PrimeDiagnostics

Platforms for the Detection of Plant Diseases

Detection methods

Reliable diagnostics for the detection of plant diseases is the basis of the cultivation of healthy crops and trade in disease-free products.

Prime Diagnostics produces reagents and systems for detection and identification of plant pathogens. We supply high-quality diagnostic reagents and services that meet the specific requirements of inspection services, research organizations and companies involved in plant breeding and crop protection. Our knowledge is rooted in plant research at Wageningen University & Research.

Different methods can be used to identify different plant pathogens, depending on the question. Each of them has strengths and trade-offs, so choosing the right technique depends on balancing the need for speed, accuracy, and cost for a specific application.

This folder will highlight the characteristics of the different detection methods we offer, and briefly explain the science behind them.

Overview of techniques

	Antibody based				DNA/RNA based	
Target	IIF	DAS-ELISA	Luminex xMAP	LFD	TaqMan	Luminex (xTAG)
Bacteria	✓	✓	✓	✓	✓	✓
Virus		✓	✓	✓	✓	✓
Oomycete				✓	✓	✓
Fungi					✓	✓
Viroid					✓	✓

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https://shop.wur.nl/primediagnostics

For questions you can contact us on
Email: primediagnostics@wur.nl
WhatsApp: +31 634 229 997

Lateral flow devices	
Speed	●●●●●
Location	In the field
Sensitivity	●●○○○
Number of pathogens per sample	1
Number of samples per test	1
Qualitative/quantitative	Qualitative
Throughput	Low
Costs	●●●●○
Added value of this platform	Simple and fast

Luminex xTAG	
Speed	●○○○○
Location	In the laboratory
Sensitivity	●●●●●
Number of pathogens per sample	1-50
Number of samples per test	1-96
Qualitative/quantitative	Semi-quantitative
Throughput	Very high
Costs	●●○○○
Added value of this platform	Multiplex, automation possible, flexible

Luminex xMAP	
Speed	●●●○○
Location	In the laboratory
Sensitivity	●●●●○
Number of pathogens per sample	1-50
Number of samples per test	1-96
Qualitative/quantitative	Semi-quantitative
Throughput	Very high
Costs	●○○○○
Added value of this platform	Multiplex, automation possible, flexible

DAS-ELISA	
Speed	●○○○○
Location	In the laboratory
Sensitivity	●●●○○
Number of pathogens per sample	1
Number of samples per test	1-96
Qualitative/quantitative	Qualitative
Throughput	High
Costs	●●○○○
Added value of this platform	Automation possible, widely applicable

TaqMan	
Speed	●●●○○
Location	In the laboratory
Sensitivity	●●●●●
Number of pathogens per sample	1-4
Number of samples per test	1-96
Qualitative/quantitative	Quantitative
Throughput	High
Costs	●●○○○
Added value of this platform	Quantitative, multiplex

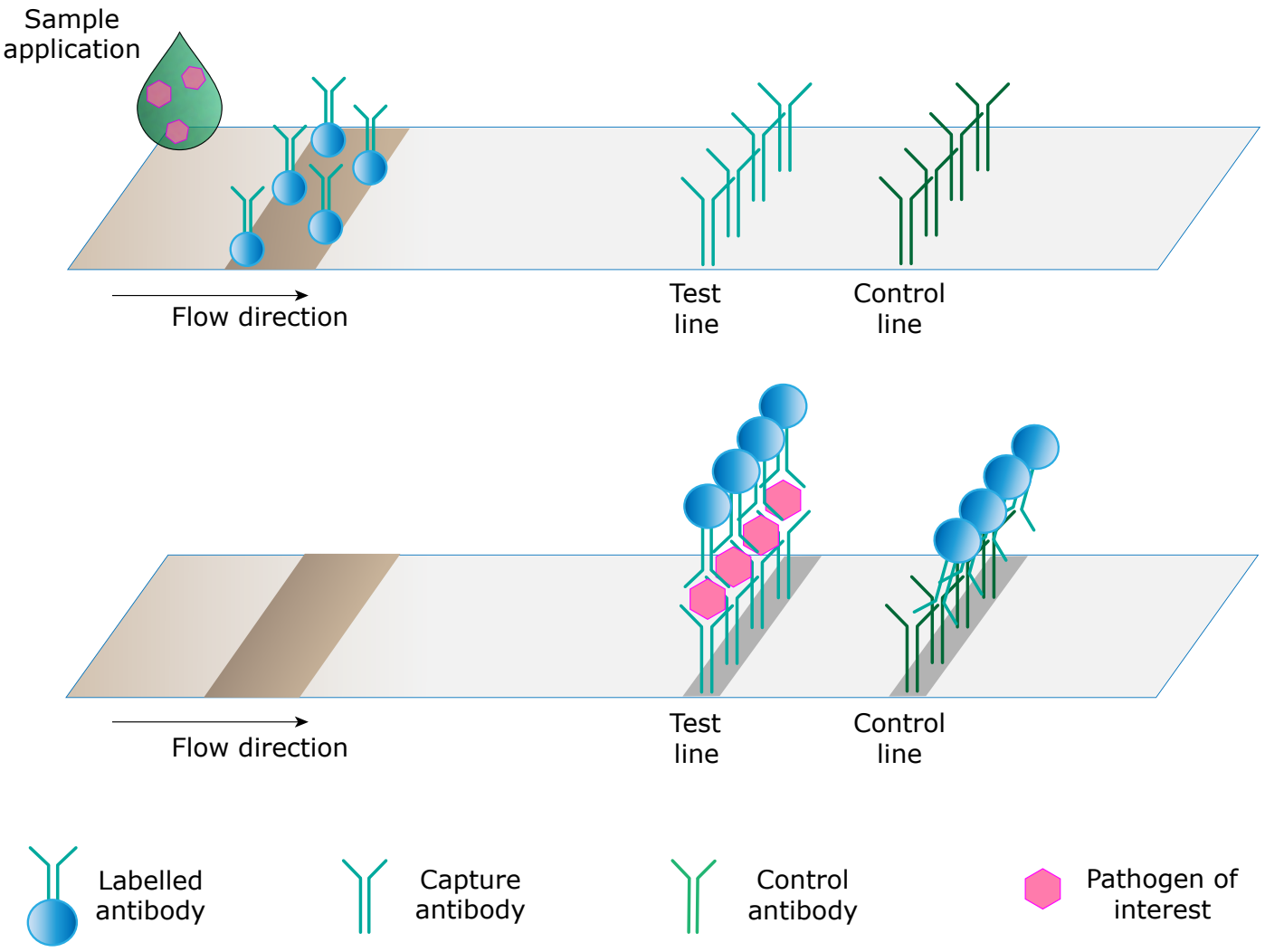
Indirect immunofluorescence	
Speed	●●●●○
Location	In the laboratory
Sensitivity	●●●●○
Number of pathogens per sample	1-2
Number of samples per test	18
Qualitative/quantitative	Quantatitive
Throughput	Medium
Costs	●○○○○
Added value of this platform	Morphological conformation

Lateral flow devices (LFD)

The Lateral Flow Device (LFD) test provides quick, typically within 10-30 minutes, qualitative results. It is an easy-to-use immunoassay for detecting viral, bacterial and oomycete plant pathogens without the need for specialized equipment, making them ideal for field use.

A sample is applied to the strip, where it encounters antibiodies that are labeled with a coloured marker. If the sample contains the pathogen, these antibodies will bind to it, forming a complex. As the sample moves further, the complex binds to a capture antibody on the test line, producing a visible colour change. The control line confirms the test worked properly. A visible test line at the test line region indicates the presence of the pathogen. If the pathogen is not present in your sample, no test line will appear, indicating a negative result.

Best used when a fast and easy-to-use test is needed, especially in field settings, though it offers a lower sensitivity and may miss low concentrations of pathogens.



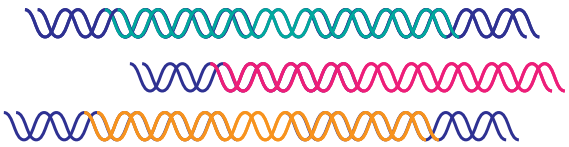
Luminex xTAG

Luminex xTAG is a multiplex PCR-based method for simultaneously detecting up to 50 plant pathogens in a single sample.

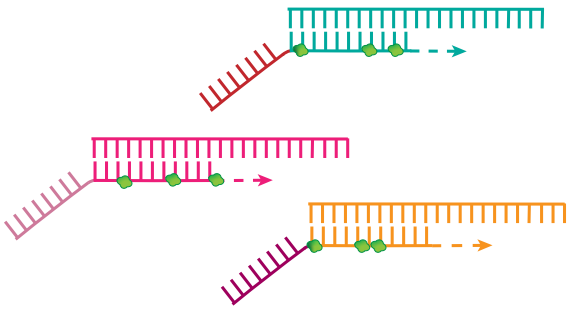
It starts with multiplex PCR, amplifying multiple DNA or RNA targets from a sample, enhancing sensitivity and enabling detection of various pathogens in one reaction. Target-specific primers are extended, incorporating biotin molecules, which bind to the amplified sequences. The biotinylated targets are then attached to colour-coded microspheres (beads), each specific to a target. These beads are analysed using a Luminex system, which measures fluorescence and identifies the target based on bead colour and signal intensity, enabling multiplexed pathogen detection.

Best used when high-throughput, accurate molecular detection of multiple plant pathogens is required, when for example no antibodies are available. Suited for complex, detailed pathogen profiling with high specificity.

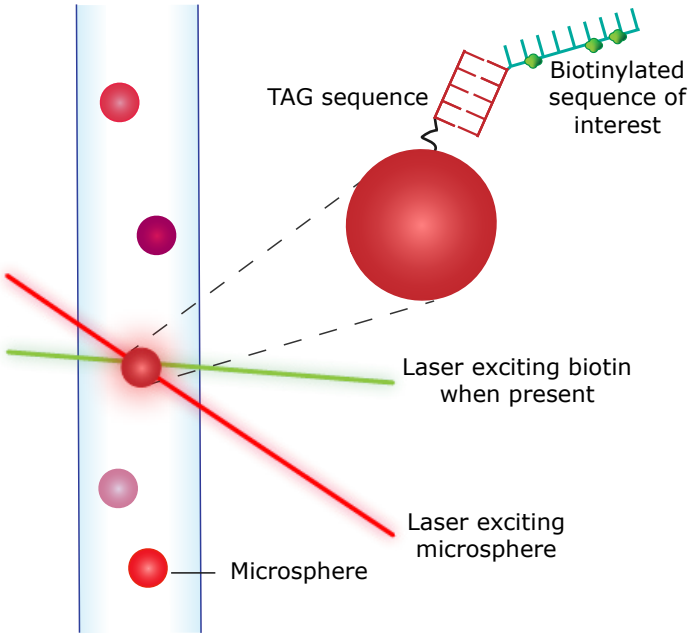
1 Multiplex PCR to pre-amplify multiple targets from DNA or RNA



2 Target Specefic Primer Extension (TSPE) and incorperation of (biotin)-dCTP



3 Measurement on Luminex machine

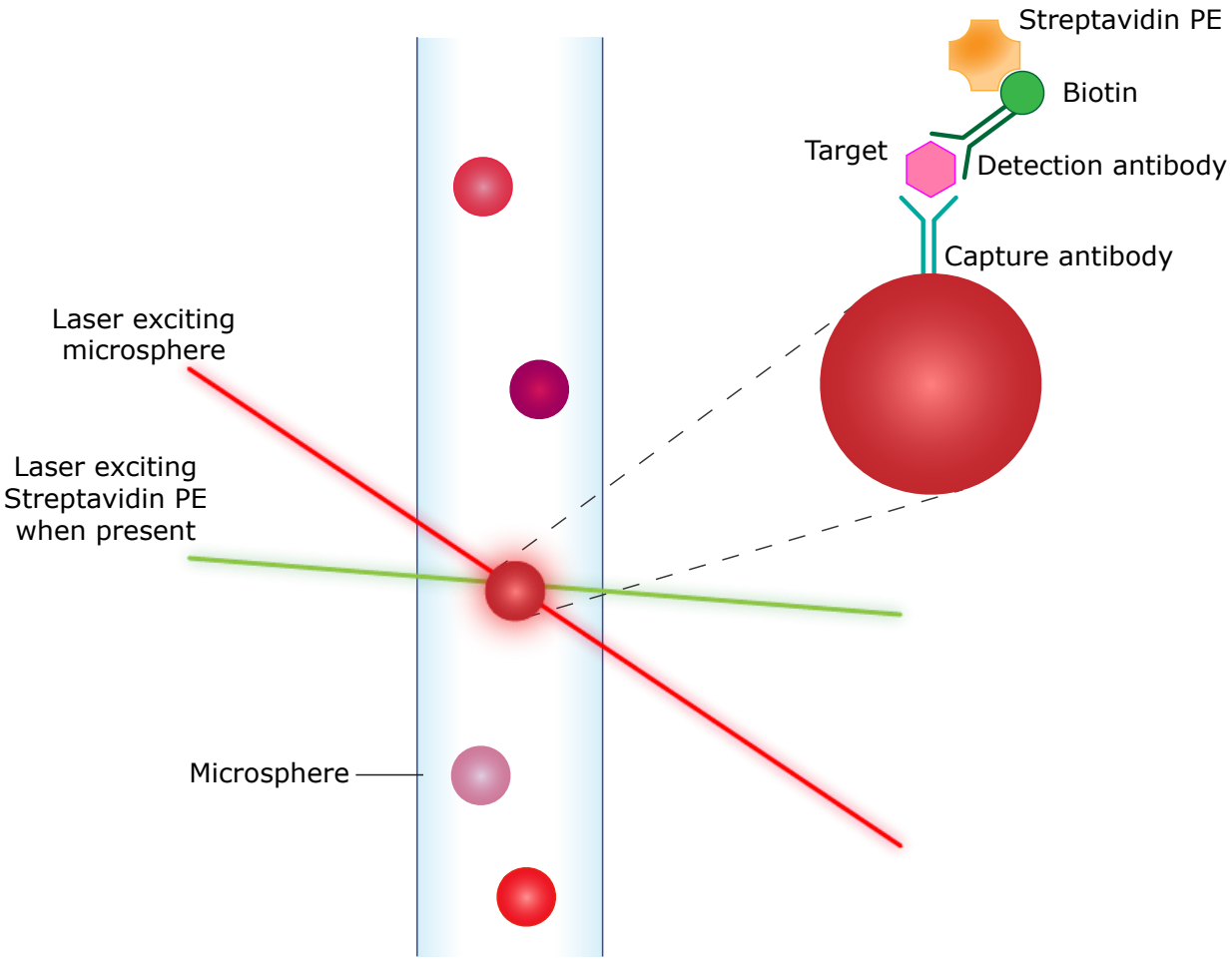


Luminex xMAP

Luminex xMAP is a multiplex assay platform that enables the simultaneous detection of multiple viruses and/or bacteria in a single sample.

This technology uses colour-coded microspheres (beads), each coated with a specific capture antibody that binds to the target pathogen's proteins. After adding the sample, the beads are analysed in a Luminex machine, which identifies the presence of pathogens based on the bead colour and fluorescence.

Best used when rapid, high-throughput, and precise detection of multiple pathogens simultaneously is needed, providing good sensitivity. Ideal for multiplex screening in plant disease diagnostics.

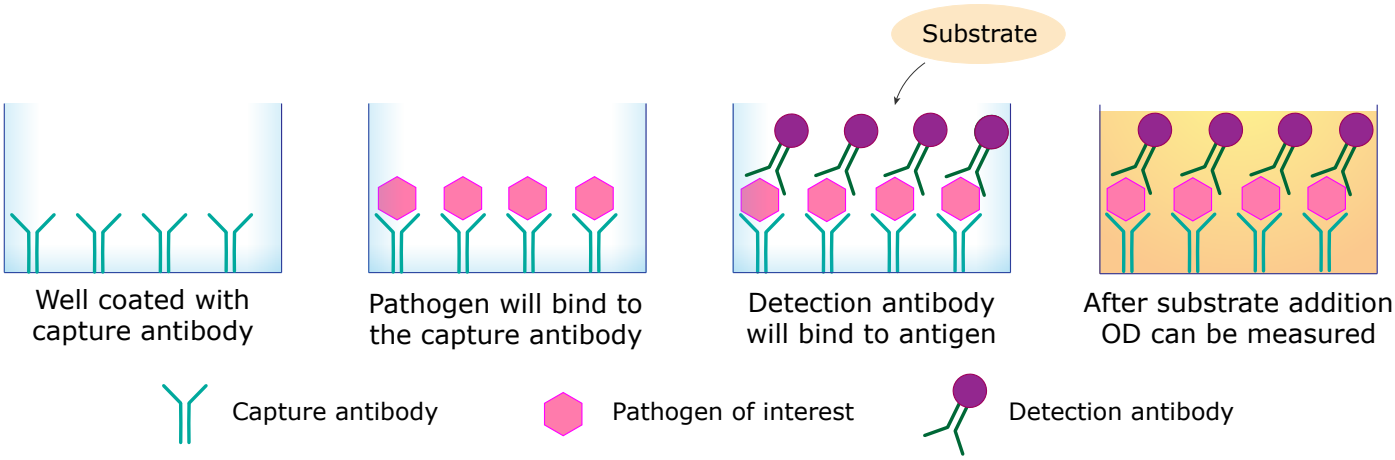


Double antibody sandwich (DAS)-ELISA

Double antibody sandwich (DAS-)ELISA is a sensitive and specific immunoassay that can be used to detect viruses and bacteria.

To start, a microplate is coated with a capture antibody, after which the sample is added. If the pathogen is present, it will bind to the capture antibody. Next, the detection antibody is added, which will bind to the pathogen if present. Together, the capture antibody and the detection antibody form a "sandwich" complex. After adding substrate, a colour change occurs if the pathogen is present.

Best used when a balance between sensitivity and cost is needed, although it is more time consuming and labor intensive than rapid tests.

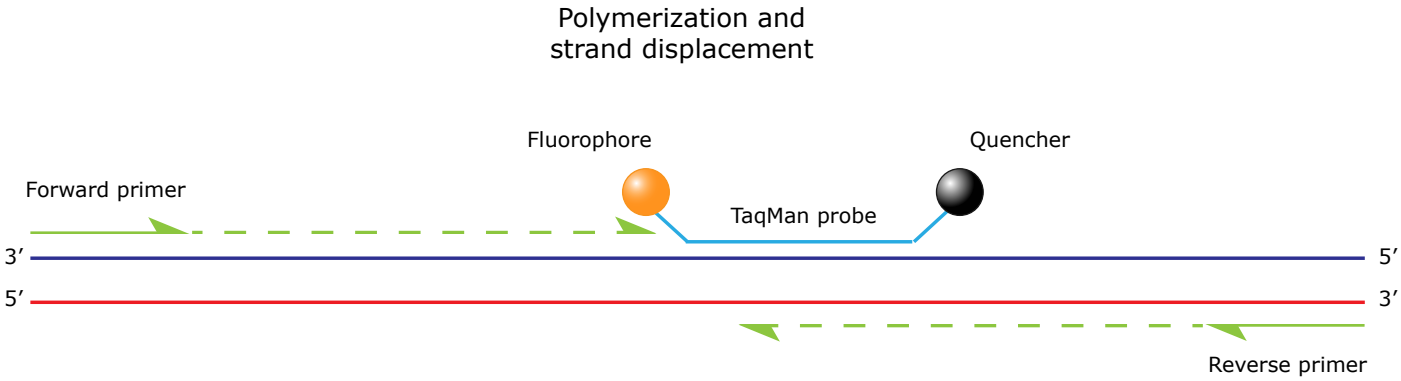


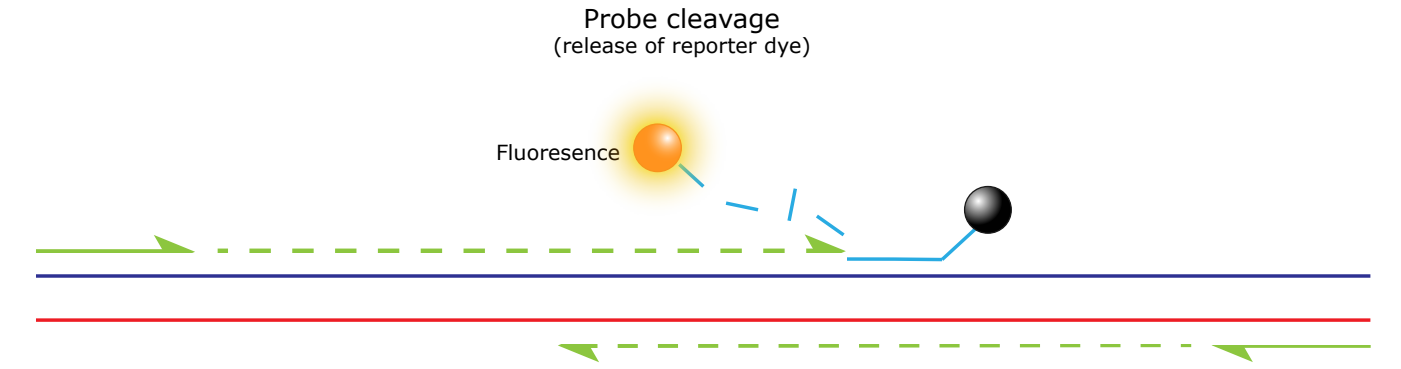
TaqMan

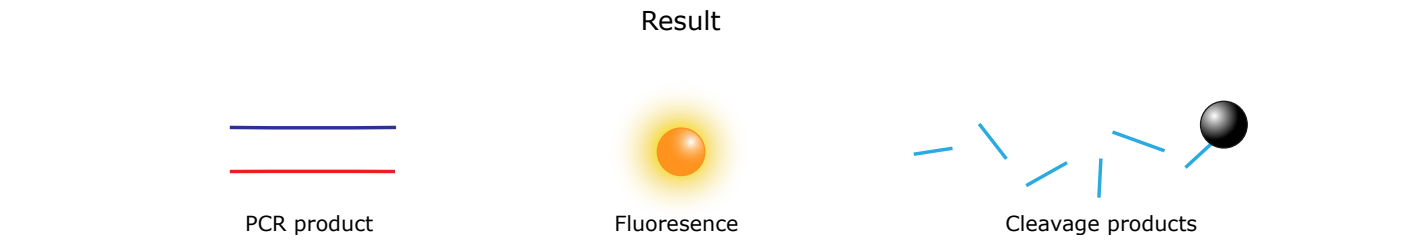
TaqMan PCR is a quantitative PCR technique used to detect and quantify plant pathogens. It can be used in a multiplex setting up to 6 targets.

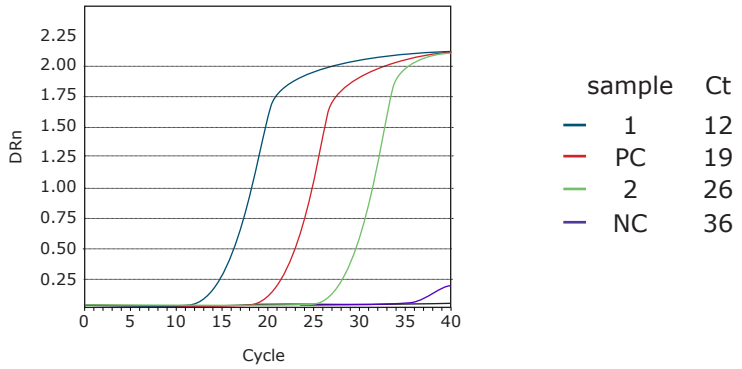
It involves amplifying the pathogen's DNA or RNA using primers and a fluorescent probe. The probe, labeled with a fluorophore and quencher, emits fluorescence when the DNA is amplified. This real-time detection allows for precise quantification of the pathogen's genetic material in a sample.

Best used when very high sensitivity and specificity are required for pathogen detection, especially for quantitative assays in diagnostic labs, where both speed and accuracy are key.







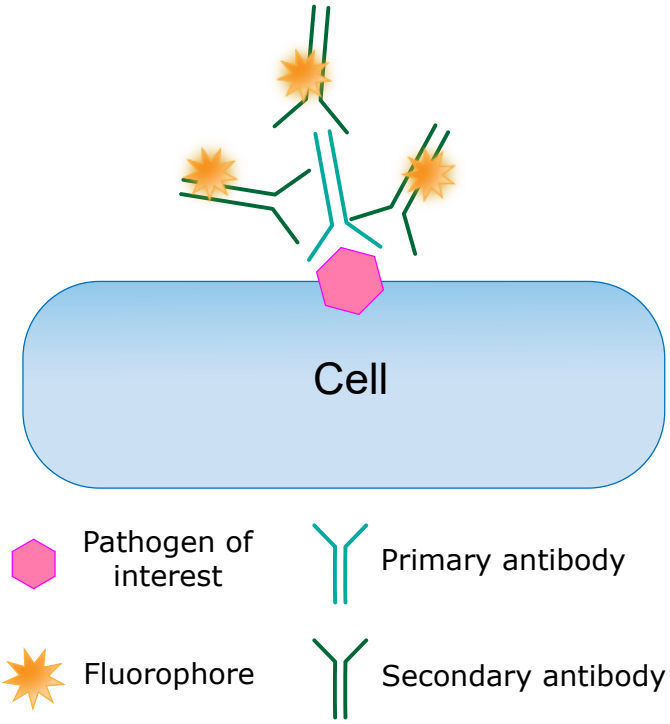


Indirect immunofluorescence (IIF)

Indirect Immunofluorescence (IIF) is a technique used to detect plant pathogenic bacteria by labeling antibodies with a fluorescent dye.

During IIF, a primary antibody will bind to the pathogen of interest in the sample. A secondary antibody, which is conjugated to a fluorescent dye, binds to the primary antibody. When the sample is exposed to UV light, the fluorescent marker on the secondary antibody emits light, which can be observed under a microscope. This light indicates the presence and location of the pathogen.

Best used when a highly sensitive method and localization of the bacteria is needed.



WAGENINGEN
UNIVERSITY & RESEARCH

primediagnostics

Droevendaalsesteeg 1
P.O. Box 16
6700 AA Wageningen
The Netherlands

email : primediagnostics@wur.nl
web site : www.primediagnostics.com
web shop : shop.wur.nl/primediagnostics