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Review Article

Phenomics Approach for Identification and Management of Plant Disease

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Abstract

Phenomics research into plants allows for the non-invasive tracking of development, efficiency, and composition. The missing link between the phenome and the genome is connecting phenotypes to their underlying genetic causes. Plant phenotypic plasticity is affected by the complex interactions between the DNA, the environment, and the management of the plant. The relationship between plants and pathogens, as well as the susceptibility of plants to illness, can be investigated by using phonemics. New opportunities exist thanks to sensing technologies for detecting specific phenotypic reactions during plant-pathogen interaction, allowing for faster selection of genetic material resistant to specific pathogens or strains and greater insight into the physiological mechanisms linking pathogen infection and host disease symptoms. Changes in plant diseases that have not yet shown apparent symptoms can also be detected using phonemics. In plant diseases, digital imaging, chlorophyll fluorescence imaging, spectral imaging, and thermal imaging are all used. Magnetic resonance, soft x-ray imaging, ultrasound, and volatile chemical detection are briefly described as examples of less common techniques. It is challenging to generate representative and reliably labelled training data at this size due to the observation of only mixed spectra of plant and fungal components. For this purpose, clear spectra are required. Contaminants on a surface can be detected at an early stage using infrared light. The temperature sensitivity and real-time detection capabilities of thermal imaging make it useful. There have been significant advances in high-throughput, low-cost analysis of genetic data and in non invasive phenotyping. We hope that our work may hasten the introduction of automated, non-destructive methods for high-throughput phenotyping of plant-pathogen interactions.

Introduction

Plant diseases create major economic losses in agriculture. Plant health monitoring and early pathogen detection are vital to avoid disease spread and enable appropriate control. DNA-based and serological techniques offer significant tools for plant disease diagnosis. DNA-based and serological techniques have revolutionised plant disease identification, but they're not very accurate at the asymptomatic stage, especially when a systemic pathogen is implicated. Two-day sample collection, preparation, and analysis Differential mobility spectrometers and lateral flow devices can detect early infections in the field; therefore, phenomics-based techniques help detect the pathogen early. Remote sensing and spectroscopy allow for highly specialised results and can help identify primary infections quickly. "Phenome" refers to the whole phenotype. Phenomics is the collection of organism-level phenotypic data. "Plant phenotyping" monitors a plant's genetic makeup as it interacts with its environment. Phenomics collects high-dimensional phenotypic data at multiple organisational levels to comprehensively characterise a genome's phenotypes. Plant phenomics assesses growth, performance, and composition using noninvasive technologies. D. Houle (2010). According to the IBEF and the GOI, agriculture will employ 58% of Indians in FY19. The current national ratio of extension employees to operational holdings is 1:1162, compared to 1:750. Because of this, resources and land are exploited carelessly every day, as instructed by the USDA's extension personnel. Despite covering a small number of operating holdings, they use severity-based management. Plant disease severity is usually rated by skilled raters who visually inspect plant tissue (Bock et al., 2010). Visual estimations are difficult for tiny, uniformly dispersed lesions and nearly impossible for a large area. Due to lab costs and the time needed to visually assess disease, the quantity of data points is limited.

Recent Available Methods to The Farmer for Disease Severity Analysis-Based Detection & Management

When a farmer visually identified that his field was infected with a disease, the illness severity had already affected 10-20% of the field. Then he has two options for disease management: A) The majority scientific method and B) the scientific method. (JGEC, S. Maity, 2018).

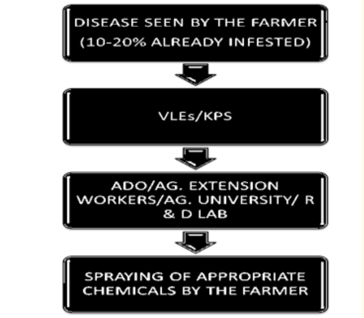
Un-Scientific Method

The majority of the farming community in the rural villages follows this method for quick solutions. But unfortunately, the lack of knowledge of the local pesticide shop owner or selecting chemicals without field visits leads the farmers and epidemics towards great devastation by increasing the cost of plant protection and it turns disease into an epidemic.

UN-SCIENTIFIC METHOD
(Approx. avg. total duration=2/3 days)

Scientific Method

Though proper detection and diagnosis can be obtained by this method but it requires a small span of time duration and the facility is not available to all.



SCIENTIFIC METHOD
(Approx. avg. total duration=10-15 days)

Image processing method vs. Traditional

Processing-Based	***Image Processing-Based Method of Disease Severity Estimation***	***Traditional Method of Disease Severity Estimation***
1.Time required	Less	More
2.Destructiveness	No	Yes
3.Trained technicians	Not required	Required
4.Availability	Available	Not available
5.Cost	Cheap	Costly
6.Early detection	Possible	Not possible

Different detection with time of use

If we considered the general disease progression cycle in four steps then.

a) **Stage-0**: Infected vectors present.
b) ***Stage-I***: Plants get infected.
c) ***Stage-II***: Pathogen established & many plants infected.
d) ***Stage-III***: Symptomatic phase & Disease spread.

And then total available methods for disease detection and severity analysis can be divided into two broad groups.

I. Traditional method.
II. Innovative Method.

Traditional method

This method can be used for stage III: symptomatic phase and disease spread stage in the field. Already disease turns epidemic and acquired required potentiality to damage the crop remarkably. Three techniques comes under traditional method.

a) Detection of Visual symptoms by Scout Teams.
b) q PCR confirmation analysis.
c) serological assays.

Innovative method: Very early (Stage-0) to symptomatic phase (Stage III) can be detected by using this method. Several techniques are there like that.

a. Volatile organic compounds.
b. Gene expression changes based on colorimetric signals (Lateral Flow Microarray).
c. Spectroscopy based methodologies.
d. Remote sensing technology.
e. Image processing based method.
f. Biosensors based on phage display & bio photonics.

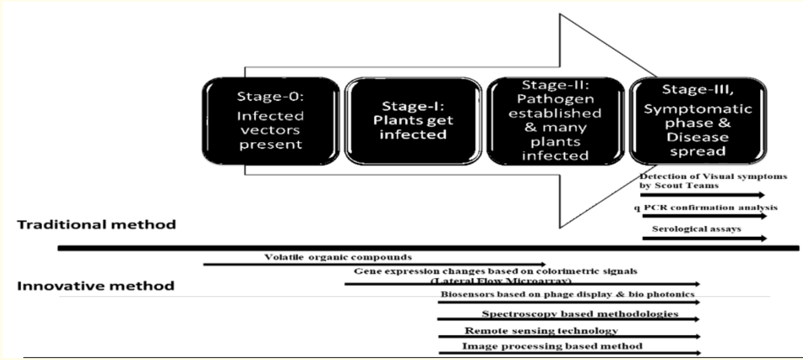
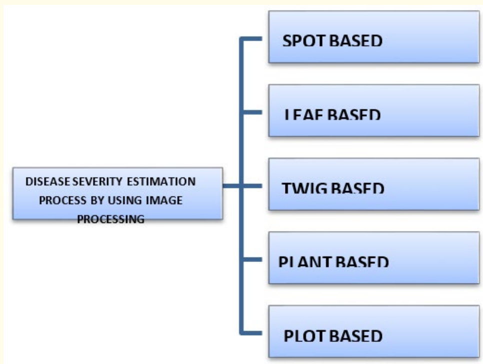


Figure 1: Traditional & innovative methods, their timing of use during plant disease.

Disease Severity Estimation Process by Using Image Processing



A) ***Spot-Based Method***: This technique is based on a single spot that shows spectral deviation from normal as the disease progresses. Here, the diseased area and the yellow halo zone are both taken into account.
B) ***Leaf-Based Method***: The total spectrum of a leaf is measured, and using an appropriate method, the mean spectrum at each wavelength is determined.
C) ***Twig-Based Method***: Total twig data calculated and deviation from normal used to determine the plant health.
D) ***Plant-Based Method***: For an orchard or any crop it can be calculated.
E) ***Plot-Based Method***: Whole plot spectrum considered.

Image Processing-Based Method

F) ***Dataset***: At first healthy to badly infected images of the foliage were collected from different angles and from different ages of the foliage. More no of the dataset leads to fewer errors.
G) ***Splitting The Data***: Then the whole data set is divided into two parts. One training dataset contains approx. 80% images and another 20% known as testing data set. (Mohanty et al.,2016).
H) ***Deep Learning***: Using any one suitable architecture among AlexNet (Krizhevsky et al.,2012), Google Net (Szegedy et al.,2015), ResNet architecture training for deep learning started.

I) ***Training/Validation Model***: Using 80% training images CNN mode was l validated. (Huan g, K.Y.2007).

J) **Performanc Metricses**: Using Accuracy (Jiang, P. et al.,2019) , CA (Fujita, E. et al.,2016), F1 (Amara, J. et al.,2017matriceses performance of developed CNN model checked with testing data set.

K) ***Visualization Technique***: This consist of how the mode differentiates disease apart from the rest of the healthy part. And using visual estimation the error was reduced as much as possible.

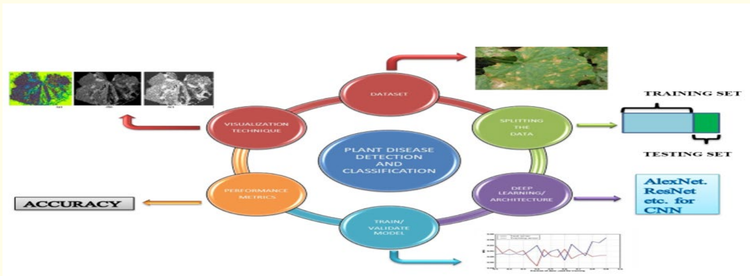


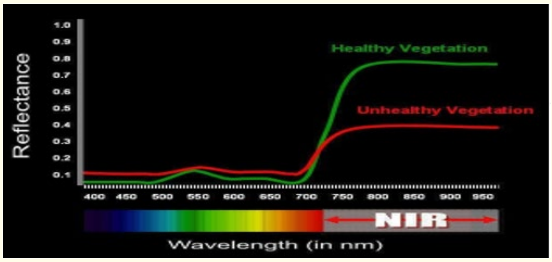
Figure 2: Overall Deep Learning process by Muhammad Hammad Saleem et al, 2019.

Fundamental Background

Bulletin ASE, European Space Agency, 2003, said that if we consider total incident light wavelengths as 100%, then 56% is used for photosynthesis purposes, nearly 11% is reflected back directly, 2% is reflected as chlorophyll fluorescence, 20% is wasted as heat energy, and nearly 11% is transmitted through the lamina. Here, we want to detect and quantify the severity of the disease by measuring the divergence of directly reflected wavelengths and photons and chlorophyll fluorescence relative to healthy foliage. From its spongy parenchymatous tissues, a healthy plant reflects the green colour wavelength (500-575 nm) and the maximum incident near-infrared (700-1500 nm) and short-wavelength infrared (1500-2500 nm) spectrums. In the case of diseased and dead leaves, however, the quantity of both spectrums varied. The reflectance of the green and infrared regional spectra is lower in sick leaves than in healthy leaves, and it is lowest in dead leaves. Taking into account the entire visible spectrum (400-700 nm), the overall reflectance of healthy foliage is less than that of diseased vegetation. Because chlorophyll and other accessory photosynthetic pigments absorb visible light in the blue (445-500 nm) and red (620-740 nm) regions for photosynthesis, In the case of the near-infrared spectrum, the cell structure of healthy and diseased vegetation differs significantly. Due to the existence of healthy spongy parenchymatous tissue in the lower surface of the leaf, the maximum incident infrared part of the spectrum is reflected; however, the reflectance of this part of the spectrum in diseased foliage is significantly reduced as spongy parenchyma degenerates.

This technique can be used for foliage-symptom-producing diseases caused by fungi, bacteria, and viruses.

Bacterial disease symptoms	***Viral disease symptoms***	***Fungal disease symptoms***
Spots	Mottling	Sooty molds
Soft spots	Distortion	Rust
Wilt	Dwarfing	Mildews
Canker		Rots
		Canker
		Spots
		Wilts



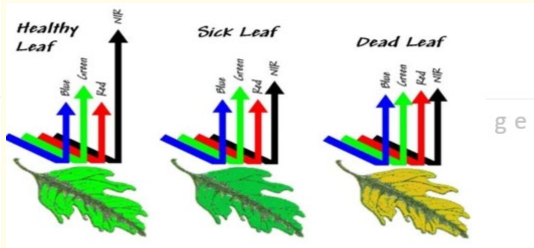


Figure 4: Reflectance vs wavelength in reference. ***Figure 5:*** Healthy to Dead via Sick leaf and Light.

Different Types of Sensors with Carrier in Different Stages of Abnormalities in Different Stages of Plant Life

RGB -Imaging

Plant pathology uses digital imagery to analyse plant health. Digital cameras provide easy-to-use RGB (red, green, and blue) images for disease detection, identification, and quantification. The light sensitivity of the camera sensor, spatial resolution, and optical and digital focus improve every year. Today, nearly every farmer and phytopathologist carries a smartphone or tablet with a digital camera sensor. Video cameras or scanners can be used to examine digital photographs of roots and inflorescences. RGB sensors of all resolutions are used to monitor plants during the growing season. Red, green, and blue pictures can identify biotic stress in plants (Bock et al. 2010). Spatial information gives critical plant disease features along with RGB and HSV (hue, saturation, value) colour information (Bock et al. 2010). Parameters for detecting and identifying plant disease symptoms include colour, grey levels, texture, dispersion, connectedness, and form (Camargo and Smith 2009; Neumann et al. 2014). Using pattern recognition and machine learning, researchers have identified plant illnesses from RGB photos (Camargo and Smith 2009; Neumann et al. 2014). Systematic selection of RGB features improves classification accuracy (Behmann et al., 2014). Plant disease assessment uses digital image analysis. ASSESS 2.0, "Leaf Doctor," Scion Image software, and custom-made modules are available (Bock et al. 2010; Wijekoon et al. 2008; http://www.plant-image-analysis.org/). In ASSESS 2.0, colour distribution is analysed in histograms to determine thresholding. A well-organized graphical user interface lets users alter parameters for healthy and sick areas. After masking the backdrop, disease severity can be retrieved as diseased pixels or a percentage. ASSESS 2.0 evaluates disease severity on single leaves and well-arranged photos. Image acquisition requires special care. Automated image analysis requires uniform focus, sharpness, and illumination for high accuracy. In natural conditions, the imaging angle (leaf orientation) and object-to-sensor distance (pixel size) affect image quality. Heterogeneous conditions and poor image quality can hinder detection and accuracy. A repeatable imaging process is the key to success.

Spectral reflectance sensors

Spectral sensors are characterised by spectral resolution (number and width of detected wavebands), spatial scale, and detector type (i.e., imaging or non-imaging sensor systems). It's twofold.

Sensors: multispectral first spectral sensor. These sensors measure object spectra in numerous broad wavebands. Multispectral imaging cameras may give R, G, B, and near-infrared data. So, the wavelength vs. reflectance graph has distinct bars. The normalised difference vegetation index (NDVI) uses NIR and red-reflected wavelengths to calculate plant greenness (Bauriegel and Herppich, 2014).

Rouse et al 1974.

$$NDVI = \frac{(NIR - Red)}{(NIR + Red)}$$

Hyperspectral sensor

Modern hyperspectral sensors have a spectrum range of 350-2,500 nm and a possible spectral resolution below 1 nm, increasing the complexity of observed data (Steiner et al. 2008). Unlike non-imaging sensors, whose average spectral Hyper-spectral imaging sensors provide spectral and spatial information about imaged objects. The graph of wavelength vs. reflectance shows a continuous line. Sensor distance affects spatial resolution. Hyper-spectral imaging combines spatial and spectral information by acquiring electromagnetic spectra at every image pixel (Bock et al., 2010). Hyper-spectral images demand a lot of disc space and computational effort to store and analyse, but they provide a plethora of information for studying plant disease characteristics. Visible (400-700 nm), near-infrared (NIR; 700-1100 nm), and short-wave infrared (1100-2500 nm) are typical plant imaging wavelengths. Hyperspectral imaging's many measurements are a big benefit. Data can be reduced to a few key spectral bands' multispectral observations. Complex algorithms can compare several wavelengths to analyse a complete spectra.

Chlorophyll fluorescence imaging

Visible disease symptoms may not be the best measure of disease severity, especially early in the infection. Stressed plants' chlorophyll fluorescence emission varies (Baker, 2008). Photosystem II emits most chlorophyll fluorescence at 685 nm (Rolfe and Scholes, 2010). Fluorescence imaging can detect stress-induced changes in chlorophyll fluorescence emission. Fv/Fm, the highest quantum efficiency of PSII, is a frequently researched fluorescence characteristic (Baker, 2008). This value is determined using Fm and Fv, the maximum and minimum fluorescence of a dark-adapted leaf (F0). Non-stressed plants keep a steady Fv/Fm value, whereas biotic or abiotic stimuli change it. Rousseau et al., 2013; Bauriegel and Herppich, 2014). This metric shifts before illness symptoms appear (Bonfig et al., 2006; Rolfe).

Thermal imaging

Pathogens have different effects on plant tissue temperature depending on the infection. Temperature reduces transpiration (Lindenthal et al., 2005). Pathogens that close plant stomata reduce transpiration and raise leaf temperature. Lindenthal et al. (2005) employed digital infrared thermography to photograph cucumber downy mildew, produced by *Pseudoperonospora cubensis*. Infrared thermography detects leaf infrared radiation to measure leaf surface temperature. Lindenthal et al. (2005) discovered that different infection stages affect leaf temperature differently. Infection with *P. cubensis* first reduces surface temperature by suppressing stomata closure. In later stages, when the virus has produced necrosis, the temperature of infected leaf tissue rises above that of uninfected tissue, possibly because injured tissue cannot perform natural cooling by transpiration. During infection, other pathogens change the leaf surface temperature. Thermal imaging of tobacco plants resistant to the tobacco mosaic virus indicated leaf temperature increases before cell death (Chaerle et al., 2001).

Volatile organic compound

Diseased plants release VOCs. If appropriately evaluated, these emissions may be useful for local disease control. Diseases change VOC emissions. Fungi, prokaryotes (bacteria and mollicutes), parasitic plants, viruses, viroids, nematodes, and protozoa cause these diseases.

Limitation

1. Analysis of disease severity using an image processing system = (IT + agriculture). Because of this, a single division cannot make any progress in the area of diagnostics using an image processing system. The successful application of AI in agriculture requires teamwork.
2. There hasn't been much study done to develop artificial intelligence (AI) in the agricultural sector.
3. Initial costs may be slightly higher. A single hyperspectral instrument set costs 18 lakhs. To process images and store data, a computer system with a lot of memory is needed.
4. Not all people have access to some data, such as hyperspectral satellite photos. This information is only accessible to authorised institutions. Decentralization is therefore crucial for achieving this goal.
5. A lot depends on the weather, including cloudiness, rainfall, etc. Environmental dangers have a significant impact on satellite

photos, UAVs, aeroplanes, and their jobs.
6. Cultural customs vary across the nation. The foliage becomes too green when there is an excess of urea or nitrogen fertilizer. Therefore, a linear relationship between fertiliser uniformity or fertiliser dose and chlorophyll growth is necessary.

Conclusion

Although this method has a lot of potential and is very accurate, it is still in its early stages and requires a lot of research and calibration work to make it more practicable and farmer-friendly with other biotic and abiotic parameters.

In recent years, there has been a rise in the prevalence of the use of sensing technologies for the purpose of identifying particular phenotypic reactions resulting from interactions between plants and pathogens. Utilization of instrumentation based on sensor data in phenomics on the other hand, effective identification of plant diseases will have an impact not only on plant pathology and genetics, but also on postharvest quality (Simko et al. 2015), agronomy, and applications such as precision agriculture. The use of remote sensing techniques in conjunction with weather monitoring systems, diagnostic assays, and appropriate epidemiological models will result in tools for the early detection of diseases in crops, allowing for timely treatment and/or treatment of plants locally, reducing the amount of chemicals required. The availability of accurate data on the assimilation, development, and health condition of plants will have a favourable effect on farm management, the creation of more environmentally friendly agricultural PR practices, and forecasting flow.

Following improvements are needed to make it a more reliable & successful method.

a) The interaction of biotic and abiotic stress has to be explored.
b) The impact of mixed infections on the optical properties of plants has to be investigated.

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