

User Manual

SignalChem Diagnostics

Pythium myriotylum One-Pot Detection Kit

To enable point-of-care testing for the immediate detection of *Pythium myriotylum* in plant samples

Catalog # PM01-K199 (Starter Kit)

Catalog # PM02-K199 (Standard Kit)

Diagnostic Reagents For Accurate And Reproducible Results

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Introduction

Testing kit designed for early rapid detection of pathogens in plants. The kit is self sufficient, so there is no need for the end user to send samples elsewhere for analysis. The kit is designed to work with an instrument (CRISPOT[®] Detection Device, Cat# CSP01-V19A, SOLD SEPARATELY), which screens for the presence of a portion of the pathogen's genome in all samples, thus identifying infected plants. It is important for the end-user to label the samples according to the plants they were extracted from to avoid confusion. The user is expected to spend around 10 minutes on sample preparation and setting up the device. Once set-up is complete, running the test on the detection device will take 25 minutes.

Disclosure

This product is not to be used as a drug, medical device, food additive, cosmetic, or household chemical. It is not to be used in therapeutic, consumer, or pesticidal applications. Not for resale.

Support

For inquires and assistance, please contact us at:

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Version History

Version 2.0

Updated: Aug 28, 2026

Table of Contents

Kit Components..... 4

Kit Components: Visual Guide..... 5

Storage and Precautions..... 6

Sample Preparation 7

Sample Preparation: Visual Guide..... 9

Kit Components

Starter Kit-Only Components

#	Component	Amount
1	Tube Rack	1
2	Adjustable Pipette	1

Not Provided

Gloves
Bleach

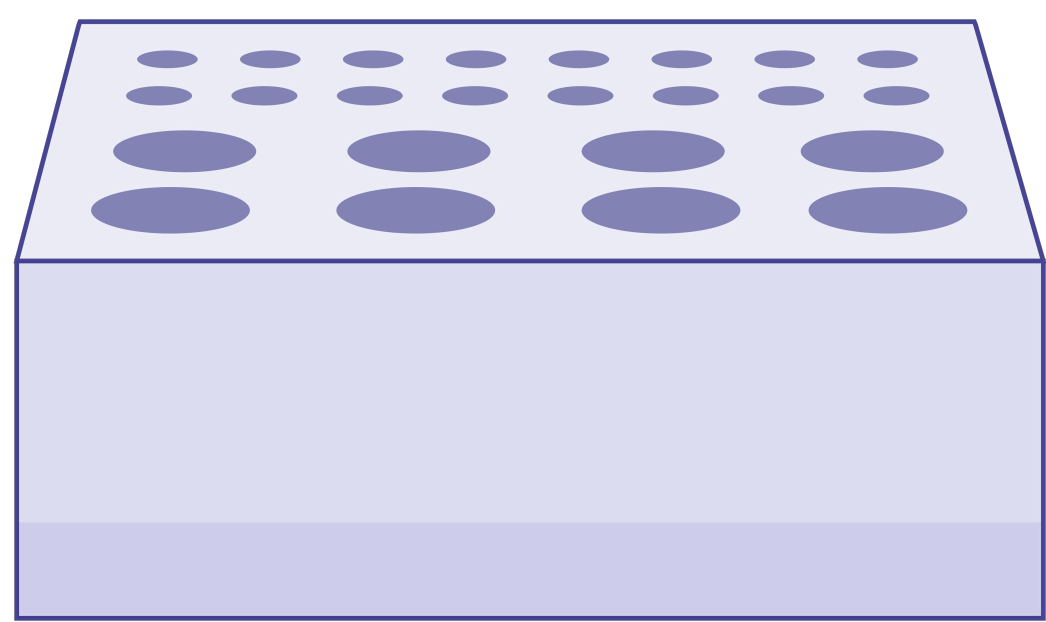
Standard Kit Components

#	Component	16 Reactions	32 Reactions	48 Reactions
3	Pipette Tips	35	70	105
4	Grommets	16	32	48
5	Grinding Rods	16	32	48
6	Lysis Buffer	16 tubes	32 tubes	48 tubes
7	Reaction Buffer	16 tubes with red cap	32 tubes with red cap	48 tubes with red cap
8	PCR Tube Strips With Master Mix	2 (8-well) strip tubes	4 (8-well) strip tubes	6 (8-well) strip tubes
9	User Manual	1	1	1

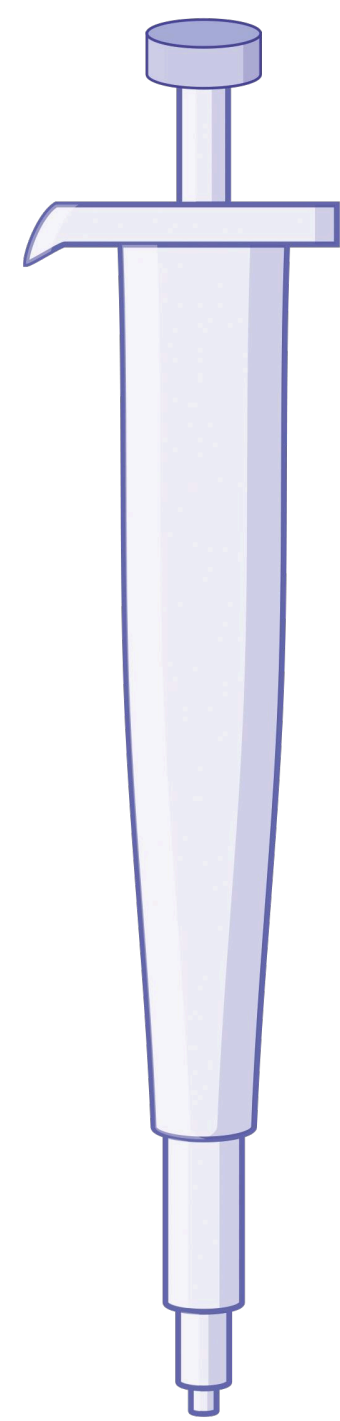
Kit Components: Visual Guide

Starter Kit-Only

1 Tube Rack

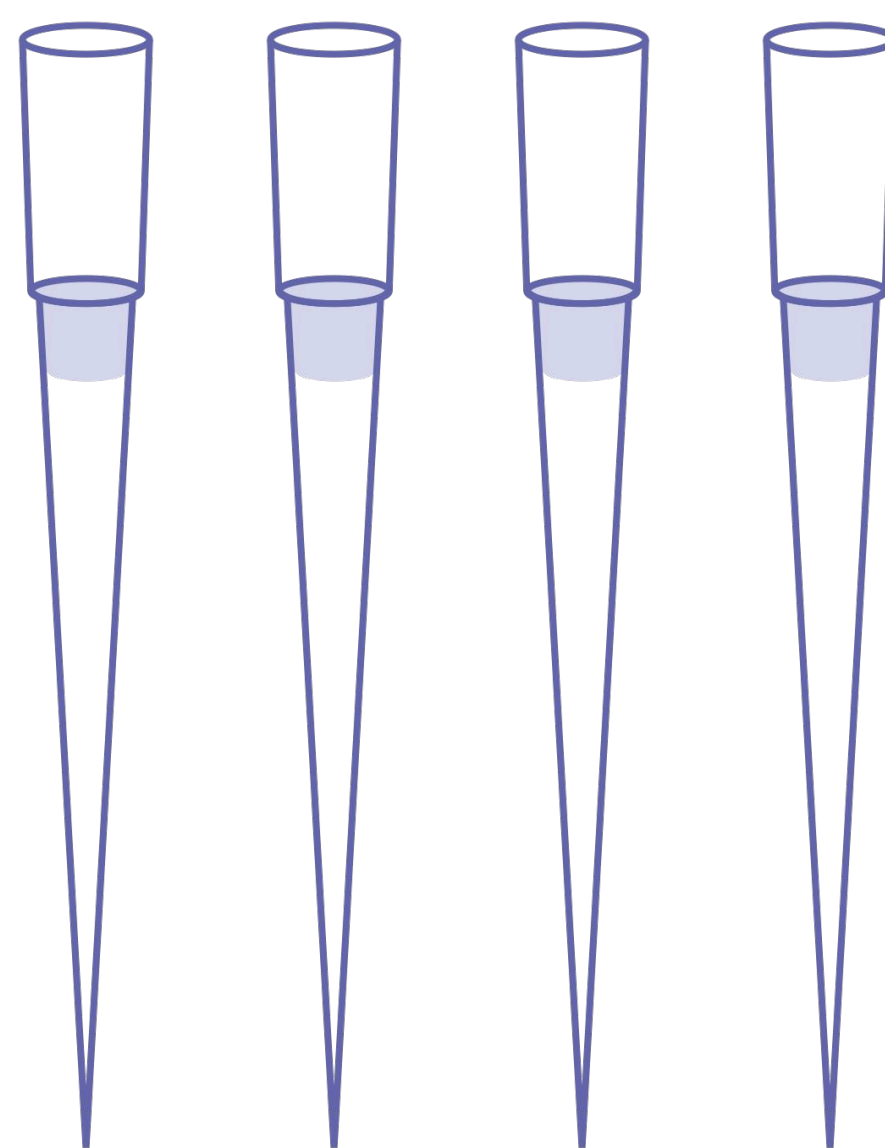


2 Adjustable Pipette

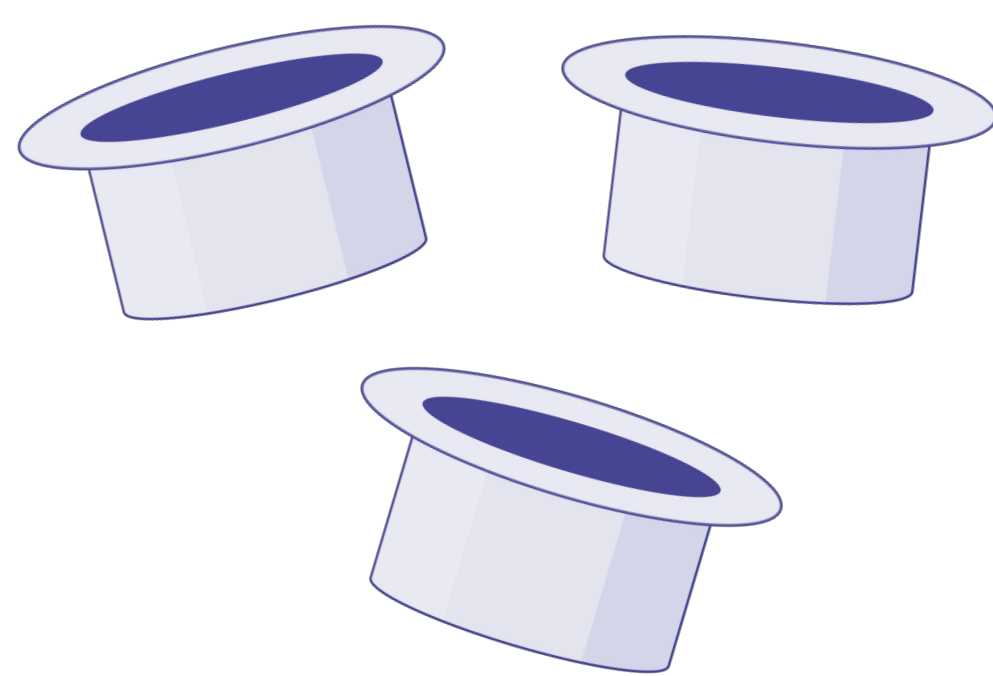


Standard Kit

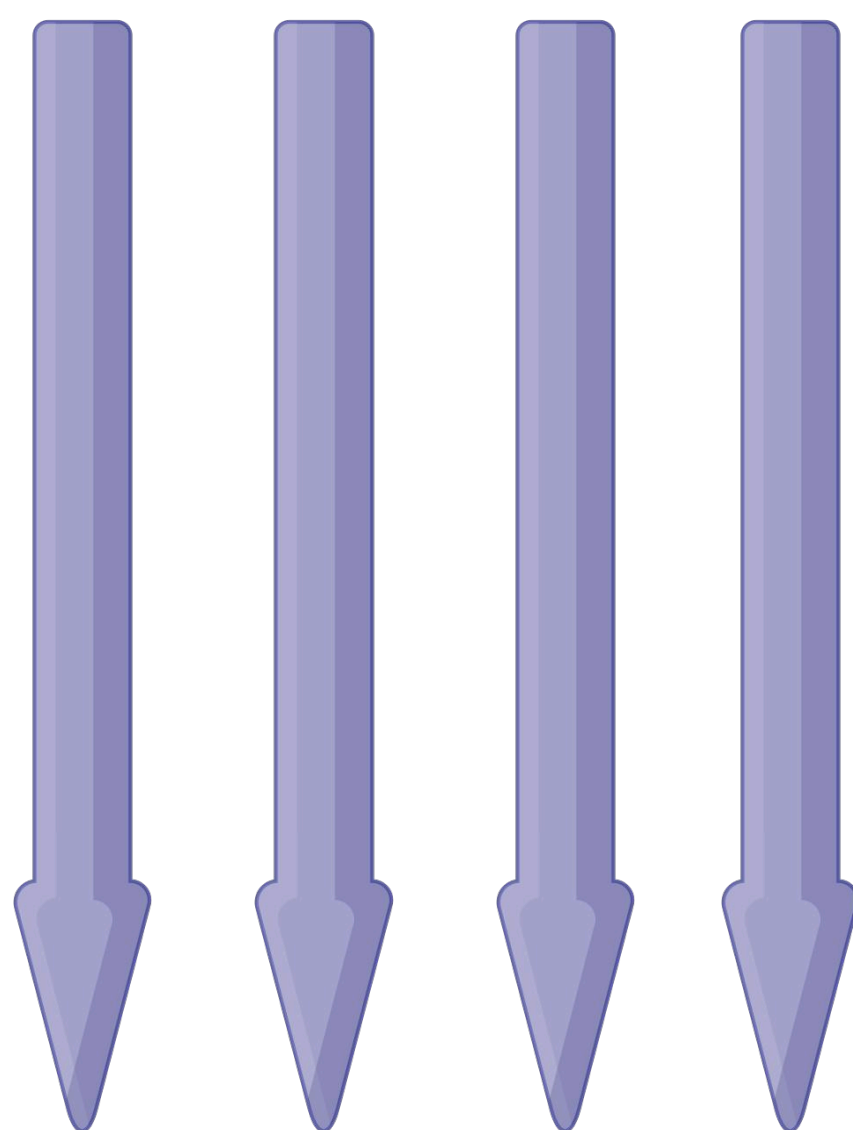
3 Pipette Tips



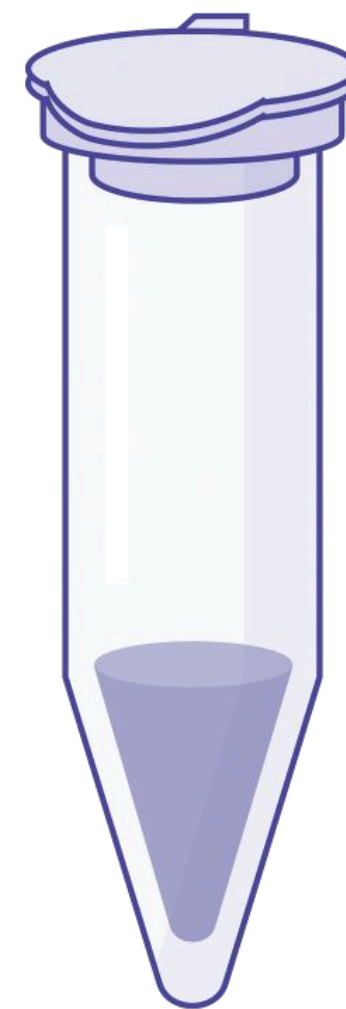
4 Grommets



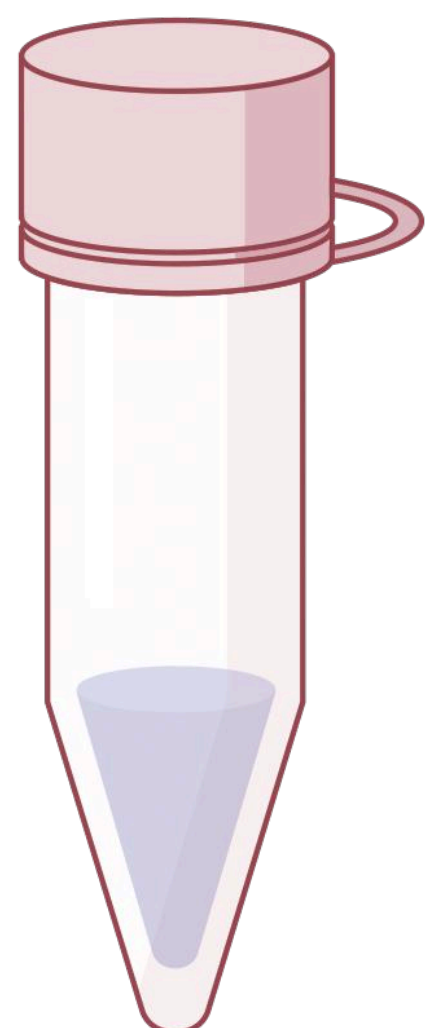
5 Grinding Rods



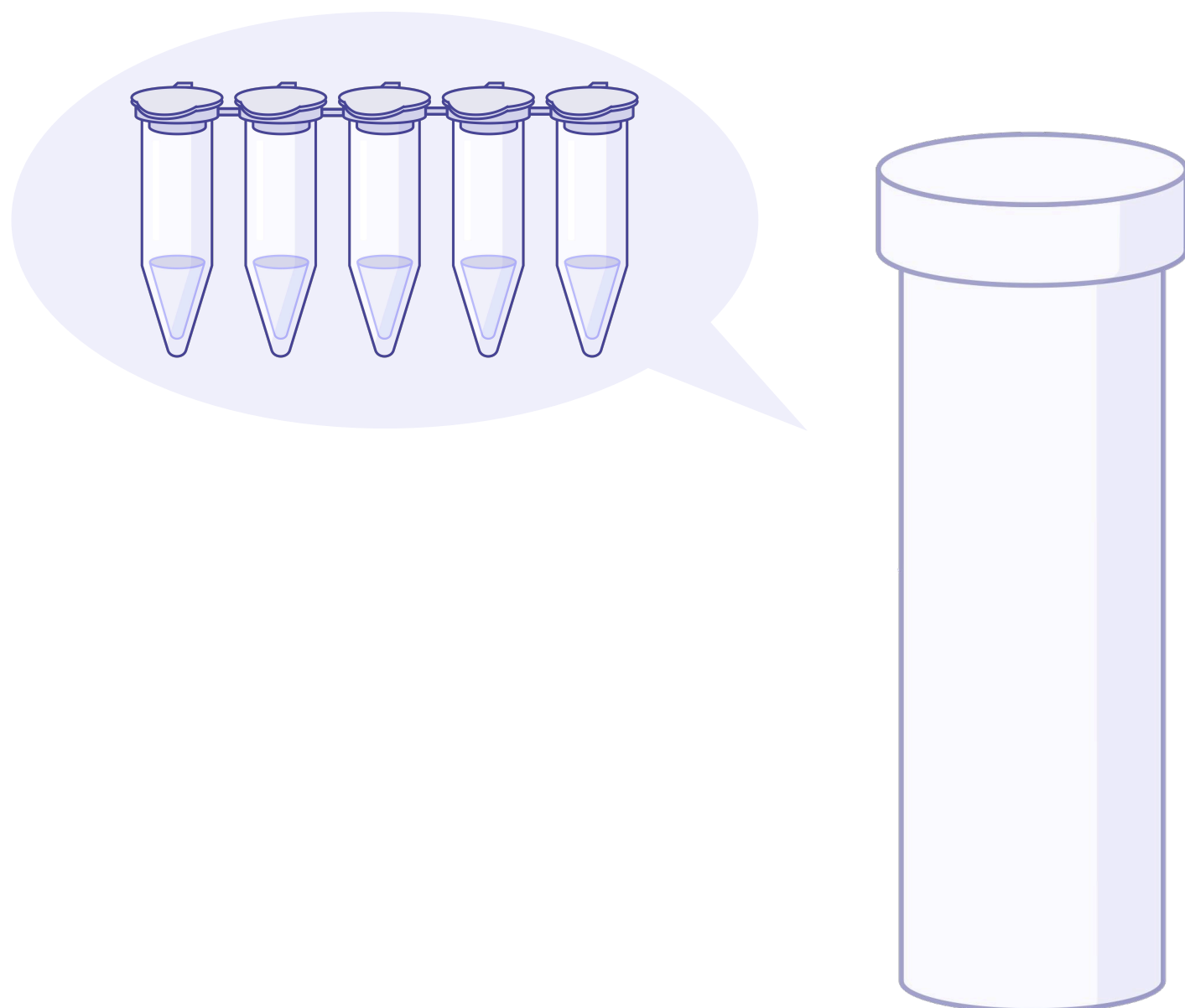
6 Lysis Buffer



7 Reaction Buffer



8 PCR Tube Strips With Master Mix in White Plastic Tube



9 User Manual



Storage and Precautions

Kit Storage

Store all kit components at 2-8°C for up to 12 months or at 8-25°C for up to 6 months.

The Master Mix in the PCR tubes (component 8) should be protected from light. The tube with desiccant cap offers light and moisture protection as long as the tube is tightly closed. Always keep the strips in the tube when not in use, as exposure to light can cause deterioration and lead to inaccurate results.

Precautions

- Ensure the working environment is clean before starting the experiment.
- Always wear a new pair of examination gloves when handling samples and performing the assay.
- Remove the appropriate number for tubes from the strip tubes containing the buffers and Master Mix based on the number of samples being tested.
- The presence of inhibitors can lead to invalid or false-negative results.
- Mutations in the target regions of the *Pythium myriotylum* genome, covered by the primers in this kit, may prevent the detection of the pathogen.
- Adhere to good laboratory practices to ensure proper use of this kit. Keep both the kit and reactions free from contamination. Carefully inspect all reagents for signs of contamination, and do not use any reagents that appear contaminated.

Sample Preparation

Protocol

1. Prepare Samples and Materials

- Determine the number of samples to test. Label each plant and record sample order.
- For each sample, collect the required components from the kit. If necessary, separate the needed number of tubes from the PCR tube strips:
 - Grommet ④
 - Lysis Buffer Tube ⑥
 - Reaction Buffer Tube ⑦
 - Master Mix Tube ⑧

2. Collect and Load Samples

- Support the leaf with a paper towel on a hard surface.
- Using a clean grommet ④, collect 3-5 leaf punches from the margins of the infected area.
 - Avoid heavily decayed tissue, excess soil, and moisture.
 - Including a small amount of brown tissue is acceptable.
- Place tissue directly into a lysis buffer tube ⑥ and grind with a grinding rod ⑤ for 30-60 seconds.
 - Use the pointed end of the grinding rod ⑤ to eject the tissue.
 - Do not exceed recommended amounts.

3. Add to Reaction Buffer

- Set pipette ② to 5 µL and attach a clean tip ③.
- Open one tube at a time, transfer 5 µL of the lysate from Step 2 into reaction buffer tube ⑦, then close and mix by shaking.
- Discard tip after each sample.

Important: Use a new tip for every sample and avoid opening multiple tubes at once.

4. Add to Master Mix

- Set pipette ② to 30 µL and attach a new tip ③.
- Transfer 30 µL of sample into master mix tube ⑧ without touching the pellet.
- Close tube, discard tip, and mix by flicking to fully resuspend pellet.

5. Methods to ensure liquid is at bottom:

- If a microcentrifuge is available, briefly centrifuge the tube to bring the entire volume to the bottom and eliminate any air bubbles.
- A downward wrist-flicking motion.
- Lightly tap the tube(s) on a surface to bring the droplets to the bottom of the tube.

Repeat for all samples using new tips each time.

CRITICAL: Check that all liquid is at the bottom of the tube before proceeding to ensure test accuracy.

Sample Preparation

Protocol (Continued)

6. Run Test

- Place test strip (or portion if <8 samples) into CRISPOT® device.
- Select Program 1 and press “Play”.

7. Read Results

- Results display as:
 - Red = Positive
 - Green = Negative
- Record immediately (write or photograph). Results are lost if buttons are pressed or device is turned off.

Using the Pipette

1. Set the volume

- Adjust the pipette to the required volume (5–50 µL) using the dial.

2. Attach a tip

- Firmly press a pipette tip onto the pipette to ensure a secure fit.

3. Aspirate (draw liquid)

- Press the plunger down to the first stop (first point of resistance).
- Insert the tip into the liquid.
- Slowly release the plunger to draw up the liquid.

4. Dispense (release liquid)

- Move the tip to the target container.
- Press the plunger down to the second stop (past the first resistance) to fully dispense the liquid.

5. Remove and discard tip

- Keep the plunger pressed while removing the tip from the liquid to prevent re-aspiration.
- Dispose of the used tip.

Warning

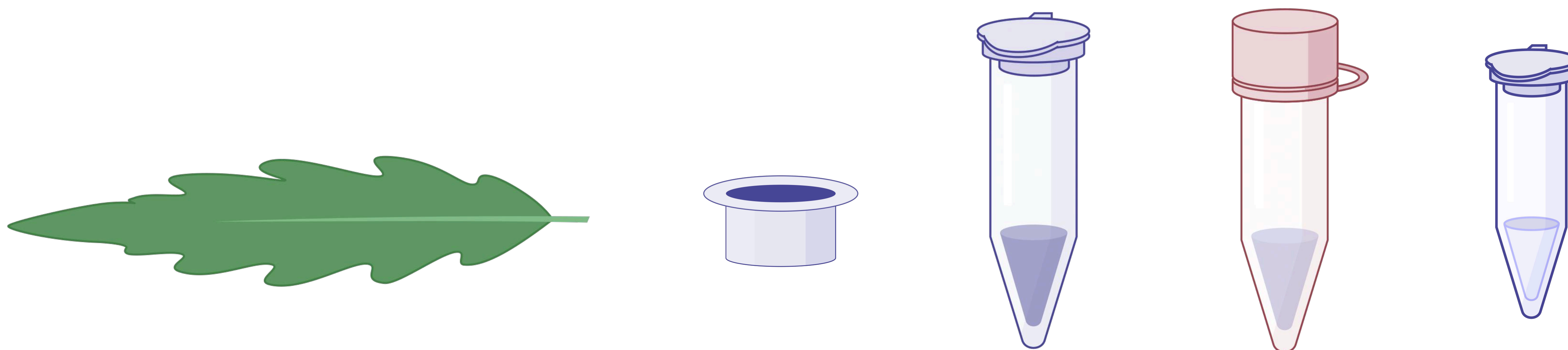
- Ensure all plant material is fully submerged in the lysis buffer before incubating in the CRISPOT® device.
- Change gloves and grommets between each sampling to avoid cross-contamination.
- Use a new pipette tip for each sample transfer to prevent cross-contamination.
- Dispose of used materials in the regular trash.

Sample Preparation: Visual Guide

Protocol

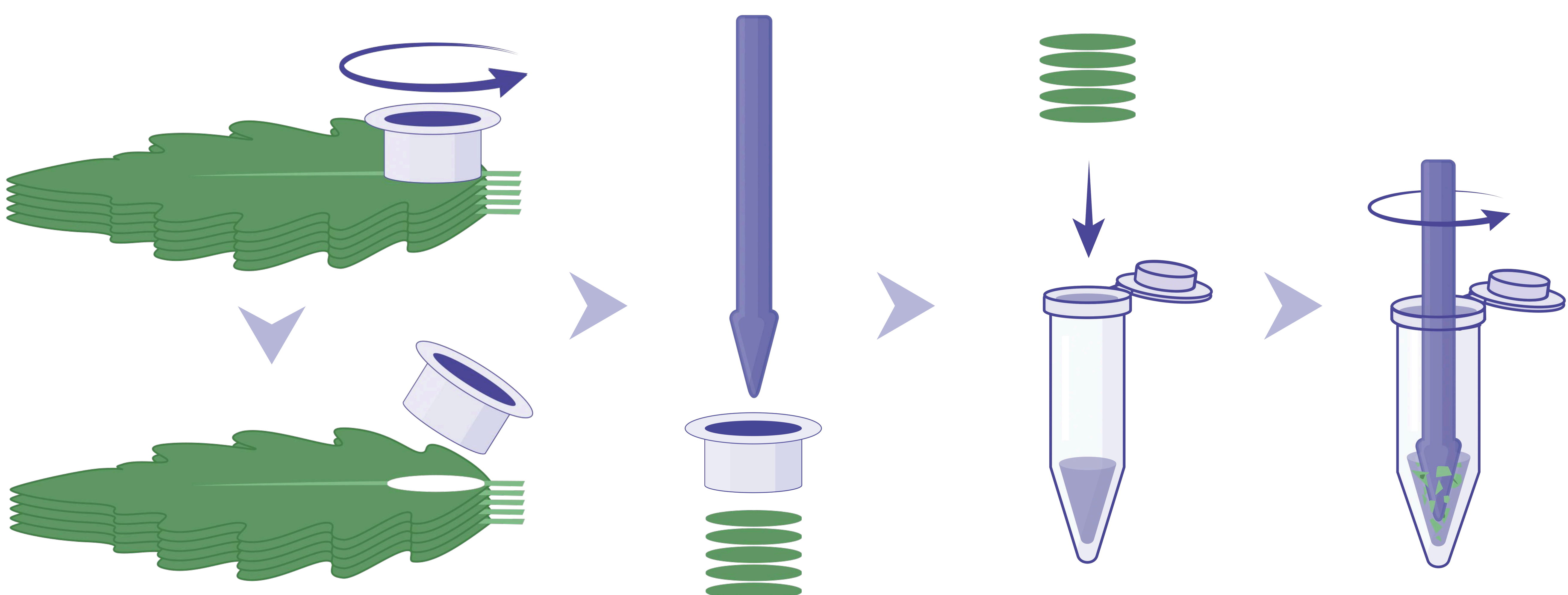
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2. Collect and Load Samples

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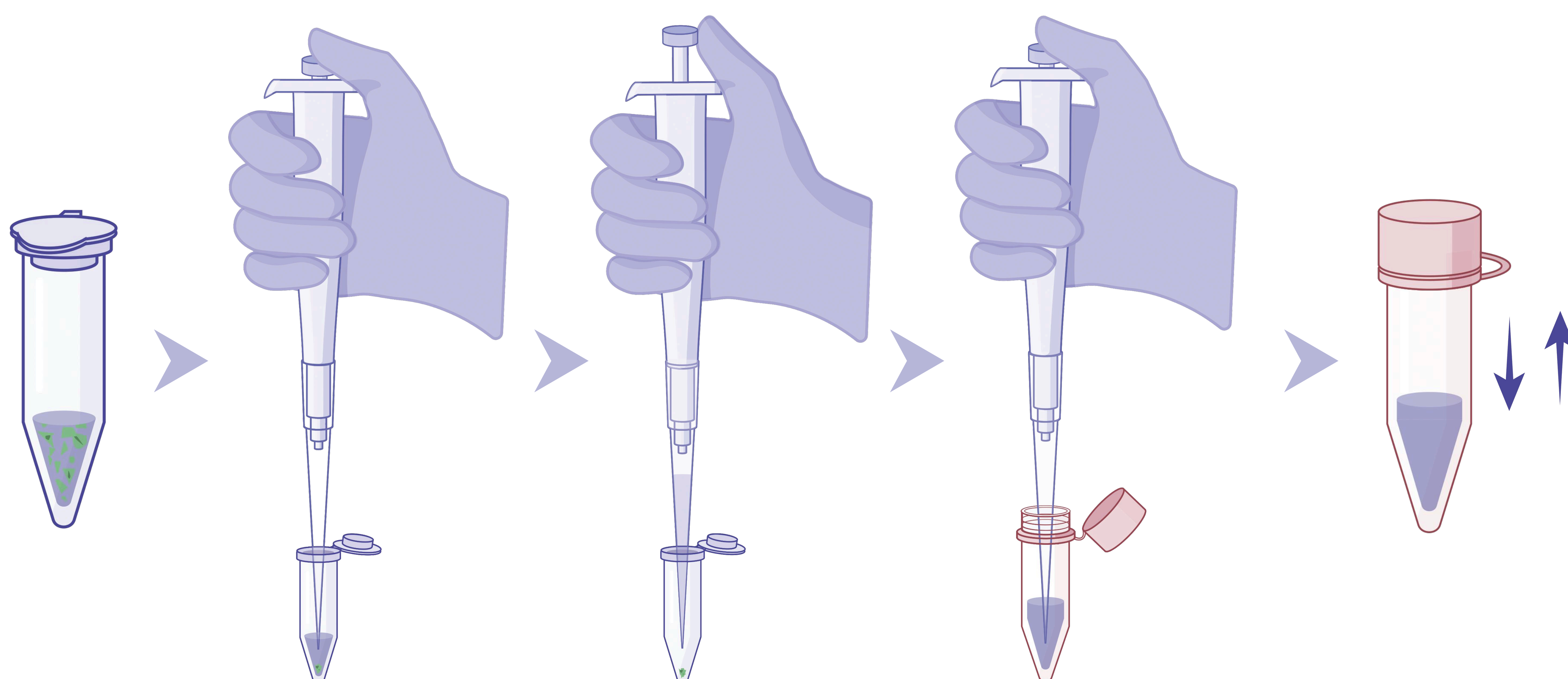
Sample Preparation: Visual Guide

Protocol

3. Add to Reaction Buffer

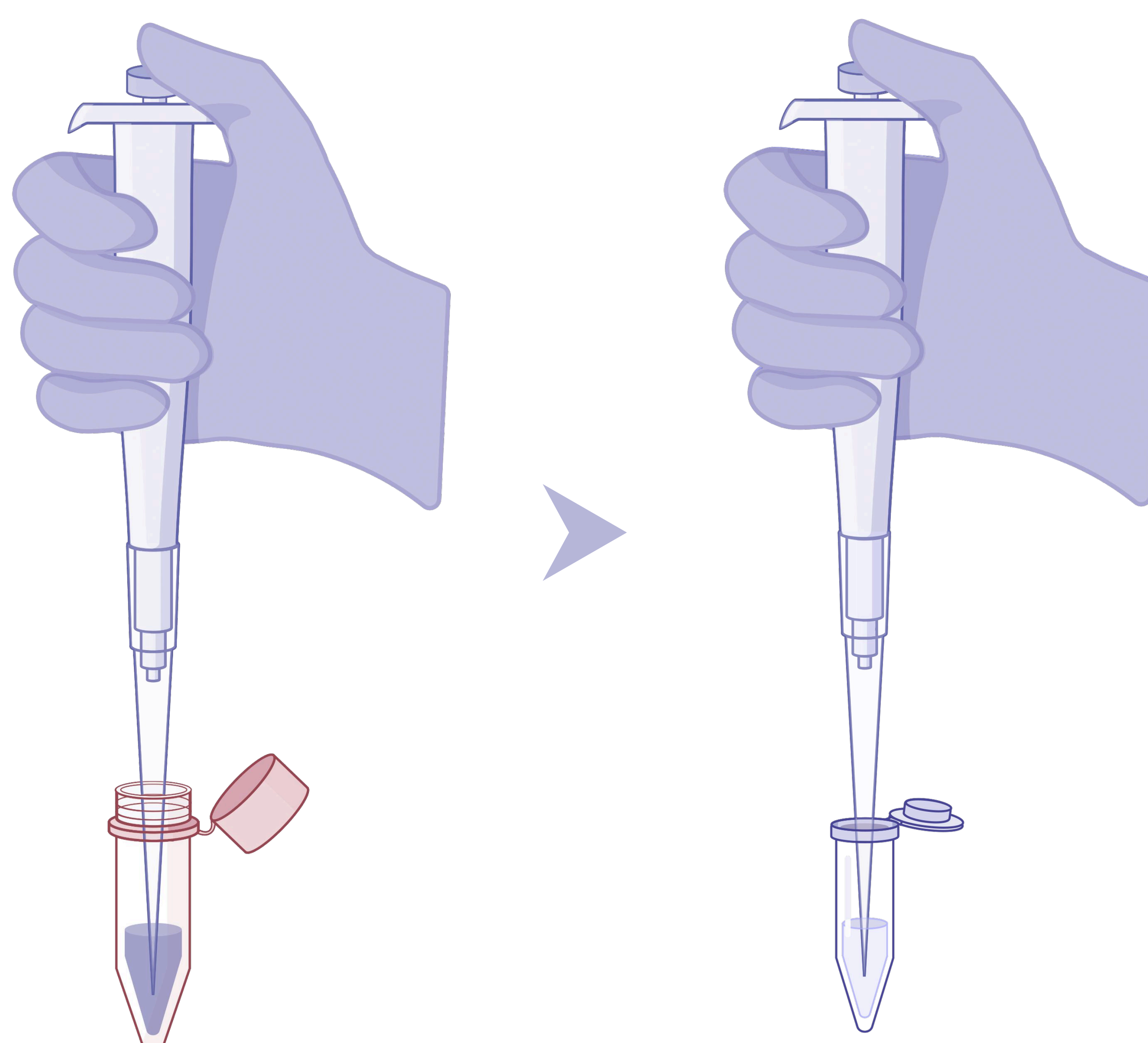
- Set pipette ② to 5 μ L and attach a clean tip ③.
- Open one tube at a time, transfer 5 μ L of the lysate from Step 2 into reaction buffer tube ⑦, then close and mix by shaking.
- Discard tip after each sample.

Important: Use a new tip for every sample and avoid opening multiple tubes at once.



4. Add to Master Mix

- Set pipette ② to 30 μ L and attach a new tip ③.
- Transfer 30 μ L of sample into master mix tube ⑧ without touching the pellet.
- Close tube, discard tip, and mix by flicking to fully resuspend pellet.



Sample Preparation: Visual Guide

Protocol

5. Methods to ensure liquid is at bottom:

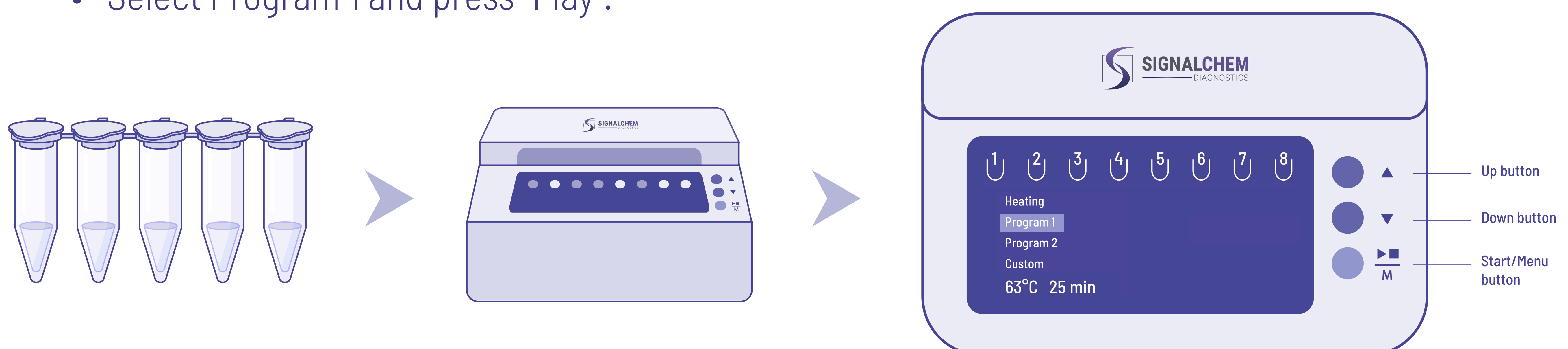
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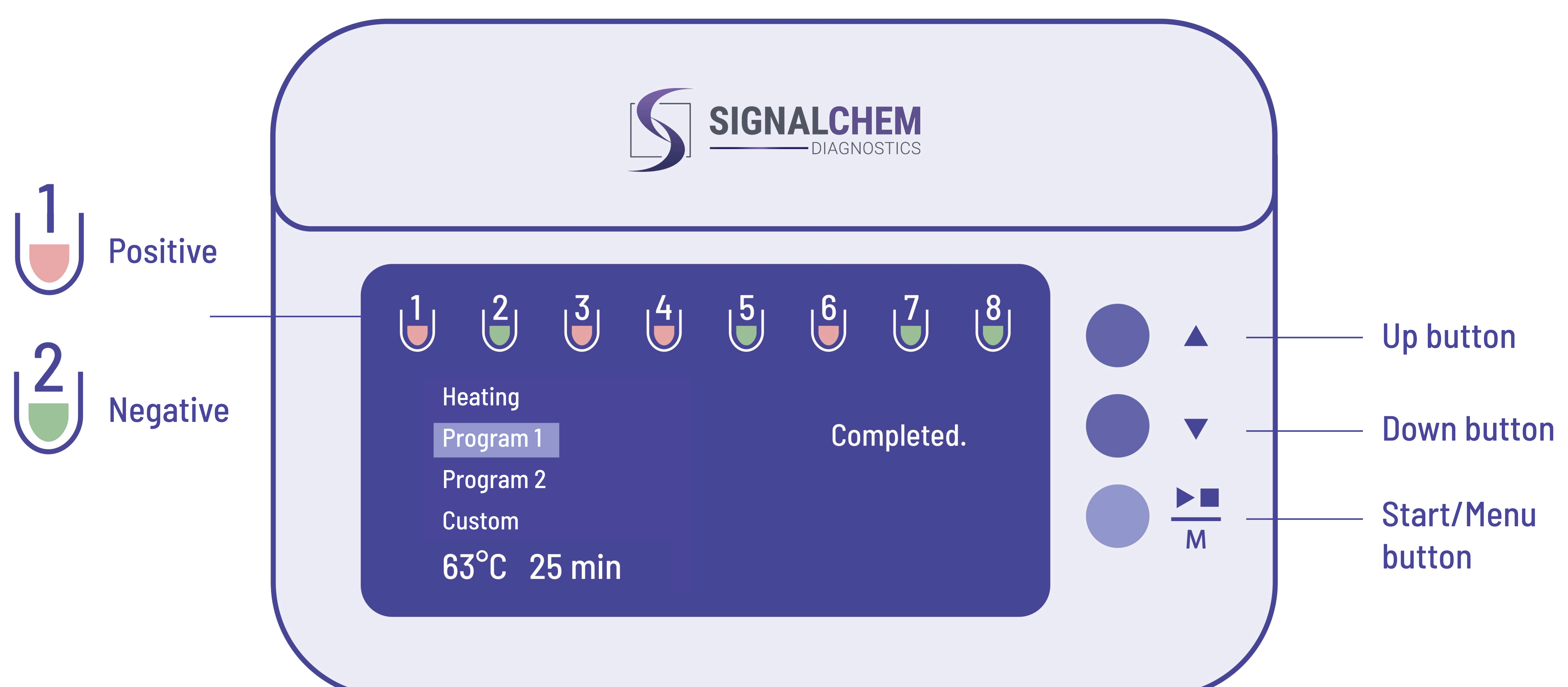
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- Place test strip (or portion if <8 samples) into CRISPOT® device.
- Select Program 1 and press "Play".

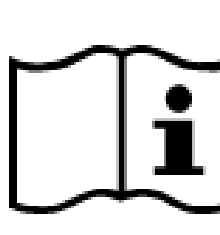


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Index of Symbols

	Do not reuse		Manufacturer		Catalog number
	Temperature limit		Batch code		Consult instructions for use
	Caution		Use by		


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