409–419

Li, L., Zhang, Q and Huang D. 2014. A Review of Imaging Techniques for Plant Phenotyping (14): 20078-20114