



Prevailing Methods for Plant Diseases Detection

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It is estimated that pathogens cause losses in crop yield to the tune of 20 to 40 percent. The average crop yield loss in maize, barley, rice and soybean is estimated to be around 12 percent whereas that in potatoes and ground nuts is estimated to be around 24 per cent. Post-harvest losses due to diseases are estimated to be 30 to 40 percent. To minimize the damage through disease in crops during cultivation and post-harvest processing and to ensure agricultural sustainability, early detection and prevention of diseases in crops are highly important. Disease detection methods play a key role in effective disease management, determination of pathogen, and in understanding the epidemiology and distribution of diseases in a field/ geographical area. They also play key role in pathogen taxonomy, plant quarantine, seed certification, food safety and evolutionary relationship of plant pathogens. The modern disease detection techniques have brought an evolutionary change in plant disease management strategies. Such methods are widely used and have proved beneficial to farmers as the early detection of plant disease is possible with minimal time span and corrective actions can be carried out at appropriate time. They helped us in overcoming the shortcomings of conventional methods currently in existence which are less reliable and lacks accuracy. Modern tools play a very important role to prevent risk of movement of pathogens and their vectors from one country to another, preventing outbreaks and potentially devastating crop diseases and also to identify variability of the pathogen.

Current Methods for Disease Detection

Nucleic acid and serological based methods are now providing essential tools for accurate plant disease detection.

ELISA (Enzyme linked immunosorbent assay): It is one of the serological methods where disease is identified on the basis of reaction to antibodies and colour change in the spectrometric assay. In this method, the target antigen from the virus, bacteria and fungi are made to bind specifically to antibodies conjugated to an enzyme followed by addition of enzyme substrate followed by colour change which is analyzed by ELISA reader.

PCR based methods: Real-time PCR platforms have also been used for on-site, rapid diagnosis of plant diseases based on the bacterial, fungal and viral nucleic acids. Nested and Multiplex PCR Multiplex PCR was proposed to enable simultaneous detection of different DNA or RNA by running a single reaction such as Potato viruses, Erwinia, Tilletia indica, Tomato spotted wilt virus, Potato cyst nematode etc.

FISH (Fluorescence in-situ Hybridization): It is used for bacterial detection in combination with microscopy and hybridization of DNA probes and target gene from diseased plant samples. The presence of pathogen specific rRNA sequences in plants is recognized with the help of this technique. It is also used to detect fungi, viruses and endophytic bacteria.

Flow Cytometry: It is a laser-based optical technique which uses an incident laser beam and measures the scattering and fluorescence of laser beam reflected from the sample. It has been used for rapid detection of food borne bacterial pathogens such as, example *Bacillus subtilis* in mushroom compost.

Thermography: Temperature changes in plant leaves and canopies are captured by thermographic cameras on emitted infrared radiation. It is also a promising tool to monitor the heterogeneity in the infection of soil borne pathogens.

Hypersepectral technique: This technique is used for plant phenotyping and crop disease identification in large scale agriculture. The imaging cameras facilitate the data collection in three dimensions, X- axes, Y- Axes for spatial and Z-axes for spectral which contributes to more detailed and accurate information about plant health across a large geographical area by measuring the changes in reflectance resulting from the biophysical and biochemical characteristics changes upon infection. For example., *Magnaporthe grisea*, *Phytophthora infestans* and *Venturia inaequalis* infection. Furthermore, Gas Chromatography: In this technique particular disease can be identified with the help of specific volatile organic compounds produced by the particular disease for example, *Phytophthora cactorum* causing crown rot disease in strawberry releases p-ethylguaiaicol and p- ethylphenol as VOCs from the infected portion of the strawberry plant/ fruit.

Biosensors based methods: A biosensor is an inquisitive device which is able to convert a biological signal into electrical signal where immobilized layer of biological material is connected with sensor which inspects the biological signal & get it converted into electrical signal. Leland C. Clark Jr. known as the father of the biosensor. Various biosensors are commercially available such as biosensors platforms based on nanomaterials, affinity biosensors, antibody-based biosensor, Nucleic acid-based affinity biosensors, enzymatic electrochemical biosensors and bacteriophage-based biosensors. A bioreceptor which can be antibody, enzymes, phages, oligonucleotides receive an analyte of interest which further processed by a transducer which may be optical, electrochemical, mechanical and cantilever and after processing it will produce a signal which could be luminescence, fluorescence, UV-VIS, magnetic and resonance through which specific disease could be detected. Novel sensors based on the analysis of host responses, e.g., differential mobility spectrometer and lateral flow devices, deliver instantaneous results and can effectively detect early infections directly in the field. Some biosensors based on phage display and biophotonics can also detect instantaneous infections although they can be integrated with other systems and if remote sensing techniques coupled with spectroscopy-based methods its allow high spatialization of results, these techniques may be very useful as a rapid preliminary identification of primary infections. However, serological and PCR-based methods are the most convenient and effectual to confirm specific plant disease, volatile and biophotonic sensors provide instantaneous results and may be used to identify infections at asymptomatic stages. The untamed potential of various biosensors for plant disease detection has been comprehensively reviewed and the advent of nanotechnology has resulted in the advancement of highly sensitive biosensors due to modern nanofabrication techniques. The specificity of the biosensors could be greatly enhanced by the use of enzymes, antibodies, DNA and bacteriophage as the specific recognition element.

Molecular techniques can reduce the time required for assay and increase the sensitivity of assay, allowing detection of pathogen before symptom expression. On the other hand, imaging techniques such as thermography, hyperspectral imaging and fluorescence imaging are proved to be susceptible to parameter change of the environment.

Modern detection tools are needed to detect pathogens before symptoms development, screen large number of samples accurately, reliably and quickly and find the ways that provide additional information. The advancement of disease detection methods needs to be more specific, rapid, cost effective and sensitive so that the prevention measures for specific disease could be taken before it clogs the way to food security of world.