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Phenomics: Approaches and Application in Improvement of Vegetable Crops

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Abstract

Increasing consumption of food, feed, fuel and to meet global food security needs for the rapidly growing human population, raise the necessity to breed high yielding crops that can adapt to the future climate changes, particularly in developing countries. To solve these global challenges, novel approaches are required to identify quantitative phenotypes and to explain the genetic basis of agriculturally important traits. These advances will facilitate the screening of germplasm with high performance characteristics in resource limited environments. High-throughput phenotyping platforms have also been developed that capture phenotype data from plants in a non-destructive manner. In this review, we discuss recent developments of high throughput plant phenotyping infrastructure including imaging techniques and corresponding principles for phenotype data analysis. Phenomics is a way of speeding up phenotyping with the help of high-tech imaging systems and computing power. It has been a practice in plant breeding for selecting the best genotype after studying phenotypic expression in different environmental conditions and also using them in hybridization programs, to develop new improved genotypes.

1. Introduction

Persistent food and feed supply needs, resources shortages, climate change and energy use are some of the challenges we face in our dependence on plants. Until 2050, crop production will have to double to meet the projected production demands of the global population. Demand for crop production is expected to grow 2.4% a year, but the average rate of increase in crop yield is only 1.3%. Moreover, production yields have stagnated in up to 40% of land under cereal production (Fischer and Edmeades, 2010). Genetic improvements in crop performance remain the key role in improving crop productivity, but the current rate of improvement cannot meet the needs of sustainability and food security.

Precise and accurate measurement of traits plays an important role in the genetic improvement of crop plants. Therefore, a lot of development has taken place in the area of phenomics in the recent past. Both forward and reverse phenomics have been evolved, which can help in identification of either the best genotype having the desirable traits or mechanism and genes that make a genotype the best. This includes development of high throughput non-invasive imaging technologies including colour imaging for biomass, plant structure, phenology and leaf health (chlorosis, necrosis); near infrared imaging for measuring tissue and soil water contents; far infrared imaging for canopy/ leaf temperature; fluorescence imaging for physiological state of photosynthetic machinery; and automated weighing and watering for water usage imposing drought/ salinity. These phenomics tools and techniques are paving the way in harnessing the potentiality of genomic resources in genetic improvement of crop plants.

These techniques have become much more advanced and have now entered the era of high throughput integrated phenotyping platforms to provide a solution to genomics-enabled improvement and address our need of precise and efficient phenotyping of crop plants.

Worldwide demand for crops is increasing rapidly due to rising global population, rising demand for biofuel and feed stocks and changing food preferences. Meeting future demand of agricultural production poses the greatest challenge to agricultural scientists and policy makers (Bruinsma, 2003) because demand for cereals, biofuels and feed stocks has already surpassed the current supply and is expected to rise further in the near future (Sticklen, 2007; Furbank *et al.,* 2009). Therefore, there is a competition among crops for arable land in order to increase their production. Rising global mean temperature by 0.8 °C since the 1850's, which is expected to increase further by 1.8–4 °C by the end of this century, will have further impact on agricultural production due to changing climate (Solomon *et al.,* 2007) and prevalence of abiotic stresses with more intensity and frequencies (Tester

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and Langridge, 2010). It has been estimated that in future average crop yields may decline across Africa and South Asia by 8% by the 2050's (Knox *et al.,* 2012). These declines in yields have been predicted about 17% in wheat, 5–16% in maize, 11–15% in sorghum, and 10% in millet across above regions under regimes of climate change (Wheeler and von Braun, 2013). Therefore, development of 'climate-smart' germplasm would be a priority to tackle these future challenges of climate change (Ziska and Bunce, 2007; Leakey *et al.,* 2009).

The use of conventional plant breeding methods has made substantial gain in crop yield worldwide. However, researchers are now observing that current breeding methods will not be sufficient to meet the projected future demand of foods (Sticklen, 2007; Furbank *et al.,* 2009; Tester and Langridge, 2010). Therefore, this has shifted our focus towards the use of genomics and gene technology advances for assisting the current breeding programs in order to increase grain yields. These developments are being utilized in trait discovery, genetic dissection of complex traits and discovery of associated genes and their deployment in varieties.

Although a large collection of germplasm of different crop species are available worldwide, phenotypic descriptions of these genome wide knockout collections are still limited. As a result, it restricted the use of genomic resources for identifying the allelic variation for a promising candidate gene in natural germplasm collection (Miyao *et al.,* 2007). The poor utilization of genomic resources could also be due to the lack of analysis of invisible traits and sometimes complex phenotypic effects of genetic modification. Hence, this science is one of the best in its kind and shares the following objectives.

- It can be used in laboratories and also in fields to analyse phenotypes in natural conditions as well as under controlled environment.
- Evaluation can be faster, and facilitates a more dynamic whole-of-life cycle measurement.
- The whole plant, pot or even entire field can be included in a single image.
- Analysis of a single image would allow quantification of several traits.
- Less dependent on periodic destructive assays.
- Improved the precision and accurate in recording the data.
- Reduce the need for replication in the field.

2. Importance of Plant Phenomics

By 2050, it is estimated that 9.1 billion people will populate the planet earth. And the need of high yielding varieties would give sustainable yields even under adverse and changing environmental conditions. Therefore, phenomics can act as a powerful tool to overcome one of humanity's greatest challenges: Hunger.

Biofuels are fuels such as ethanol and biodiesel that are produced from plant matter, and are called 'feedstocks'. Such crops could not only be eaten by people instead can be turned into fuel. These crops could compete with food crops for the best agricultural land. Hence, researchers are in need to make trials for alternative plant species as feedstock that can grow on less productive lands. But, then these crops will need to tolerate a wide range of environmental stresses, such as low water availability, salinity or low nutrient supplies, to be able to grow successfully in such 'marginal' lands. It was estimated that half of the increase in yield of the main agricultural crops has come through plant breeding processes (Kuhr *et al.,* 1985; Byerlee *et al.*, 1993).

Phenotyping can be a laborious process, taking many days, weeks or even months. The high-tech automated plant analysis systems of plant phenomics have speeded up traditional, time consuming methods of plant analysis. Phenomics is focused on coming up with practical solutions to problems that will affect us and our future. This technology can be used for study plants from the small scale to studying individual cells or leaves to up to the large scale of an entire ecosystem.

Phenomics technology allows plants, which are only one month old, move through a series of chambers, each one automatically taking different images and measurements. Within a few hours, the high-tech imaging and computing systems automatically measure the plants and the necessary data are given. The young plants with the characteristics the researcher looking for can be selected and grown on to mature for seed; thereby, the rest can be discarded. Figure 1 showing the general step involve in phenomics.

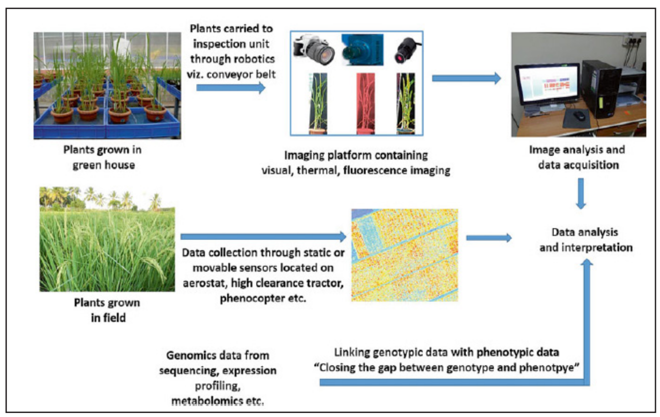

Figure 1: Steps involve approaches in phenomics

3. Origin of Plant Phenotyping

Plant phenotyping has been a part of crop and variety selection since the time of human civilization when humans selected the best individuals of a crop species for domestication. Subsequently it has become common practice in plant breeding for selecting the best genotype after studying phenotypic expression in different environmental conditions and also using them in hybridization programs in order to develop new improved genotypes (Fisher, 1925; Annicchiarico, 2002;

Pearson *et al.,* 2008). Ecologists used phenotyping to study phenotypic of genotypes during the middle of the 20th century and suggested the role of the genotype and environmental conditions in the expression of plant phenotypes under which it develops (Suzuki *et al.,* 1981). Subsequently, developments in ecology in relation to phenotyping are the trait-based approaches, in which phenotypic characteristics of a wider range of different species are evaluated either in the field (Reich *et al*., 1992) or under laboratory conditions (Grime and Hunt, 1975; Poorter *et al.,* 1990). They were used to derive different strategies by which the ecological niche of species could be described (Grime, 1979) and to analyze the interdependence of various traits (Wright *et al*., 2004).

4. Phenomics

The word 'phenome' refers to the phenotype as a whole (Soul, 1967) *i.e.,* expression of genome for a trait in a given environment while in phenomics we get high-dimensional phenotypic data on an organism at large scale. Actually phenomics is used as analogy to genomics. However it differs from genomics. In genomics, complete characterization of a genome is possible while in phenomics, complete characterization of phenome is difficult due to the change in the phenotypic expression of traits over the environmental conditions (Houle *et al.,* 2010).

5. Phenotype vs. Phenomics

Phenotype of a plant can be described on the basis of morphological, biochemical, physiological and molecular characteristics. Different parameters are measured to describe these characteristics. Johannsen (1911) has coined the terms 'genotype' and 'phenotype'. He demonstrated substantial variation in quantitative traits to which he called 'phenotypical' in genetically-identical material and thus proved that variation in a given observed traits is not controlled entirely by genetics. Therefore, use of statistical analysis has been suggested for identifying the differences among genotypes because phenotypic variation within a genotype can obscure phenotypic differences among genotypes. This leads to origin of 'pheno' word. After 1950, 'phenotyping' as a noun, 'to phenotype' as a verb and 'phenome' as the collective noun were introduced, which have been accepted scientifically and are being utilized commonly in literature.

6. Forward and Reverse Plant Phenomics

Plant phenomics is the study of plant growth, performance and composition. Figure 2 showing the use of forward and reverse phenomics in genetic improvement. Forward phenomics uses phenotyping tools to discriminate the useful germplasm having desirable traits among a collection of germplasm. This leads to identification of the 'best of the best' germplasm line or plant variety. Use of high-throughput, fully automated and low resolution followed by higher-resolution screening methods have accelerated plant breeding cycle by screening a large number of plants at seedling stage. Thus interesting traits can be identified rapidly at early stage and there is no need to grow plants up to the maturity stage in field. Now it is possible in forward phenomics to screen thousands of plants in pots running along a conveyor belt, and travelling through a room containing automated imaging systems such as infra-red or 3D cameras. The pots are labelled with barcodes or radio tags, so that the system can identify which pots contain plants with interesting traits. The selected plants can then be grown up to produce seed for further analysis and breeding.

The reverse phenomics is used where the best of the best genotypes having desirable trait(s) is already known. Now through reverse phenomics, traits shown to be of value to reveal mechanistic understanding are dissected in details and subsequently the identified mechanisms are exploited in new approaches. Thus in reverse phenomics, we discover mechanisms which make 'best' varieties the best. This can involve reduction of a physiological trait to biochemical or biophysical processes and ultimately a gene or genes. For example, in case of drought tolerance, researchers try to work out the mechanisms underlying the drought tolerance and find out the gene or genes that are responsible for it. These genes are screened in germplasm or the gene can be bred into new varieties.

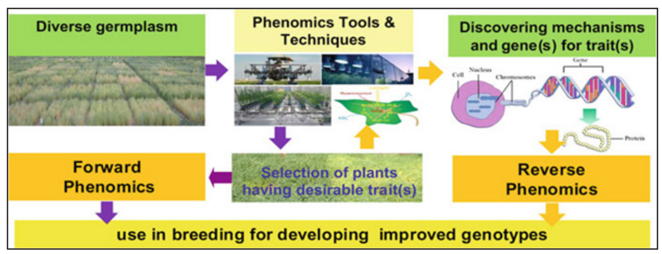

Figure 2: Forward and Reverse Plant Phenomics

7. Imaging Technology

In plant science, visible light imaging has been broadly adopted due to its low cost and simplicity. Using this imaging system, with a similar wavelength (ranging from 400 to 700 nm) perception as the human eye, two-dimensional (2D) images can be used to analyze numerous phenotypic characteristics and to record the changes in plant's biomass (Tackenberg, 2007; Bylesjo *et al*., 2008; Duan *et al.,* 2011; Golzarian *et al.,* 2011). To spread the spatial and volumetric information of phenotype images, three-dimensional (3D) imaging approaches have been developed, which could provide more accurate estimations of the morphological features (Clark *et al.,* 2011; Paproki *et al.,* 2012). Phenomics borrows imaging techniques from medicine to allow researchers to study the inner workings of leaves, roots or whole plants (Figure 3).

8. Three-Dimensional (3D) Imaging

Digital photos of the top and sides of plants are combined into a 3D image. Measurements that can be taken using a 3D image include: shoot mass, leaf number, shape and angle, leaf colour and leaf health. Technically, pots of plants move on a conveyor belt through an imaging chamber and 3D models are automatically generated by a computer program. Obtained images are transferred to the software and required editions as colour improving and optimization are made. Digital images have advantages such as simple recording, transmitting, and storing in a database. However, algorithms are necessary to gather and analyze the huge amount of data (Tsaftaris *et al.,* 2009).

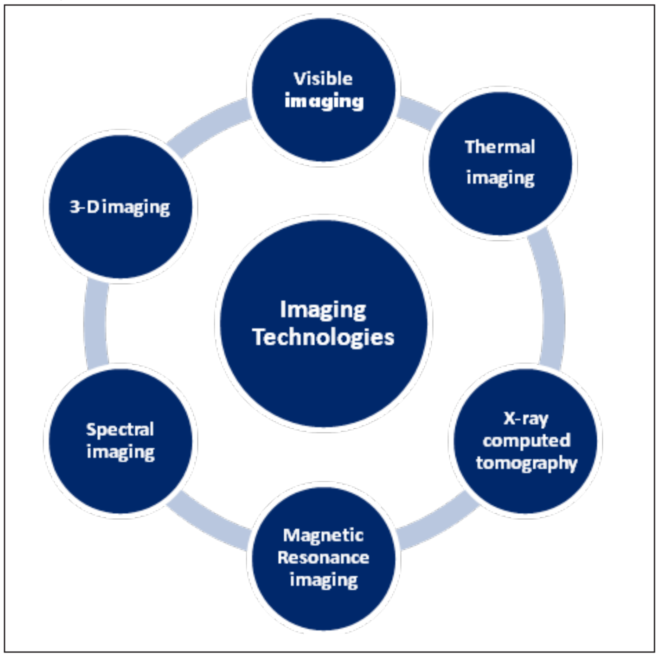

Figure 3: Imaging Technology

The Tray Scan system holds, tray labelled with a barcode so that plants can be easily identified and 3D images are taken appropriately. Early detection of diseased plants with modern vision techniques can significantly reduce costs. The virus often infects many tissues, if not the whole plant. In contrast, bacteria affects the vascular bundles especially in lower parts of the stem, causing symptoms as black-rot in the stem, and general symptoms like yellowing, necrosis and wilting of the leaves on the affected stems.

In 2016 a new experiment was set up, using 49 bacterial diseased potato plants and 20 control plants grown in the field. During the growing season, each week hyperspectral imaging and a full 3D scan was made, as well as a top view RGB-depth scan was done. Preliminary results show that plants affected by bacterial diseases are distinguishable from healthy plants using a combination of the three data modalities (Gerrit *et al*., 2017).

9. Far-Infrared (FIR) Imaging

This system uses light in the FIR region of the spectrum (15 µm to 1 mm) to study the temperature. Temperature differences can be used to study the salinity tolerance, water usage, photosynthesis efficiency *etc.* Cooler plants have better root systems and take up more water. Far infrared light can be shown on plants growing in high-tech growth cabinets to create ‘heat maps’ of each leaf. These heat maps help to study the temperature differences.

10. Near Infra Red (NIR) Imaging

The NIR region of the spectrum is used to measure water content and its movement in leaves and soil. Shortwave infrared hyperspectral (950-1650 nm) imaging system was explored to detect sour skin (*Burkholderia cepacia*) a major postharvest disease in onions on both the healthy and infected onions. Principal component analysis (PCA) revealed that neck area of the onion at two wavelengths (1070 and 1400 nm) was most indicative of the sour skin. Using the pixel number of the segregated areas, Fisher’s discriminate analysis recognized 80% healthy and sour skin-infected onions were recognised. The result of this study can be used to further develop a multispectral imaging system to detect sour skin-infected onions on packing lines (Wang *et al.,* 2012).

11. Fluorescence Imaging

Fluorescence occurs when an object absorbs light of one wavelength and gives off light of a different wavelength. A computer program converts the resulting fluorescence into falsecolour signals to allow instant analysis of plant health. Chlorophyll fluorescence is used to study the effect of different genes or environmental conditions on the efficiency of photosynthesis and stress monitoring.

The measurement of chlorophyll fluorescence of greenhouse-grown cucumber (*Cucumis sativus* L. cv. Mustang) fruit can be a useful tool in monitoring senescence, temperature stress, and desiccation during storage (Lin, 2000). Pre-symptomatic monitoring clearly opens perspectives for quantitative screening for disease resistance. These non-destructive imaging techniques were able to visualize infections at an early stage before damage appeared. Under growth-room conditions, a robotized set-up captured time series of visual, thermal, and chlorophyll fluorescence images from infected regions on attached leaves (Humplík *et al*., 2015).

12. Magnetic Resonance Imaging (MRI)

The MRI uses a magnetic field and radio waves to take images of roots in the same way as same way as it takes images of organs and soft tissues in medical applications. MRI allows the 3D geometry of roots to be viewed just as if the plant is growing in the soil and also describes 3D representation of water movement. *In situ* magnetic resonance imaging of plant roots was studied to check the different developmental stages of *Pisum sativum* (pea) seed germination. The *Pisum sativum* leaf metabolism was profiled, using 1D and 2D nuclear magnetic resonance (NMR) spectroscopy to monitor the changes induced by drought-stress under both glasshouse and simulated field conditions. Significant changes in resonances were attributed to a range of compounds, identified as both primary and secondary metabolites, highlighting metabolic pathways that are stress responsive. The metabolites present at higher concentrations in drought-stressed plants under all growth conditions included proline, valine, threonine, homoserine, myoinositol, γ-aminobutyrate (GABA) and trigonelline (nicotinic acid betaine). Such changes may be expected to impact both on plant performance and crop endues (Adrian *et al.,* 2008).

13. Spectral Reflectance

Spectral reflectance is the fraction of light reflected by a non-transparent surface. Depending on the part of the electromagnetic spectrum that is used to analyse reflectance, different matter can have different patterns of reflectance. This is called the spectral signature. It is used to determine the chemical composition of plants such as: levels of chlorophyll and other pigments in leaves, water-soluble carbohydrates and nitrogen in leaves, stems, and the chemical composition of plants *etc*. A video camera is used for plant detection. An on-the-screen display generates a video sequence and includes the measured data. As a consequence, the data can be interpreted together with an image and a clear correlation.

Phenomics researchers can use spectral reflectance technology to monitor several plant properties in the field at the same time. This means that researchers can determine the biochemical composition of crop plants without having to destroy the plants by harvesting. Researchers can use spectral reflectance to tell if a plant is stressed by saline soil or drought, well before it can be seen by eye.

14. Application of Phenomics in Field

Currently, the plant phenotyping community seems somewhat divided between high-throughput, low-resolution phenotyping and in-depth phenotyping at lower throughput and higher resolution (Dhondt *et al*., 2013). Phenotyping systems and tools applied in different scales are focused on different key characteristics– automated phenotyping platforms in controllable environment and high-throughput methodologies in field environment highlights high-throughput, while phenotyping covering the organ, tissue and cellular level emphasizes in-depth phenotyping and higher resolution (Figure 4). In this part, we systematically introduced the crop phenotyping approaches covering from cellular and tissue level to field level, and discussed the application and practical problems of which technologies in crop researches.

The hi-tech computer based imaging systems cannot only be used in controlled conditions or in a limited area, but also be used in field's at large scale. The phenomic remote sensing technology allows researchers to study plants in the field. Measurements can be taken on many plants at once and over a whole growing season. Phenonet sensor network, phenomobile, phenotower, is some of the important tools which allow plant phenotyping in the field to study large number of plants, simultaneously.

15. Phenonet Sensor Network

A network of data loggers collects informationfrom the field of crops and sends it through the mobile phone network to researchers at the lab. It saves daily visits to field sites, which is especially useful if the site is in a remote area. Sensors include; far infrared thermometer, weather sensor, soil moisture sensor, thermistor (soil temperature). The phenonet's modules are self-contained PVC tubes containing a battery, sensors, radio transmitter and microprocessor. They take measurements every 10 seconds, and average the measurements over 5 minutes.

16. Phenomobile

The phenomobile is a modified golf buggy that moves through a field of plants, taking measurements as it travels along. It can travel 3–5 km per hour. Phenomobile reveals the temperature of the leaves and also the other characteristics using the sensors. The system is currently providing a throughput around 150 micro-plots per hour, allowing sampling around 1000 micro-plots within a day. The system may also run during the night while variables are significantly reducing the data volume. The phenomobile carries equipment to measure: digital cameras, far infrared cameras, stereo-imaging system of two digital cameras with three lasers to create 3D reconstructions of plots - measure leaf area, the volume (biomass) of plants*,* height and plant density and a laser that shines red light onto the plants in combination with spectral reflectance - crop's chemical composition.

17. Phenotower

The phenotower is used to take images of crops 16 m above the ground level. The phenotower allows researchers to take images of many plants at once. The data is used to compare canopy temperature, leaf greenness and ground cover between different plant lines at the same time.

18. Multicopter

The multicopter can take images of a field from a few centimeters above the ground to a height of up to 100 meters. Multicopter will be equipped with a computer, a GPS, and colour and infrared cameras. The infrared and colour images can be used to identify the relative differences in canopy temperature. It indicates plant water use most efficiently. The Multicopter is currently in a testing phase with a normal camera, with plans for further development.

19. Rhizotron

Root observation using rhizotrons are similar to the observation of roots in soil-filled pot, except that clear acrylic glass panels allow visual monitoring of root growth at the surface of the glass. Variation in root growth and morphology among the tested crop plants can be traced on the outside surface of the acrylic glass using a marker pen, different colours may be used

(a) Phenonet Sensor Network

(b) Phenomobile

(c) Phenotower

(d) Multicopter

Figure 4: Instrumental design phenomics tools in open field condition

to indicate the presence of roots at successive time intervals, followed by scanning for root quantification.

Rhizotrons can be designed and constructed to meet specific research needs, such as for deep rooted crops and long growth periods having plants. Sampling of the root exudates around individual root tips is done using the anion exchange membrane (AEM). To analyse the root growth and architecture, researchers float the plants in water on a clear plastic tray, and use a flatbed scanner to take high-resolution,

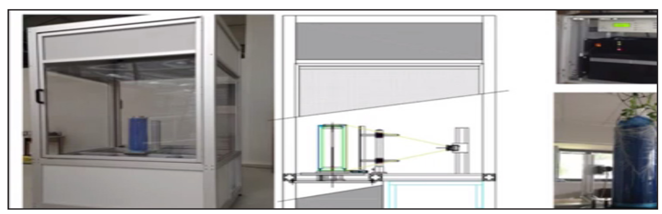

Figure 5: Rhizotron

high contrast images. A computer program transforms the images into knowledge by calculating root length and diameter, and analysing root branching patterns (Figure 5).

20. Future Prediction

In the near future, there is an urgent need to develop more adaptable, less expensive and sophisticated data analysis infrastructures for analyzing high-dimensional phenotype data sets in the phenomics area. In case more efficient statistical methods are being developed, multidisciplinary simulation models might support the proper experiment design and an improved acquisition of phenotype data. These aspects will support the promotion and explanation of plant growth, development, or responses to adverse environments. In this review, we have discussed different imaging techniques, phenotyping platforms, image analysis pipelines and phenotype data analysis methods for the high-throughput plant study. Based on our discussion we suggest that scientists should address the future challenges to enable the development of optimal digital phenotyping platforms. These challenges are, *e.g.,* the reduction of phenotyping and other related laboratory costs, the development of an efficient

Table 1: Phenomics programmes being carried all over the world

Sl. No.	Institute	Description
1	IBERS	Located at Aberystwyth University
2	International Plant Phenomics Network	A space for news about international phenomics activities
3	Lepse	Laboratory of plant ecophysiological responses to environmental stress located at Montpellier
4	National Plant Phenomics Centre	Based at IBERS, only one in its kind in UK and only one in a few in the world to enable non- destructive imaging being practice
5	Tools and Resources from- CPIB (Center for Plant Integrative Biology)	University of Nottingham
6	Purdue University Institute for Plant Sciences	Located at USA
7	The Australian Plant Facility	The high resolution plant phenomics center at Canberra

Table 2: Facilities of phenomics programmes in India

- 3rd International Plant Phenotyping Symposium was held in Chennai, India on February 2014.
- Inauguration of Plant Phenomics Facility at Central Research Institute for Dryland Agriculture (CRIDA), Hyderabad, was done on 1st July 2014. This unique state of the art facility has been installed under National Initiative on Climate Resilient Agriculture (NICRA) project launched by ICAR to develop adaptation and mitigation strategies to deal with climate change impacts on Indian agriculture.
- ICAR Sponsored Short Course on Non-destructive Phenotyping and Phenomics for Dissection of Abiotic Stress Tolerance, Gene Discovery and Crop Improvement (14-23 July, 2014) was organized by Department of Plant Physiology Indian Agricultural Research Institute.
- Inauguration of Plant Phenomics National Facility at ICAR in Indian Institute of Horticultural Research, Hessaraghatta, Bengaluru was done on 1st November 2015
- Hon'ble Prime Minister Shri. Narendra Modi dedicated the "Nanaji Deshmukh Plant Phenomics Centre" at ICAR-IARI, New Delhi to the Nation on 11th October 2017

Table 3: Phenomics application in Vegetable crops

Imaging Techniques	Species	Trait Studied
MRI	Tomato truss	Water translocation in tomato truss fruit through xylem
	Tomato	Water translocation in tomato fruit through xylem
	Bean (*Phaseolus vulgaris L.*)	Morphometric parameters in 3D; Water content, Root structure
	Sugar beet (*Beta vulgaris*)	Morphometric parameters in 3D; Water content Below ground symptoms caused by cyst bean, cyst nematode and *Rhizoctonia solani*
Visible Light Imaging	Bean 49	Projected area, Growth dynamics, Shoot biomass, Yield traits, Root architecture, Imbibition and germination rates, Early embryonic axis growth, Height, Size morphology, Flowering time
	Legume (*Medicago truncatula*)	Projected area, Growth dynamics, Shoot biomass, Yield traits, Panicle traits, Root architecture, Imbibition and germination rates, Early embryonic axis growth, Height, Size morphology, Flowering time
Fluorescence Imaging	Bean (*Phaseolus vulgaris L*)	Photosynthetic status (variable fluorescence), quantum yield, non-photochemical quenching
	Sugar beet	leaf health status, shoot architecture
	Tomato	Drought resistance study, plant health
	Chicory plant	Cold stress study

Table Continue...

Table 3: Phenomics application in Vegetable crops

Imaging Techniques	Species	Trait Studied
3D Imaging	Pepper	Shoot structure; leaf angle distributions; canopy structure; root architecture; Height
Laser Imaging	Sugar beet	Shoot biomass and Structure; leaf angle distributions; canopy structure; Root architecture; Height; Stem
MRI	Sugar beet	Morphometric parameters in 3D; Water content
	Beta vulgaris	Water content in roots
PET	Radish	Shoot to root translocation of photoassimilate
	Sugar beet	Shoot to root translocation of photoassimilate
	Beta vulgaris	Transport partitioning, sectorality, flow velocity

Table 4: Next Generation Phenotyping Studies Using Automated Devices in Vegetable Crops

Plant Species	Traits	Type of Analysis	Instrument
Tomato and Brinjal	*Alternaria solani* and *Epilachna vigintioctopunctata*	NIR Hyperspectral imaging	ASD FieldSpec Pro FR spectrometer
Tomato	*Rhizopus stolonifer spores*	NIR Hyperspectral imaging	Homemade built
Tomato	Leaves damaged by leaf miner	NIR Hyperspectral imaging	Nexus FT-NIR spectrometer
Tomato	Surface defects detection	NIR Hyperspectral imaging	ImSpector V10
Tomato	Ripeness	NIR Hyperspectral imaging	ImSpector V10
Leafy vegetables	Chlorophyll content	NIR Hyperspectral imaging	ASD Fieldspec FR spectroradiometer
Spinach	Quality during storage	NIR Hyperspectral imaging	EMCCD Luca-R cameraHyperspec®VNIR
Spinach	Crop canopy under water	NIR Hyperspectral Imaging	Specim V10 spectrometer
Watermelon	Lycopene, -Carotene, and Total Soluble Solids	NIR Hyperspectral Imaging	NIR On-Line® X-One
Lettuce	Plant traits under extreme temperature and salinity stressm treatments	NIR Hyperspectral Imaging	Series VNIR Micro-Hyperspec Sensor; Fluor Cam 800 MF
Tomato	Growing of truss	MRI, NMR	Homemade built
Tomato	Maturity	MRI	1 T MR system
Cucumber	Internal freeze damage	MRI	9.4 T MR system
Bean	Pod water content	NMR	Homemade built
Tomato	Qualitative	VIS/NIR	LabSpec 5000
Tomato	Antioxidants	VIS/NIR	HandHeld 2™
Tomato	Lycopene and physicochemical parameters	VIS/NIR	Varian Cary 500
Tomato	Varietal discrimination	VIS/NIR	USB2000 spectrometer
Tomato	Transgenic lines discrimination	VIS/NIR	FT-NIR spectrometer
Tomato	Harvest time	VIS/NIR	AgroSpec VIS-NIR spectrophotometer
Tomato	Drought stress	Chlorophyll fluorescence	Handy FluorCam FC 1000-H system
Bean	Photosyntetic traits, morphological parameters and shoot architecture	PAM fluorimetry	Growscreen Fluoro

Table Continue...

Table 4: Next Generation Phenotyping Studies Using Automated Devices in Vegetable Crops

Plant Species	Traits	Type of Analysis	Instrument
Chinese cabbage	Quality	PAM fluorimetry	FluorPenFP 100 fluorimeter
Melon	Grafting compatibility	PAM fluorimetry	Imaging-PAM fluorometer
Tomato	Fruit ripening	Chlorophyll and polyphenol content	Multiplex sensor
Leafy vegetables	Quality in post-harvest storage	Chlorophyll content	SPAD-502; Agriexpert CCN 6000
Pepper	Canopy and plant architecture	RGB	Homemade built
Tomato	Canopy	3D imaging	Digital camera
Tomato, Eggplant	Fruit morphology and shape	Tomato analyzer	Scanner imaging

data storage and less expensive analytical tools, as well as the improvement of the statistical methods to explore the plant dynamic phenotypic components and their properties.

21. Conclusion

For making successful genetic improvement in crop plants, plant breeders first identify the desirable genotypes having target traits by screening a collection of germplasm accessions. These target traits then are combined together through hybridization. This cycle of selection hybridization- selection has been implemented on the basis of visual observation since domestication of crop plants. Though visual screening is easy and precise for qualitative and highly heritable traits, its use is less precise for quantitative traits and those traits, which are difficult to observe visually (physiological and biochemical traits). Moreover, vast amount of genomic resources have been developed in a number of crop species in the past. The available gene sequences and molecular markers could still not be associated with any traits due to the lack of phenotyping of germplasm collections. For utilizing these genomic resources and identification of desirable plants, the precise phenotyping of germplasm accessions for challenging traits is required in various crop species.

In the recent past, various techniques and methodologies have been developed for screening biotic, abiotic, physiological and biochemical traits in crop plants. These technologies have become very advanced in the era of digital science. These plant phenomics developments are actually helping to make simply plant physiology in 'new clothes'. Thus this trans-disciplinary approach promises significant new breakthroughs in plant science. Phenomics provides the opportunity to study previously unexplored areas of plant science, and it provides the opportunity to bring together genetics and physiology to reveal the molecular genetic basis of a wide range of previously intractable plant processes. The challenges ahead in plant-based agriculture will require the scale of quantum advances we have seen in information technology in the past 20 years and we need to build on these advances for security of global food, fiber and fuel.

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