



# ACIAR review Cassava point-of-care diagnostics CMD & CWBD

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# DNA/RNA based diagnostic methods (Amplification)

## I. Sampling

- i. Isolation of nucleic acids
- ii. Needs to quantify
- iii. Needs expertise

## II. Amplification

- i. Needs thermocycler
- ii. Needs reliable power supply
- iii. Needs a laboratory

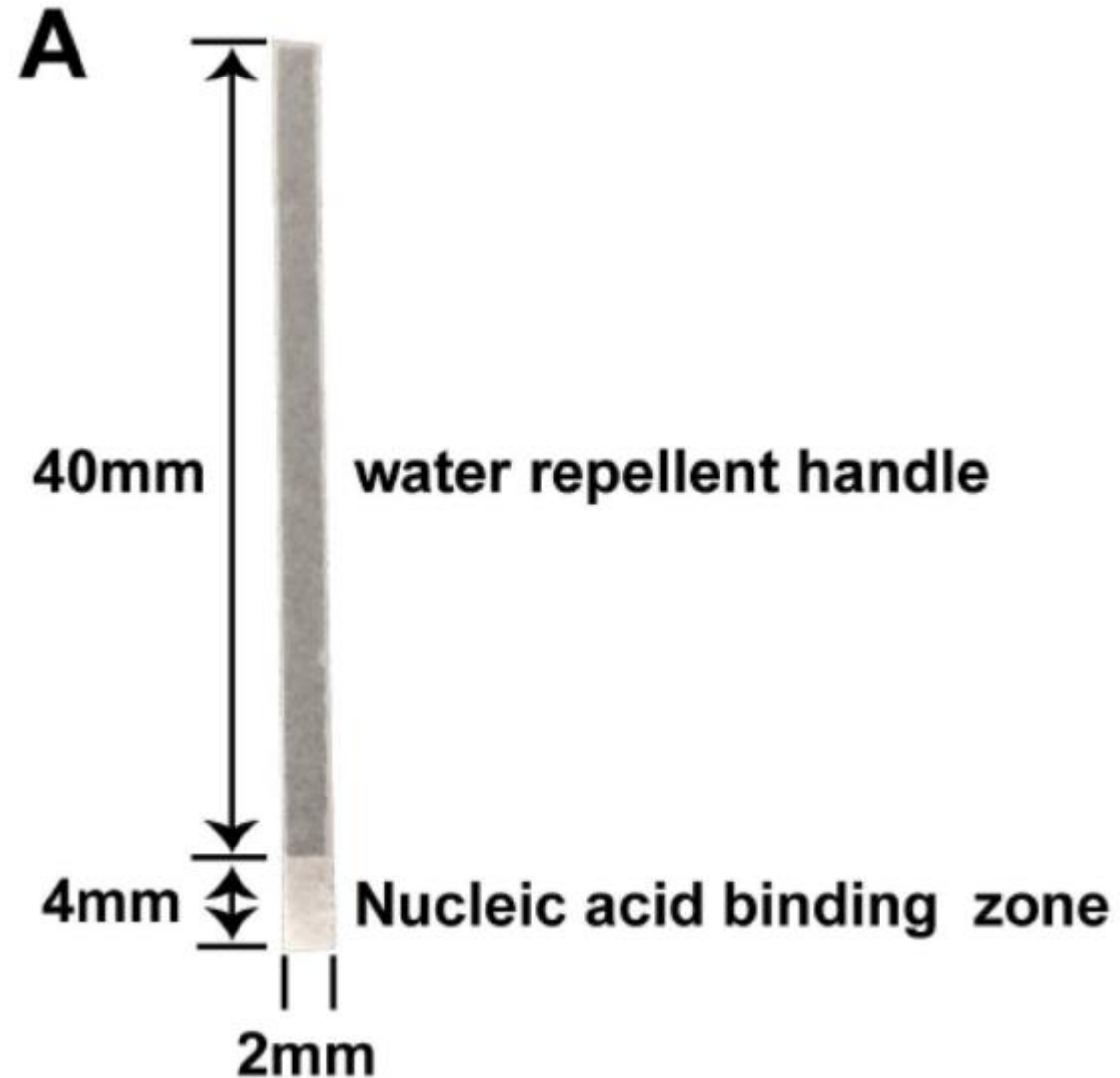
## III. Visualization of results

- i. Needs equipment
  - a. Electrophoresis
  - b. Fluorescence



# Sampling: DNA dipstick

DNA dipstick

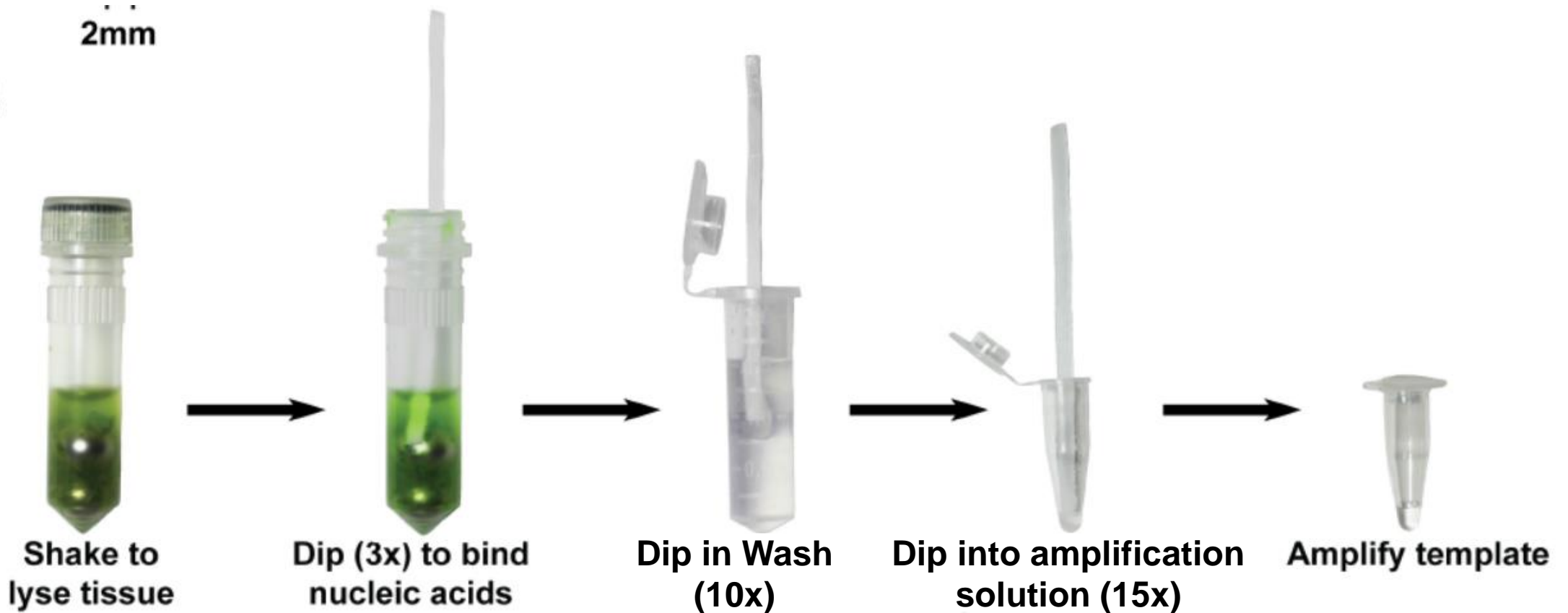




# Sampling: DNA dipstick

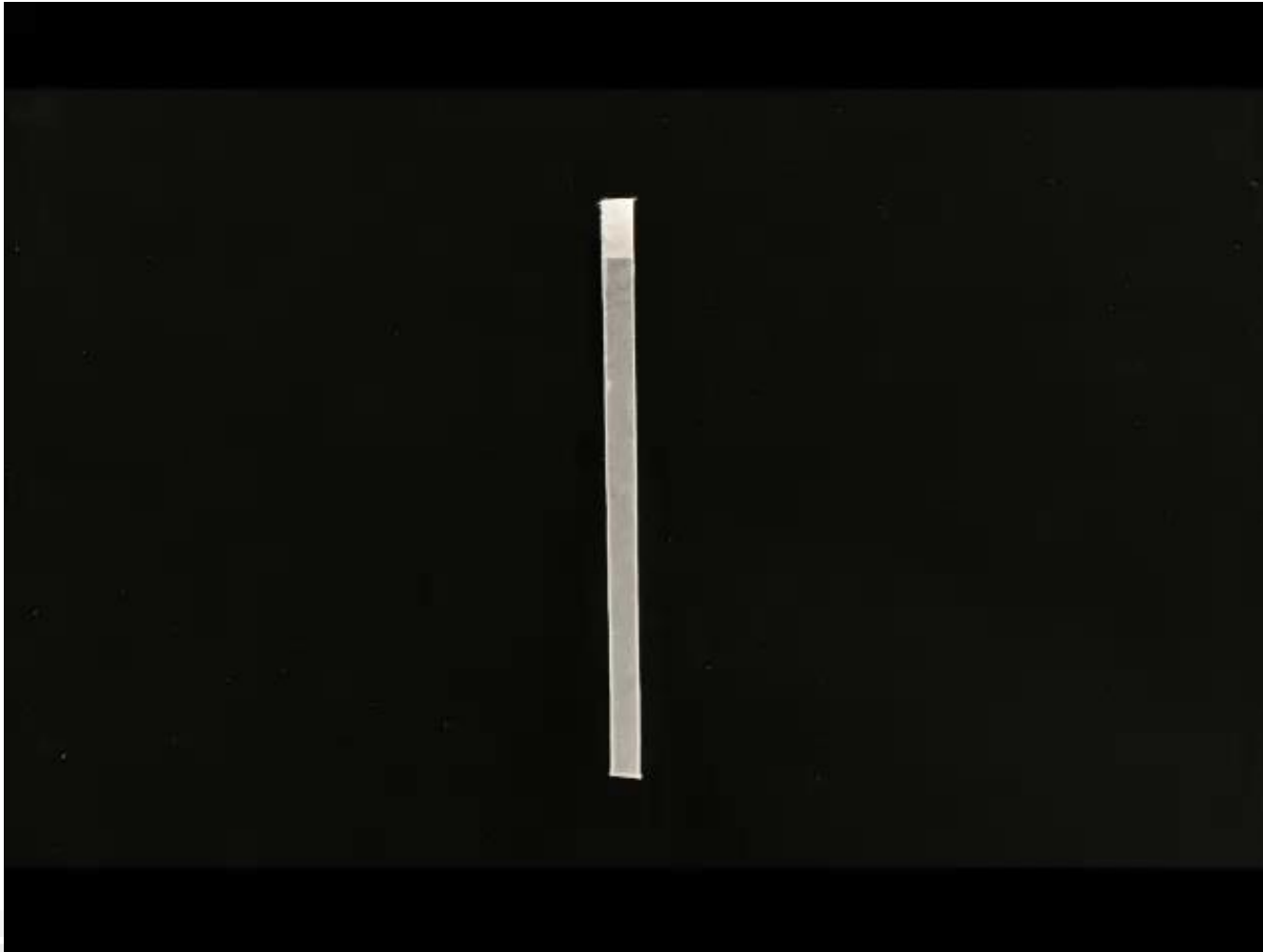
**B**

2mm





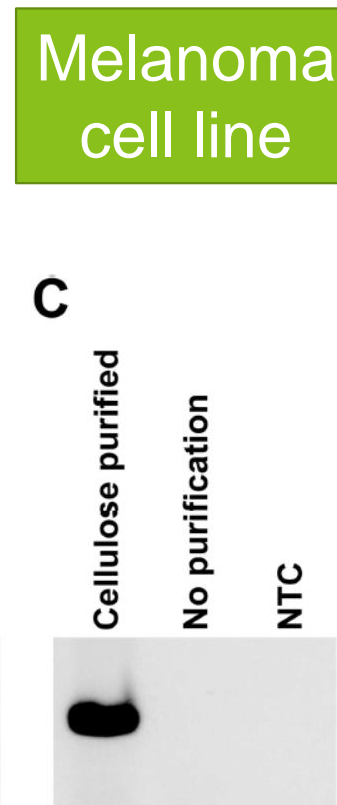
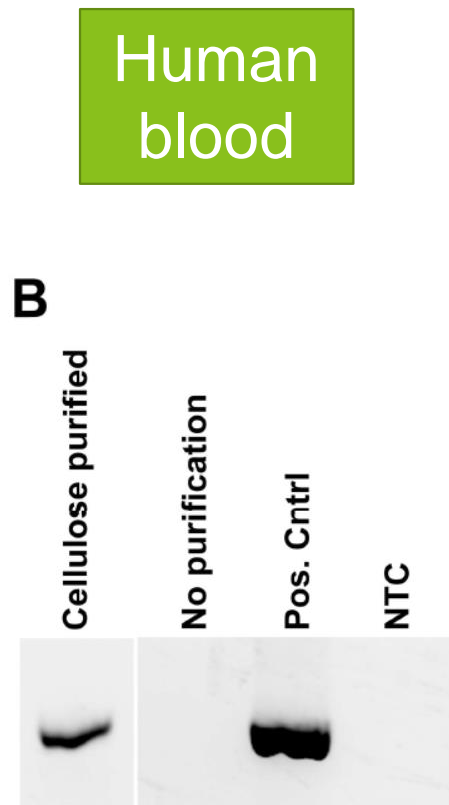
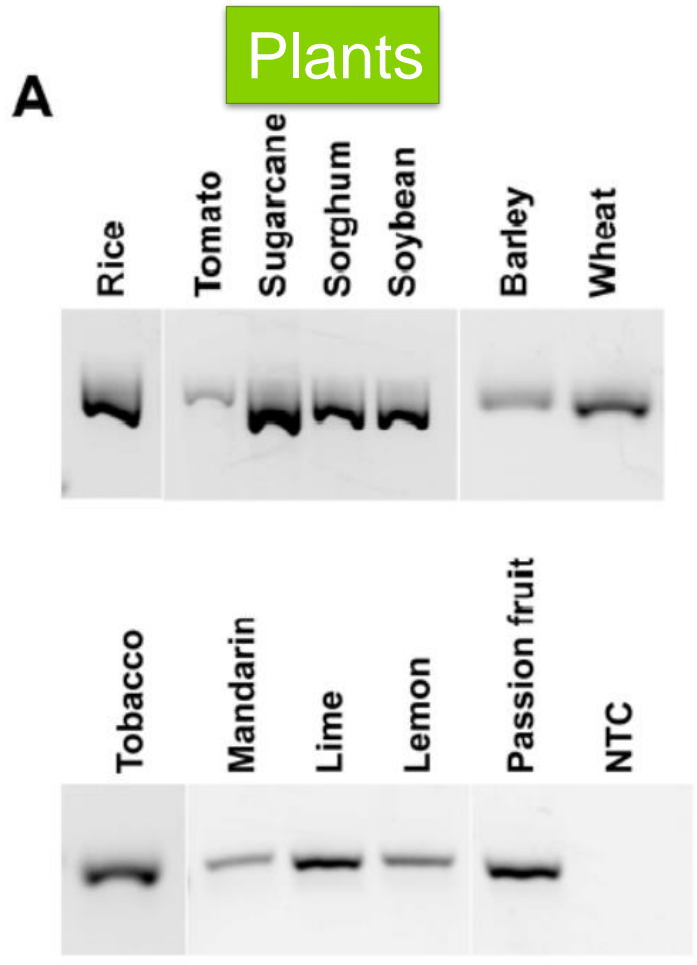
# Sampling: DNA dipstick





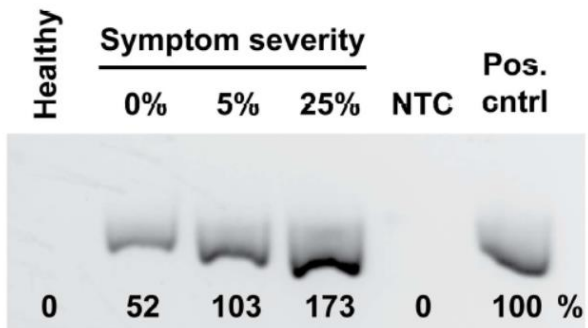
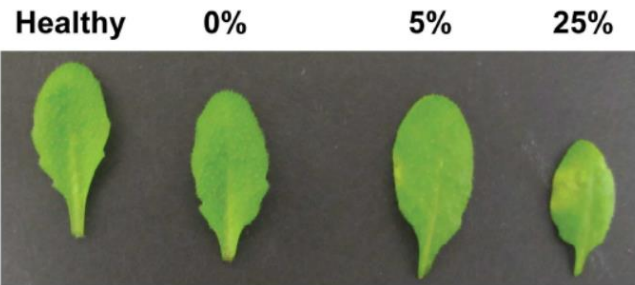
# Dipstick: Crops and human samples

## DNA dipstick

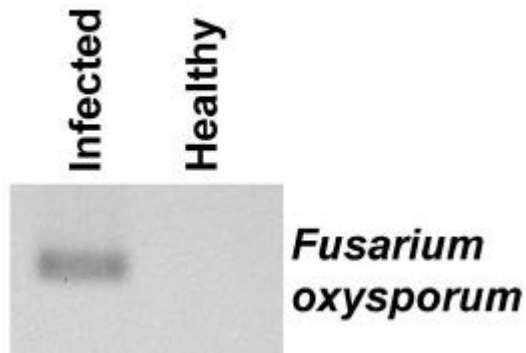


# Dipstick: plant and animal pathogens

*Pseudomonas Syringae*

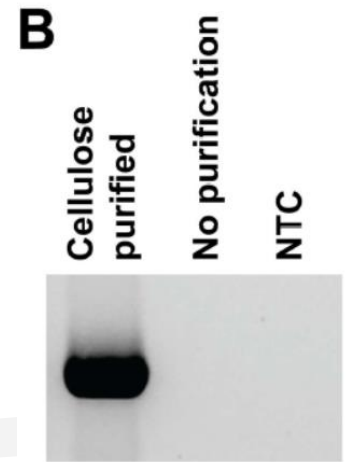


*Fusarium oxysporum*

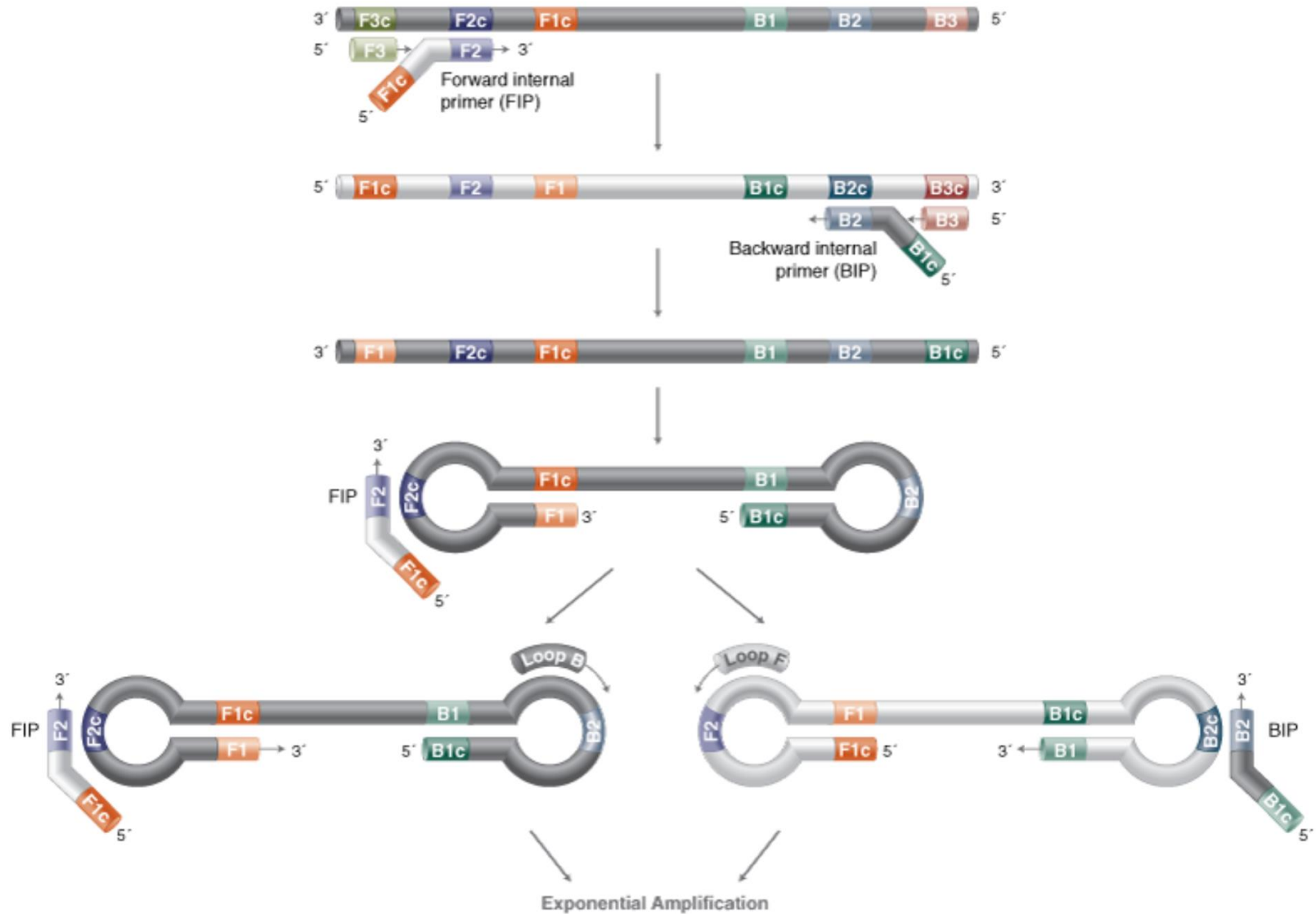


*Actinobacillus pleuropneumoniae*

Pig lung swab

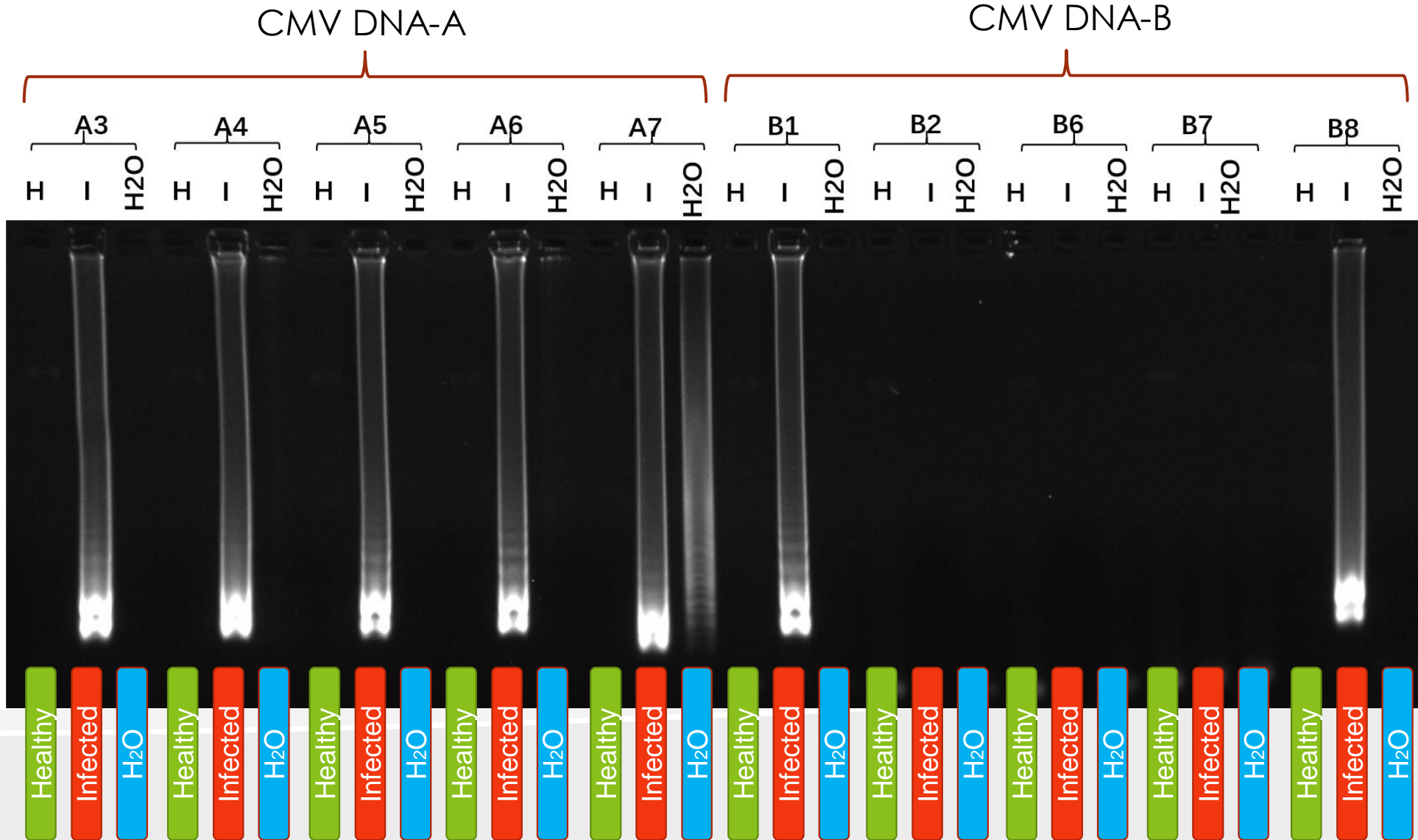


# LAMP amplification



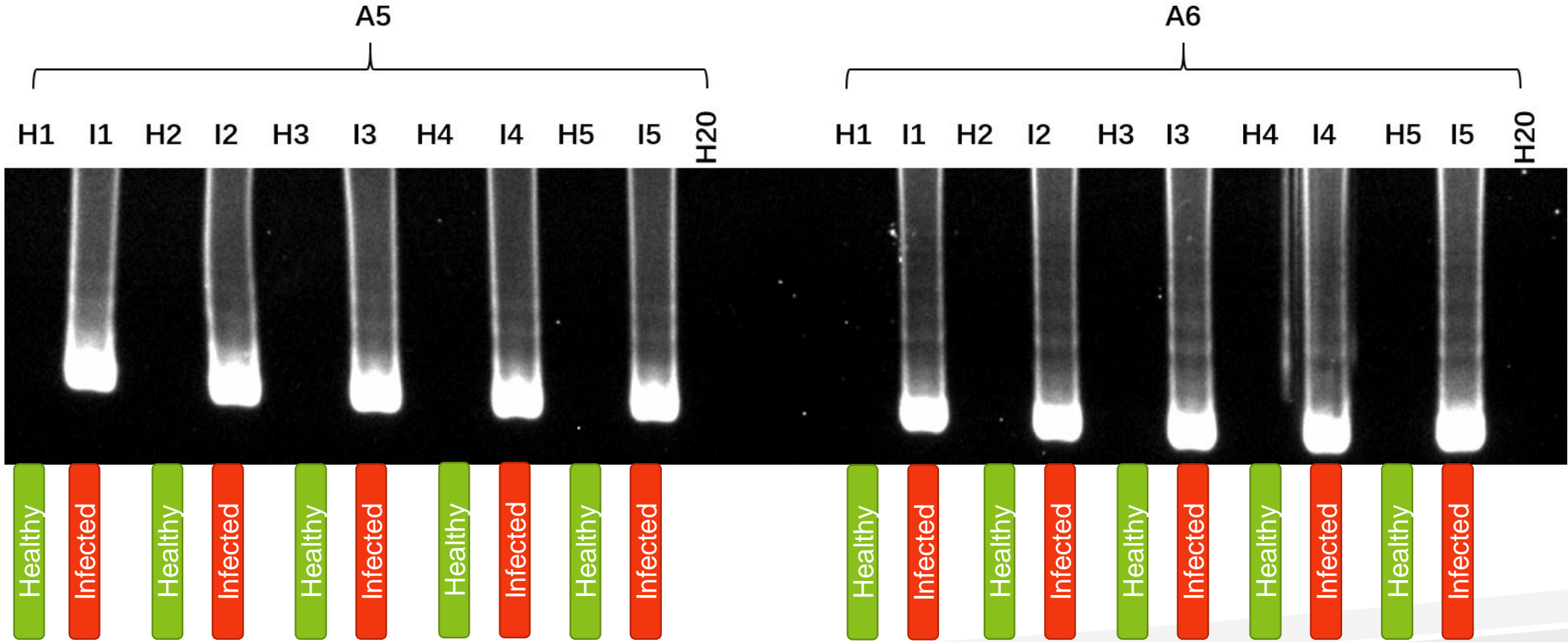


# Identification of suitable LAMP primers for CMV



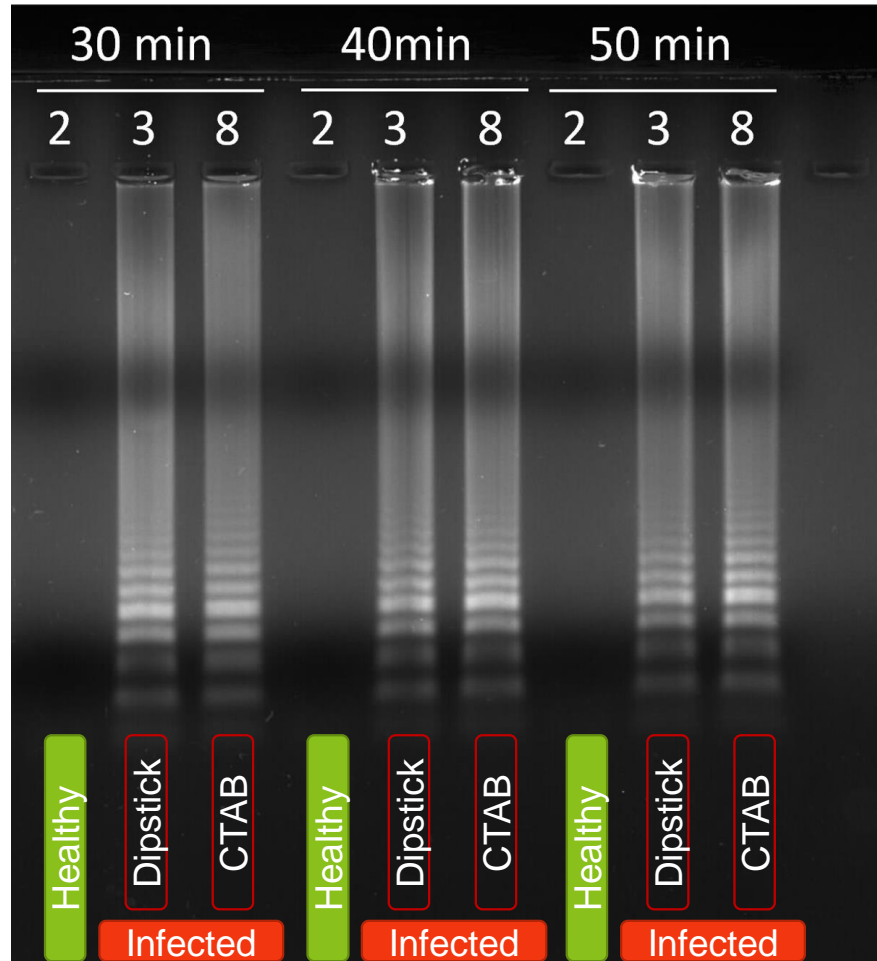


# Reliability CMV



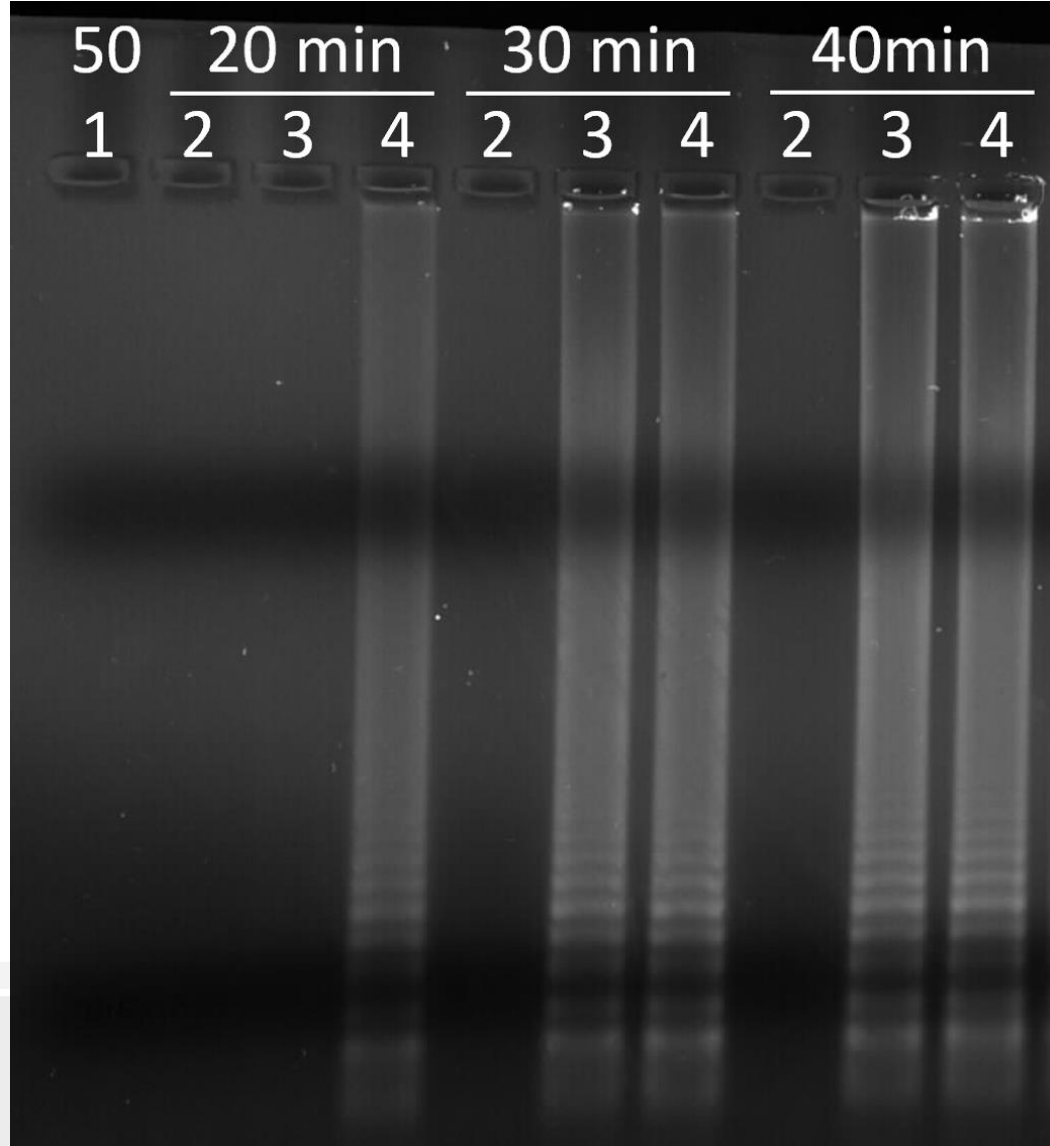


# Dry leaves – Comparison to CTAB purified DNA





Dry leaves



## Comparison to PCR

Asymptomatic sample produced a very weak band by PCR (50% of the time) using CTAB purified DNA



# Stems-sensitivity

Healthy

1:20

1:40

1:80

1:160

1:320

1:320

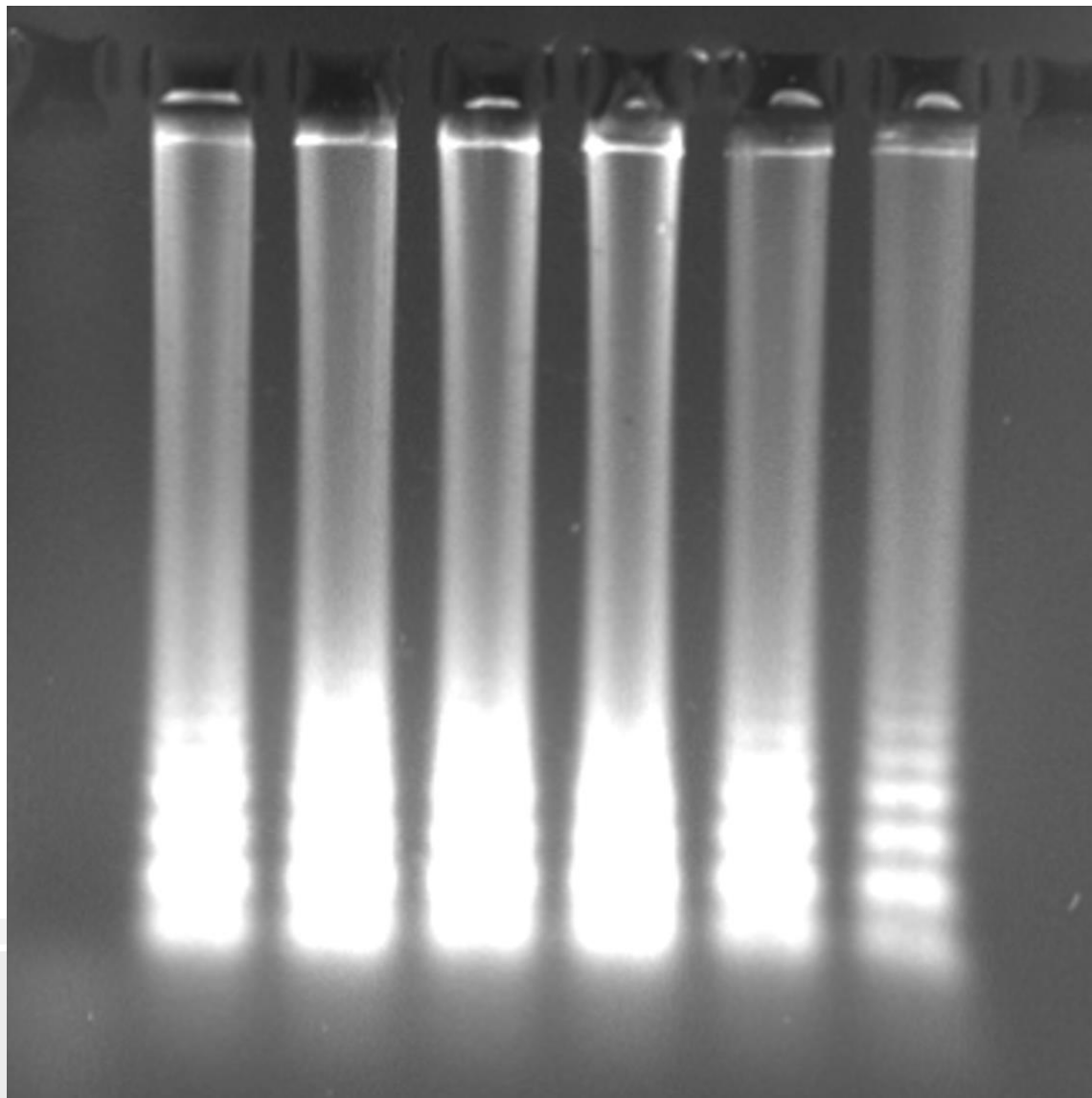
1:640

Infected



Easy Return  
Mini Cheese Dishwasher Safe Stainless

**USD 0.39**



Clean seed systems



# Sampling: DNA dipstick





# PLOS BIOLOGY

advanced sea

 OPEN ACCESS  PEER-REVIEWED

METHODS AND RESOURCES

## Nucleic acid purification from plants, animals and microbes in under 30 seconds

Yiping Zou , Michael Glenn Mason  , Yuling Wang, Eugene Wee, Conny Turni, Patrick J. Blackall, Matt Trau, Jose Ramon Botella 

Published: November 21, 2017 • <https://doi.org/10.1371/journal.pbio.2003916>

1,143  
Save

186  
Citation

148,425  
View

1,994  
Share



# Amplification & Visualization

## Diagnostic Droid





# Diagnostic Droid

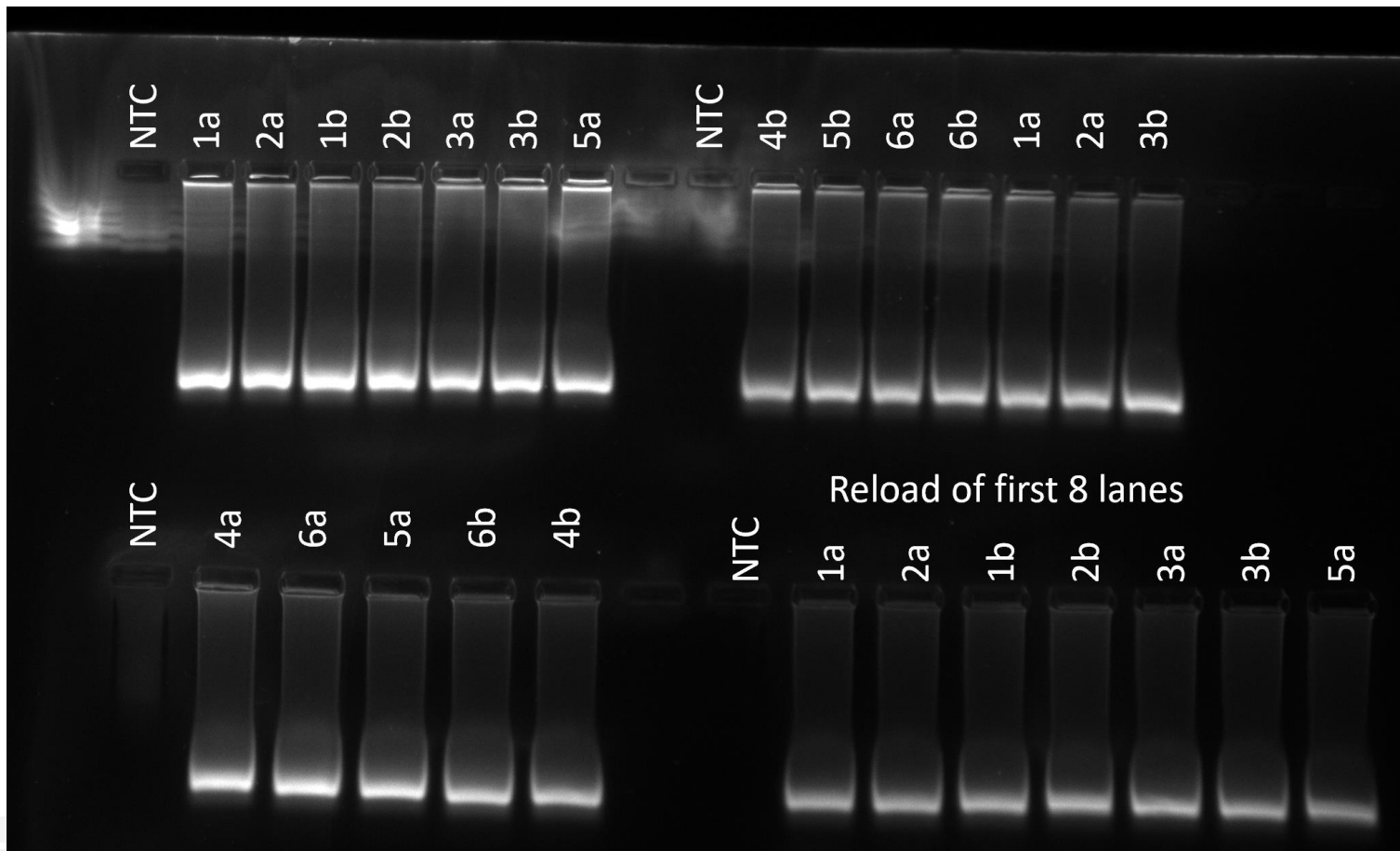
1. Analyses up to 8 samples
2. Heated lid
3. LED screen for communication
4. Data is recorded by the machine, interpreted and a diagnosis is provided in the screen
  1. Instant message of positive sample
5. SD card record data for further analysis





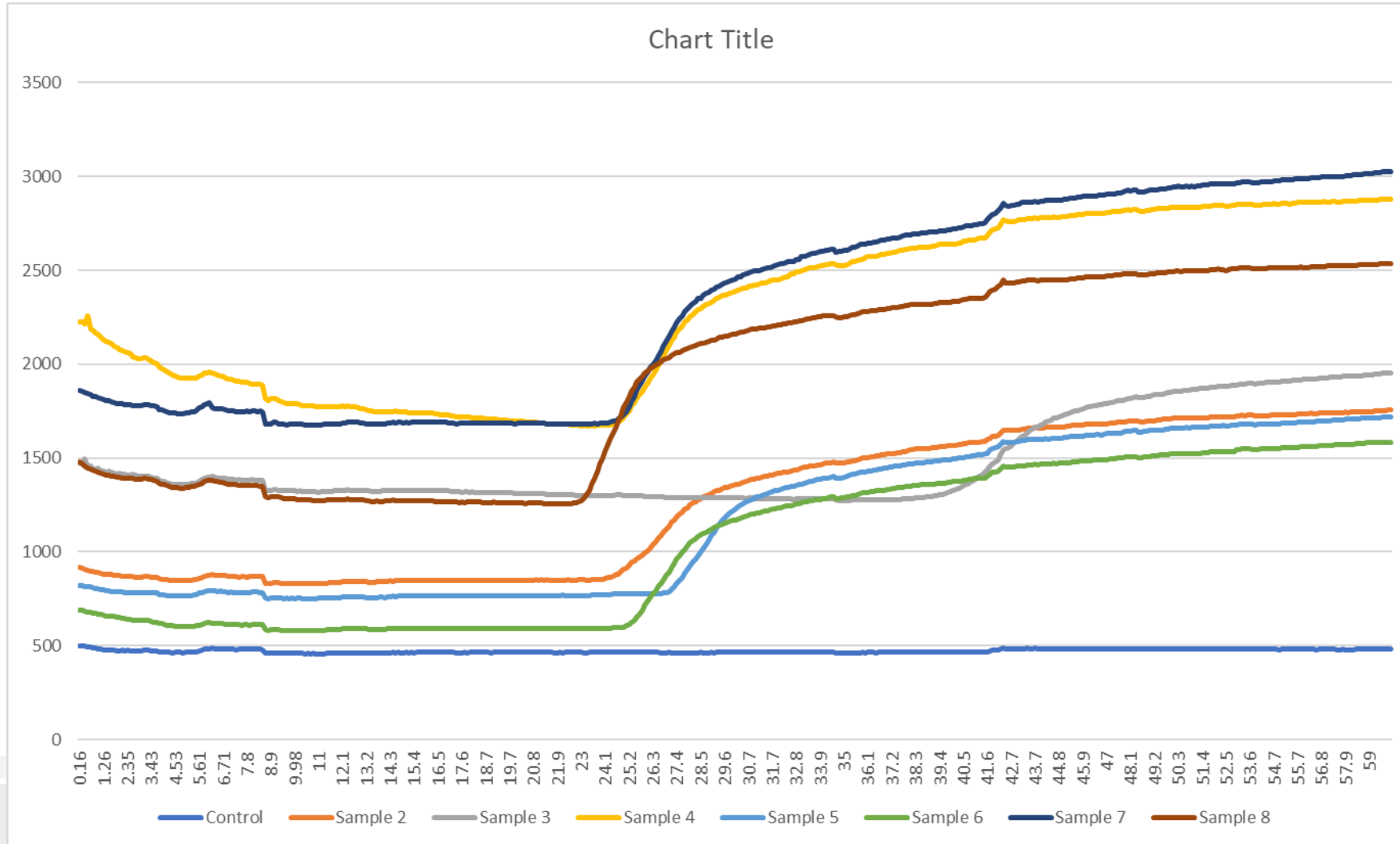


# Workshop results 26/Sept/2023





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## CWBD

- Developing and testing LAMP primers
- Promising results but need confirmation
  
- Could be applied to Vascular Streak Dieback in cocoa



Questions?