

A Note on Techniques for Analysis Plant Disease Detection

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ABOUT THE STUDY

A multitude of PCR-based diagnostic techniques are available in modern plant pathology. As a result, the availability of data produced from phytopathogen genome sequencing is critical. Because the primers ensure the specificity of PCR, they must be chosen and designed correctly for any PCR analysis to be successful. The selection of primers is the first step in PCR diagnostics. Because the genomes of viruses and viroids are generally short, complete sequencing data is readily available in databases, and appropriate primers are readily available. The genomes of bacteria, oomycetes, and fungi have collected less information, despite the fact that the amount of data is expanding. General approaches to selecting specific known DNA target pieces are available for these diseases, and procedures based on screening random DNA sections have been developed.

DNA encoding ribosomal RNA (rDNA) is commonly employed as a target sequence in bacteria, oomycetes, and fungi. rDNA is suitable for diagnostic use due to a number of factors. Each cell has many copies of rDNA, increasing the sensitivity of detection. The genes are found in all organisms and have highly conserved 5.8 S regions, making rDNA universal. Internal Transcribed Spacer (ITS) regions, for example, are very varied. The conserved sequences can be utilized to create universal primers for group identification of microorganisms within a taxon (for all oomycetes, fungus, or bacteria), but the existence of variable regions allows for race, strain, and isolate distinctions. Beta-tubulin genes, which are linked to fungicide resistance, are another target sequence employed for fungal detection. The target pieces are usually derived from DNA found in bacterial plasmids and pathogenicity-associated genes.

Random Amplified Polymorphic DNA-PCR (RAPD-PCR), also known as Arbitrary Primed Polymerase Chain Reaction (AP-PCR), is one of the approaches utilized when the target

nucleotide sequence is uncertain. The RAPD-PCR is commonly used to study DNA polymorphism, gene mapping, and demographic and evolutionary biology, either alone or in combination with RFLP. Because it enables for the screening and discrimination of sequences specific for closely related species, strains, races, and isolates, the RAPD-PCR is important for plant pathogen diagnosis.

RAPD-PCR uses annealing of single primers, as opposed to the described PCR analyses, which use two primers to restrict the generated sequence. After amplification, the primer binds to the genomic DNA's random complementary sequences, yielding a RAPD-PCR product of variable length that is partially or wholly homologous to the arbitrarily primed sequence at both ends. The DNA polymorphism, which is caused by insertions, deletions, and base substitutions, affects the RAPD-PCR product, which shows up as the presence or lack of bands in the gel following RAPD-PCR. This approach can be used to amplify gene products from a variety of organisms, and the pattern of the bands after electrophoresis will be unique to each creature. To find a band that is specific for a target, many different primers must be examined. For the synthesis of extremely specific primers, particular bands might be employed.

Finally, it's worth noting that PCR isn't the only method for detecting amplification. The ligase chain reaction, for example, has been used to detect several plant diseases. It is based on the ability of a DNA-dependent DNA-ligase to ligate a DNA strand in the presence of Adenosine Triphosphate (ATP) and Mg²⁺ ions when the phosphodiesteric bond is ruptured. In 1989, Wu and Wallace proposed this approach. High specific activity in ligation of single-stranded ruptures at the template, which forms the second complementary strand, and low specific activity in simultaneous ligation of two ruptures in both strands or rupture in single-stranded DNA are two characteristics of DNA ligase work.

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Received: November 11, 2021; **Accepted:** November 25, 2021; **Published:** December 02, 2021

Citation: Steven R (2021) A Note on Techniques for Analysis Plant Disease Detection. J Plant Biochem Physiol. 9:276.

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