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5. Diagnostic Tools and Techniques for Tuber Disease Identification

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Abstract:

Tuber crops are staple foods for millions of people worldwide, providing a significant source of calories and essential nutrients. After cereals (49%) as the primary crop group that provides dietary energy to people, tuber crops (with 5.4% energy) rank second in significance. There are five major tuber crops in India, they are potatoes, sweet potato, taro, cassava and yam. Many diseases that can seriously affect yields and quality of these crops. Timely and accurate detection of diseases in tuber crops is essential for implementing effective control and management measures. Early detection makes it possible to take swift action, limiting crop losses and stopping the spread of diseases. In this chapter, we discussed about the detection and diagnostic techniques for the identification of disease of tuber crop.

Keywords:

Diseases, Tuber Crops, Detection, Identification etc.

5.1 Introduction:

Tuber crops, such as potatoes, sweet potatoes, yams, and cassava, play a crucial role in global food security and livelihoods. Tuber crops are staple foods for millions of people worldwide, providing a significant source of calories and essential nutrients. In several developing countries, they are the main source of food and help generate revenue. After cereals (49%) as the primary crop group that provides dietary energy to people, tuber crops (with 5.4% energy) rank second in significance (Nayar, 2014). About 67 million hectares are used to grow tuber crops worldwide, and they produced 887 million tons in 2017 (FAOSTAT, 2019). There are five major tuber crops in India, they are potatoes, sweet potato, taro, cassava and yam. Among these all are tropical crop except potato (More *et al*., 2019). Many diseases that can seriously affect yields and quality of these crops. Biotic factors, such as bacteria, fungi, viruses, and nematodes, as well as abiotic factors like environmental stress, contribute to the onset and spread of diseases. Timely and accurate detection of diseases in tuber crops is essential for implementing effective control and management measures. Traditional methods of disease detection, such as visual inspection, often lack accuracy and efficiency, leading to delayed responses and increased crop losses. In recent years, advancements in technology and diagnostic techniques have provided innovative tools for the early detection and precise diagnosis of tuber crop diseases. Molecular techniques, such as polymerase chain reaction (PCR) and DNA sequencing, have proven to be powerful tools for identifying the specific pathogens responsible for the diseases.

Additionally, remote sensing technologies, including satellite imaging and drone-based surveillance, offer the potential for large-scale monitoring of crop health. Early detection makes it possible to take swift action, limiting crop losses and stopping the spread of diseases. Additionally, accurate diagnosis aids in the selection of appropriate control strategies, whether through the use of resistant varieties, cultural practices, or the application of agrochemicals. In this chapter we discuss about the diagnostic tools for disease identification in tuber crops.

5.2 Diagnostic Methods:

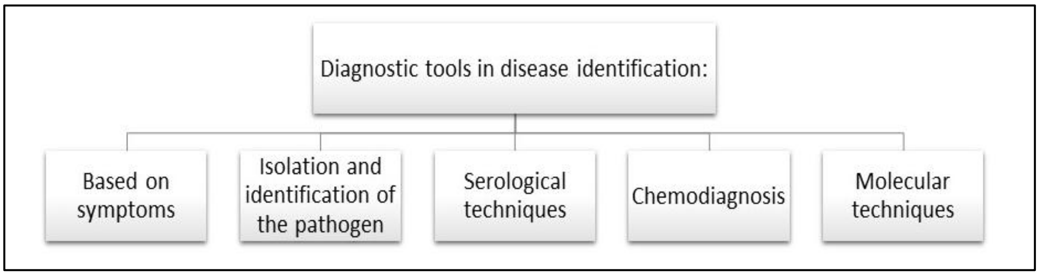

Figure 5.1: Diagnostic Methods

5.2.1 Based on Symptoms:

Identification of diseases in tuber crops involves recognizing specific symptoms and understanding the patterns of expression associated with various pathogens. Different tuber crops may exhibit distinct symptoms when affected by diseases. Accurate identification of disease symptoms requires a combination of visual observation, field surveys, and sometimes laboratory analyses. The pathogen affecting plants parts can be identified by visualizing under microscope. Some of the major disease symptoms of tuber crops were discussed below.

1. Late Blight (*Phytophthora infestans*):

Symptoms: Dark green to black lesions with a water-soaked appearance on leaves. Lesions can rapidly enlarge, and a fuzzy, white mold may develop on the undersides of leaves. Infected tubers exhibit a dark, firm rot (Arora *et al.*, 2014). This disease mainly infects potato. Late blight of potato cause epidemics in Ireland and it is known as Iris famine in 1845.

Figure 5.2(a): Water-soaked black lesion appearance on leaves

Figure 5.2.(b): Tuber exhibits irregular reddish brown to purplish rotting

2. Early Blight (*Alternaria solani*):

Symptoms: Brown, concentric rings on older leaves, starting as small lesions. Lesions may coalesce, leading to large, irregularly shaped necrotic areas. Lower leaves are typically affected first (Campo Arana *et al*., 2007).

Figure 5.3: Brown Concentric Rings on The Leaves

3. Bacterial Soft Rot (*Erwinia*):

Symptoms: Soft, watery, and foul-smelling decay in tubers. Stems and leaves may also show wilting and yellowing. Bacterial ooze may be present on infected tissues (Webb and Wood, 1974).

Figure 5.4: Soft Watery and Rotting of Tubers

4. Common Scab (*Streptomyces scabies*):

Symptoms: Raised, corky lesions on the surface of tubers. Lesions are often tan to brown and may coalesce to cover significant portions of the tuber surface. Scabs can affect the marketability of the crop (Lawrence *et al*., 1990).

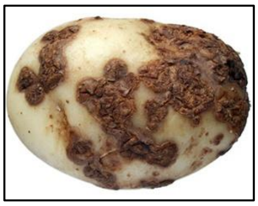

Figure 5.5: Raised, corky lesions on the surface of tubers

5. Powdery Scab (*Spongospora subterranea*):

Symptoms: Powdery pustules on tuber surfaces, leading to irregular-shaped craters. Affected tubers may have a rough appearance. The disease can affect yield and quality (Van de Graaf *et al*., 2005).

Figure 5.6: Powdery Pustules on Tuber Surfaces

6. Cassava Mosaic Disease (*Cassava Mosaic Virus*):

Symptoms: Yellow mosaic patterns on leaves, stunted growth, and distortion of leaf shape. Severe infections can lead to reduced tuber production (Hillocks and Thresh, 2000).

Transmission: Spread by whiteflies (*Bemisia tabaci*).

Figure 5.7: Mosaic Patterns on The Leaves

7. Anthracnose (*Colletotrichum gloeosporioides)*.

Symptoms: Dark necrotic lesions on leaves, stems, and petioles. Lesions may have a target-like appearance. In severe cases, can lead to defoliation (Sangpueak *et al*., 2018).

8. Pink Rot (*Phytophthora erythroseptica*):

Symptoms: Pinkish-brown discoloration of tuber vascular tissue. Infected tissue is soft and may have a foul odor. Tubers may rot during storage (Salas *et al*., 2000).

Figure 5.8: Pinkish-Brown Discoloration of Tuber Vascular Tissue

9. Yam Mosaic (Yam mosaic virus (YMV) and Yam mild mosaic virus (YMMV)).

Symptoms: Mottling, yellowing, and distortion of leaves. Reduced tuber size and yield (Eni, 2015).

Transmission: Spread by aphids and through infected planting material.

Figure 5.9: Yam Mosaic

10. Potato virus Y:

Symptoms: Potato virus Y (PVY) is a Potyvirus, causes stipple streak. The necrotic strain generally causes mild foliage symptoms, but necrosis in the leaves of susceptible potato varieties.

5.2.2 Isolation and Identification of The Pathogen:

The infected plant samples and tubers were collected from the field and the isolation of the pathogen was carried out by tissue segmentation method. The cultural characteristics were noted.

Based on the cultural characteristics the causal organism of the disease will be identified. Koch postulates for pathogenicity need to prove in order confirm the causal organism of particular disease. It is time consuming process.

5.2.3 Serological Techniques:

Serological diagnosis for tuber crop diseases involves the detection and identification of pathogens by analyzing the specific antibodies or antigens associated with the diseases. This method relies on the principles of immunology and involves various techniques to identify and quantify the presence of pathogens.

1. Enzyme-Linked Immunosorbent Assay (ELISA) (Clark *et al*., 1986):

Principle: ELISA is a widely used serological technique that utilizes the binding specificity of antibodies to detect the presence of pathogens.

Procedure: Tissue extracts or sap from tuber crops are applied to a solid support (microplate), and specific antibodies linked to enzymes are used to detect the presence of target antigens. Enzyme activity is measured, indicating the amount of the pathogen.

Applications: ELISA is used for the detection of various pathogens, including viruses, bacteria, and fungi affecting tuber crops.

2. Western Blotting (Goudou-Urbino *et al*.,1996):

Principle: Western blotting separates proteins from a sample based on their molecular weight and then detects specific proteins using antibodies.

Procedure: Tuber crop extracts are separated using gel electrophoresis, transferred to a membrane, and probed with specific antibodies. The presence of pathogen-specific proteins is revealed through chemiluminescence or colorimetric reactions.

Applications: Western blotting is particularly useful for confirming the presence of specific proteins associated with diseases.

3. Dot Immunobinding Assay (DIBA) (Njukeng *et al.*, 2005):

Principle: DIBA is a simple and rapid technique that involves dotting a small volume of plant sap or extract onto a solid support and detecting the presence of pathogens using specific antibodies.

Procedure: Dots containing the sample are incubated with pathogen-specific antibodies, and the reaction is visualized through colour development.

Applications: DIBA is suitable for on-site testing and initial screening of tuber crops for the presence of pathogens.

4. Immunofluorescence Assay (IFA):

Principle: IFA involves labelling pathogens with fluorescent dyes conjugated to antibodies, allowing for the visualization of the pathogens under a fluorescence microscope.

Procedure: Tuber crop samples are treated with specific antibodies labelled with fluorescent dyes. The labelled pathogens can be observed and quantified based on the intensity of fluorescence.

Applications: IFA is valuable for visualizing pathogens within plant tissues and determining their distribution.

5. Lateral Flow Devices (LFDs) (El-Badry *et al.*, 2009):

Principle: LFDs are rapid and user-friendly diagnostic tools that utilize antigen-antibody interactions to detect the presence of specific pathogens.

Procedure: Plant extracts or sap are applied to the LFD strip, and the strip displays colored lines in the presence of the target pathogen.

Applications: LFDs are suitable for on-site detection, providing quick results without the need for specialized equipment.

5.2.4 Chemodiagnosis:

Tetrazolium chloride test (Lozano, 1975) used to detect Cassava Mosaic Virus.

Principle: The Tetrazolium chloride test is a chemical assay that involves the use of tetrazolium chloride to detect the activity of dehydrogenase enzymes, which are often associated with cellular respiration.

Procedure: The thin section of petiole or midrib of the leaf is taken and soak in 5ml of 1% tetrazolium chrolide for 30 min to 1hour. After the incubation if the section turns into brick red colour it indicates the presences of the virus.

5.2.5 Molecular Techniques:

Molecular techniques have become powerful tools for the diagnosis of diseases in tuber crops. These methods allow for the detection and identification of pathogens at the molecular level, providing accurate and sensitive results. Here are some commonly used molecular techniques for disease diagnosis in tuber crops

1. Polymerase Chain Reaction (PCR):

- **Principle:** PCR amplifies specific DNA sequences, allowing the detection of pathogens by targeting their genomic DNA.
- **Applications:** PCR is widely used for the detection of viruses, bacteria, and fungi affecting tuber crops. Specific primers designed for the target pathogen enable the amplification of pathogen-specific DNA fragments.

2. Reverse Transcription PCR (RT-PCR):

- **Principle:** RT-PCR is used for the detection of RNA viruses by first converting viral RNA into complementary DNA (cDNA) using reverse transcriptase.
- **Applications:** RT-PCR is commonly employed for the detection of RNA viruses, such as Potato virus Y (PVY) or Potato virus X (PVX), in tuber crops.

3. Quantitative PCR (qPCR):

- **Principle:** qPCR is a real-time PCR technique that allows the quantification of the amount of target DNA or RNA in a sample.
- **Applications:** qPCR is used for both qualitative and quantitative analysis of pathogens. It provides information on the level of infection, aiding in disease management decisions.

4. Loop-Mediated Isothermal Amplification (LAMP):

- **Principle:** LAMP is an isothermal nucleic acid amplification technique that amplifies DNA with high specificity and efficiency under isothermal conditions.
- **Applications:** LAMP is suitable for on-site diagnosis due to its simplicity and rapidity. It can be used for detecting various pathogens in tuber crops.

5. Microarrays:

- **Principle:** Microarrays allow the simultaneous detection of multiple pathogens or genes in a single assay by hybridizing DNA or RNA to specific probes immobilized on a solid surface.
- **Applications:** Microarrays are useful for screening a broad range of pathogens in a single experiment. They are employed for the simultaneous detection of multiple viruses, bacteria, or fungi in tuber crops.

5.2.6 Other Techniques:

1. Remote Sensing and Imaging:

- Remote sensing technologies, including satellite imagery and unmanned aerial vehicles (UAVs or drones), can be used to monitor large agricultural areas for signs of stress or disease.
- Hyperspectral imaging can help detect subtle changes in plant health that may indicate the presence of diseases.

2. Sensor Technologies:

- Field-deployable sensors, such as those based on electronic nose technology, can detect volatile organic compounds emitted by infected plants. These compounds may serve as biomarkers for specific diseases.

3. Data Analytics and Artificial Intelligence:

- Machine learning algorithms can be trained on large datasets of plant disease images and sensor data to develop predictive models for disease detection.

5.3 Conclusion:

The challenges posed by various pathogens require innovative and integrated approaches to effectively identify, manage, and mitigate the impact of diseases on tuber crops. Advancements in molecular techniques, such as PCR and DNA sequencing, have revolutionized our ability to pinpoint the specific pathogens responsible for tuber crop diseases, enabling targeted and precise interventions. The advent of remote sensing technologies, coupled with satellite imaging and drone-based surveillance, offers a scalable and efficient means of monitoring crop health on a larger scale. These tools empower agricultural stakeholders with timely information, allowing for swift and informed decision-making.

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