

## Biotechnological Approaches for Crop Disease Management

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ARTICLE ID: 09

### Introduction

Any abnormal condition that damages a plant and reduces its productivity or usefulness to man is called disease. The occurrence and prevalence of plant diseases vary from season to season, depending on the presence of the pathogen, environmental conditions and crops or varieties grown. Major disease outbreaks among food crops have led to famines. In history, three famine, Irish famine, Bengal famine and coffee rust were causes millions of people to die. Plant disease were two types: Parasitic disease which are caused by living organism which was also called as pathogen. Non parasitic disease which are caused by external condition to plant.

Biotechnology is the genetic manipulation and multiplication of any living organism through new technologies resulting in the production of improved new organism and products can be used in a variety of ways.

### Role of biotechnology in plant disease management

- To obtain pathogen-free mother plants through rapid clonal propagation.
- New plants to which genes have been incorporated through genetic engineering.
- The main vehicle for transferring genes from donor to recipient, in plant pathogens, particularly the bacterium *Agrobacterium tumefaciens* and the *cauliflower mosaic virus*.
- The study of plant genes for resistance to disease and pathogen, genes for virulence to pathogen added by genetic engineering techniques.

### Detection of pathogen

The traditional method of identifying plant pathogens is through visual examination. This is often possible only after major damage has already been done to the crop, so treatments will be of limited or no use. For this reason, the availability of fast, sensitive and accurate

methods for detection and identification of plant pathogens is increasingly necessary. The different types of polymerase chain reaction (PCR) are the most common DNA amplification technology used for detecting various plant pathogens. With the applications of bioinformatics as a modern technology in plant pathology, identification of specific motifs, DNA sequences has become possible, which ultimately increase the accuracy of modern techniques in plant disease diagnosis. The newly emerged proteomic technology is also a promising tool for providing information about pathogenicity and virulence factors.

### Methods for Plant Disease Detection

1. **Direct Detection Methods:** Polymerase Chain Reaction, Fluorescence in-situ Hybridization, Enzyme-Linked Immunosorbent Assay, Microarray technique, Flow Cytometry
2. **Indirect Detection Methods:** Gas Chromatography, Thermography, Fluorescence Imaging, Hyperspectral Techniques
3. **Using Portable Sensors:** Biosensor Platforms Based on Nano materials, Affinity Biosensors, Enzymatic Electrochemical Biosensors, Bacteriophage-Based Biosensors

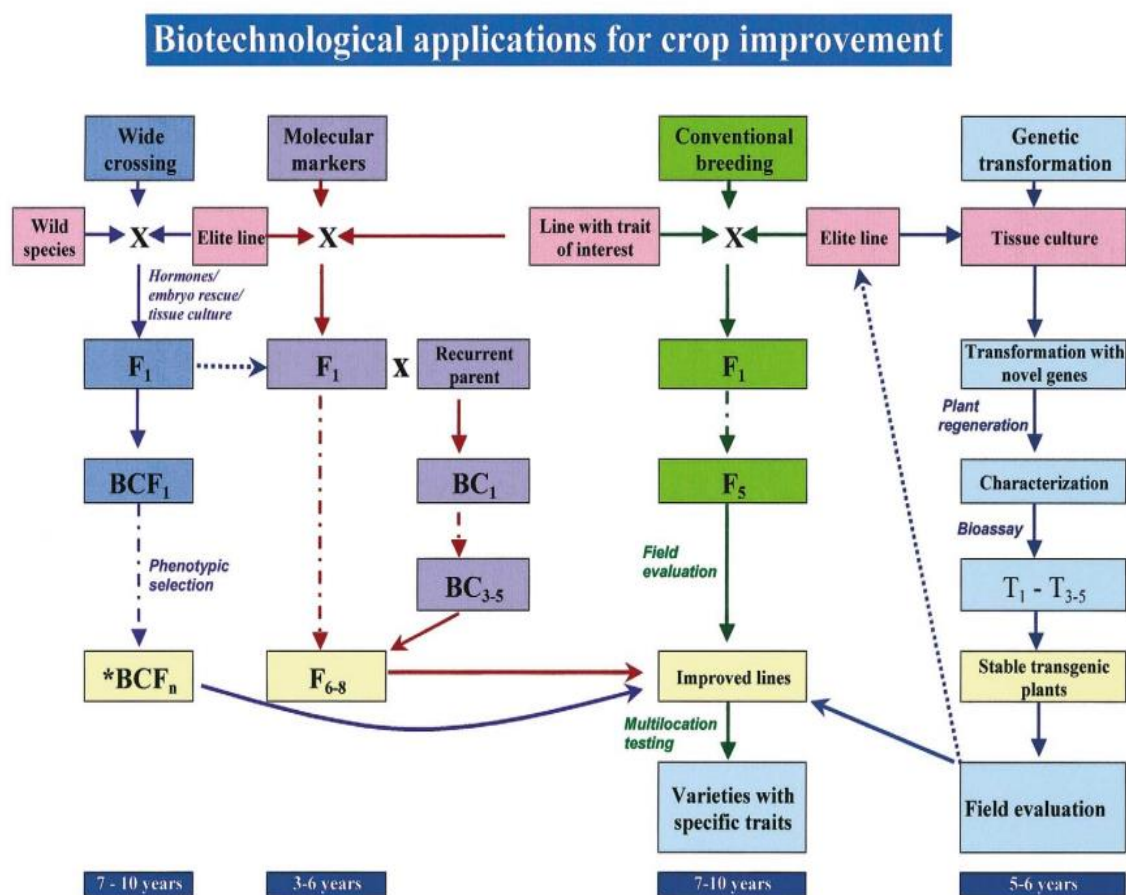
### Biotechnological approaches

- ✚ Molecular markers approach
- ✚ Plant tissue culture
- ✚ Genetic engineering
- ✚ RNA interference
- ✚ Monoclonal antibody techniques
- ✚ Epigenome modification

1. **Molecular markers approach:** There were lot of markers available for detection of resistance gene to pathogen. Insight into marker technology,
  - ✚ **Marker-assisted selection:** is an indirect selection process where a trait of interest is selected based on a marker. Leaf rust resistance in barley, leaf blight in tomato, white mold in beans etc.
  - ✚ **Marker-assisted Backcrossing:** in which the goal is to incorporate a major gene from an agronomically inferior source (the donor parent) into an elite cultivar or breeding line (the recurrent parent). TMV 2 variety of groundnut which is resistance to leaf spot and rust developed by MABC by selecting resistance loci linked

markers. **Marker-assisted Gene Pyramiding:** Gene pyramiding, which aims to assemble multiple desirable genes into a single genotype. Marker-based gene pyramiding is now the method of choice for breeding line development targeted at improving traits controlled by major genes. In rice pyramiding of *xa5*, *xa7*, *xa13* and *xa21* gives BLB resistance.

- ✚ **Marker-assisted Recurrent Selection:** The objective of marker assisted recurrent selection (MARS) is to increase the frequency of favorable marker alleles in a population before inbred line extraction. MARS combined with GS improved population with different resistance markers.



## 2. Plant tissue culture: It include

- ✚ **protoplast fusion** is one of the methods that can be used to circumvent problems in introgression genes for resistance. By this method, factors that contribute to crossing barriers between species can be avoided and viable hybrids (Cybrids) have been recovered even between distantly related species.

- ✚ **Meristem culture** method in which apical meristem is used to produce disease free plants. Apical meristem is a dome of tissue located at the extreme tip of a shoot. There is a lack of vascular tissue formation which is the main reason for disease free propagation.
  - ✚ **Soma clonal variations** is a technique under plant tissue culture in which plant itself generate variations *via.* genetic or epigenetic changes.
3. **Genetic engineering:** Plant genetic engineering rely on conventional transgenic approaches, cisgenic approaches and the more recent genome-editing technologies. Genetic engineering is the technology by which it is possible to isolate particular gene from one organism, insert them into the genome of another organism and make them to express at right time. Cells of plants can be cultured in special nutrient medium and whole plants can be regenerated from cultured cells. Genetic engineering involves gene transfer, gene silencing, gene mutation and regulation of transcriptional factors.
  4. **RNA interference:** RNA interference refers collectively to diverse RNA based processes that all result in sequence-specific inhibition of gene expression at the transcription, mRNA stability or translational level. The production of small RNAs (21-26 nucleotides) that act as specific determinants for down-regulating gene expression and the requirement for one or more members of the Argonaute family of proteins. RNAi operates by triggering the action of dsRNA intermediates, which are processed into RNA duplexes of 21-24 nucleotides by a ribonuclease III-like enzyme called Dicer. Once produced, these small RNA molecules or short interfering RNAs (siRNAs) are incorporated in a multi-subunit complex called RNA induced silencing complex (RISC). The siRNAs within RISC acts as a guide to target the degradation of complementary messenger RNAs (mRNAs). The host genome codifies for small RNAs called miRNAs that are responsible for endogenous gene silencing.
  5. **Monoclonal antibody techniques:** In this technique there is the fusion of myeloma cells (cancer cells) with antibody producing white blood cell (B- lymphocytes). The resulting hybrid celled is called a hybridoma. Techniques have been developed to produce large quantities of identical antibodies. These new antibody-forming hybrid cells, hybridomas, can now be grown culture indefinitely. Each hybridoma clone produces only one type of antibody *via.* selection techniques. The clone that produces

the desired antibody can be chosen. Monoclonal antibodies can be obtained from the liquid of hybridoma cultures and can be used to detect, identify and measure the antigens that induced their production.

- 6. Epigenome modification:** Epigenetic refers to change in phenotype without change in genotype. It can be done by DNA methylation, histone modification and RNA mediated modification. Numerous studies indicate that DNA methylation plays a part in the pathogen-induced immune system and can strongly influence the resistance response in different plant species. DNA methylation at CG, CHG and CHH (H = A, T or C) contexts through distinct pathways.

