

# HTXImaging M3+ Sprayer

MSF Training

HTXimaging parts and  
turning on procedure

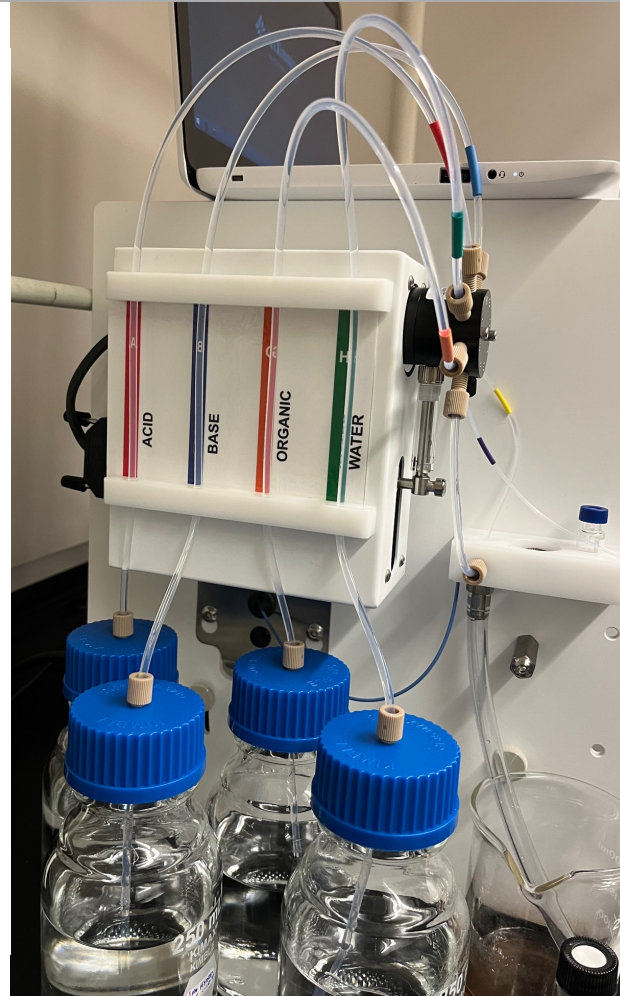
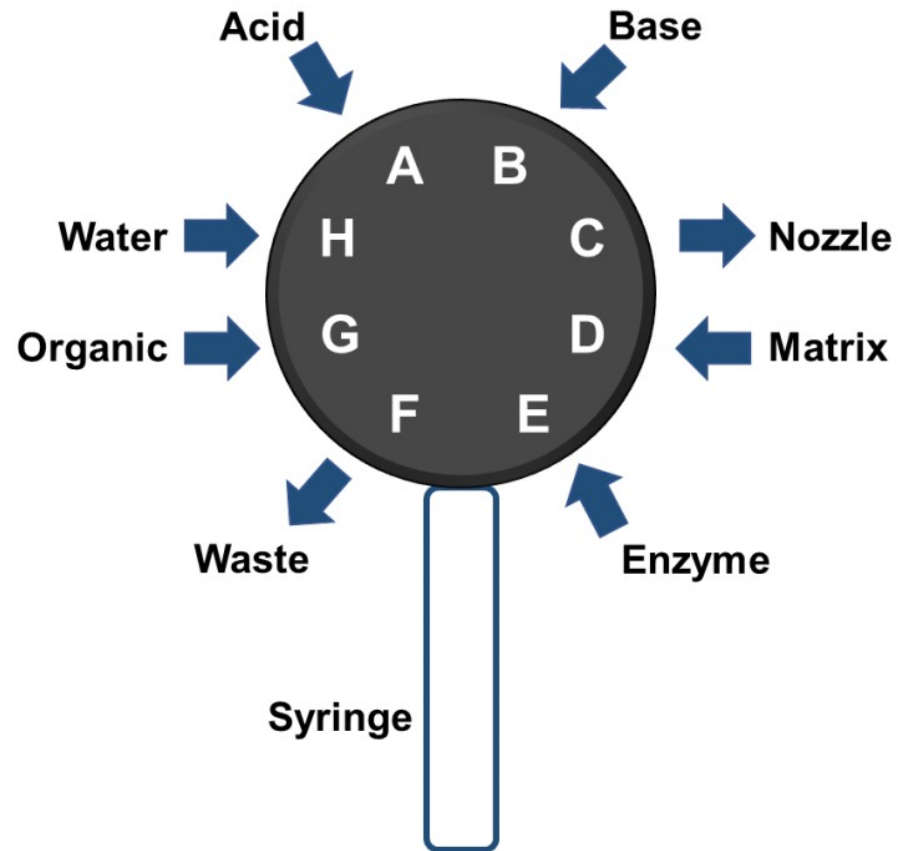


# Solvent delivery

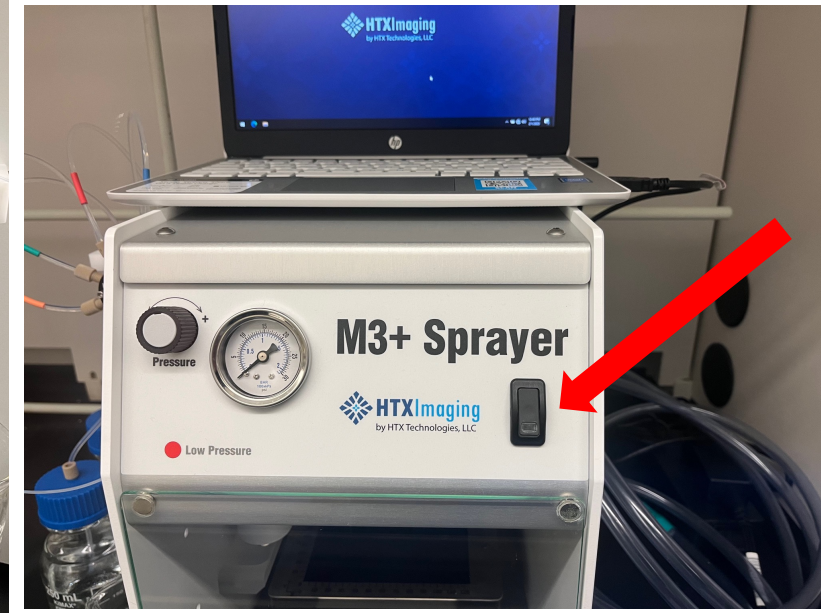
8 ports rotate valve, each with color- and letter-coded tubes. A 1 mL syringe withdraws or delivers solvent from or to each port.

Acid (A) = Red  
Base (B) = Dark Blue  
Nozzle (C) = Light blue  
Matrix (D) = Yellow

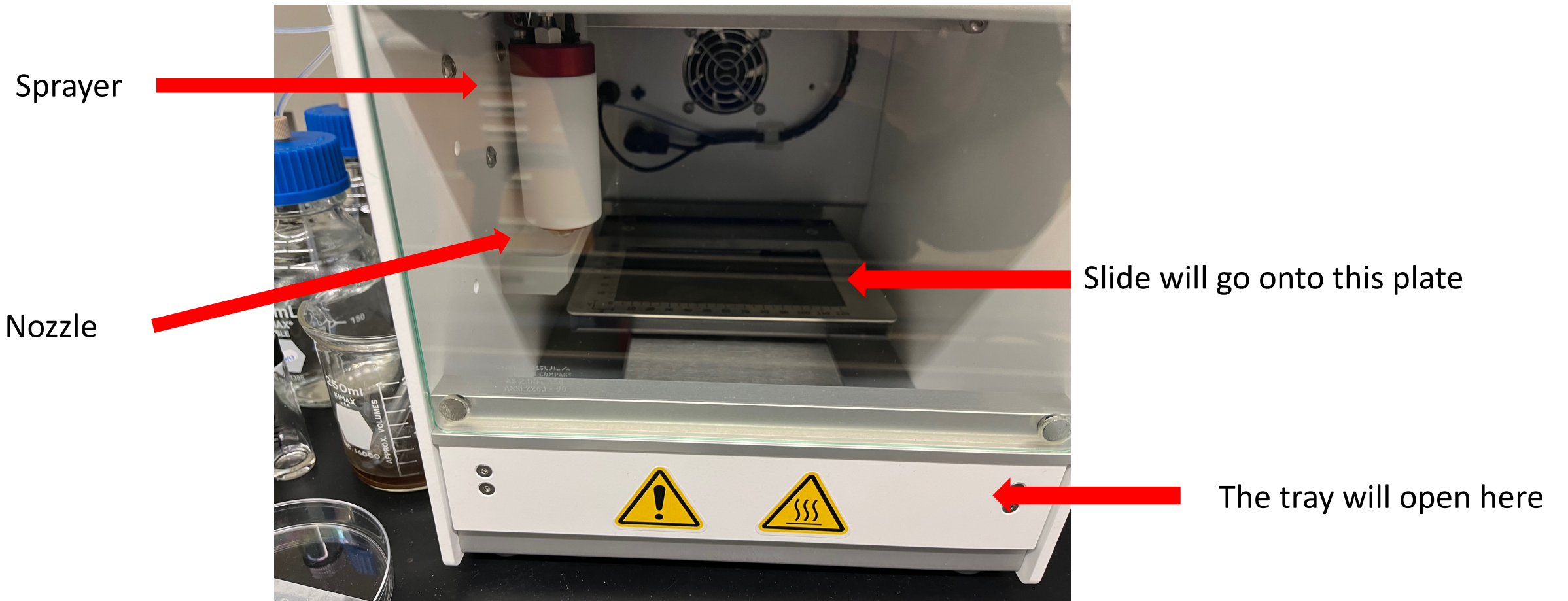
Enzyme (E) = Purple  
Waste (F) = Clear tube  
Organic (G) = Orange  
Water (H) = Green



The instrument may be turned off. Turn it on by pressing on the switch highlighted in the picture below. **Turn on before opening software**



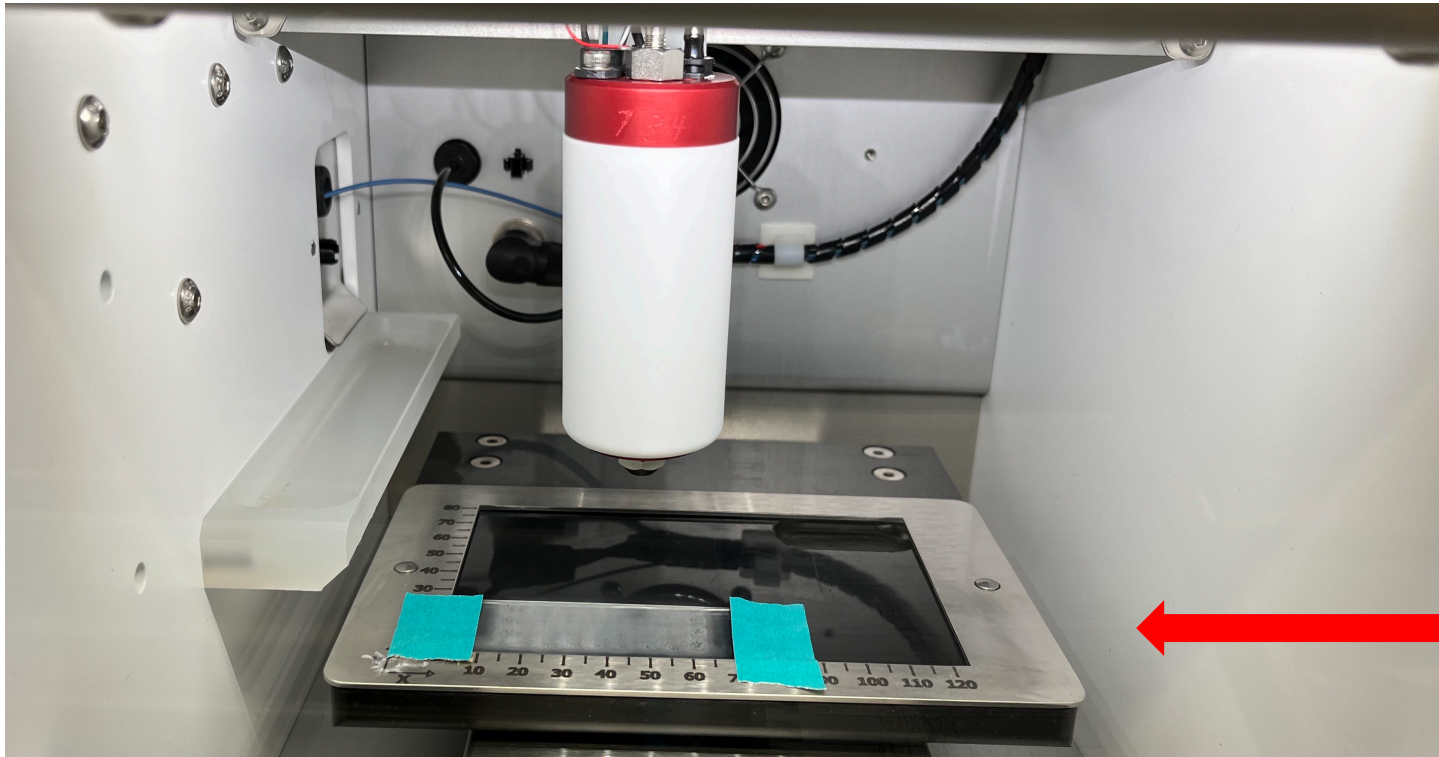
# Spray Chamber





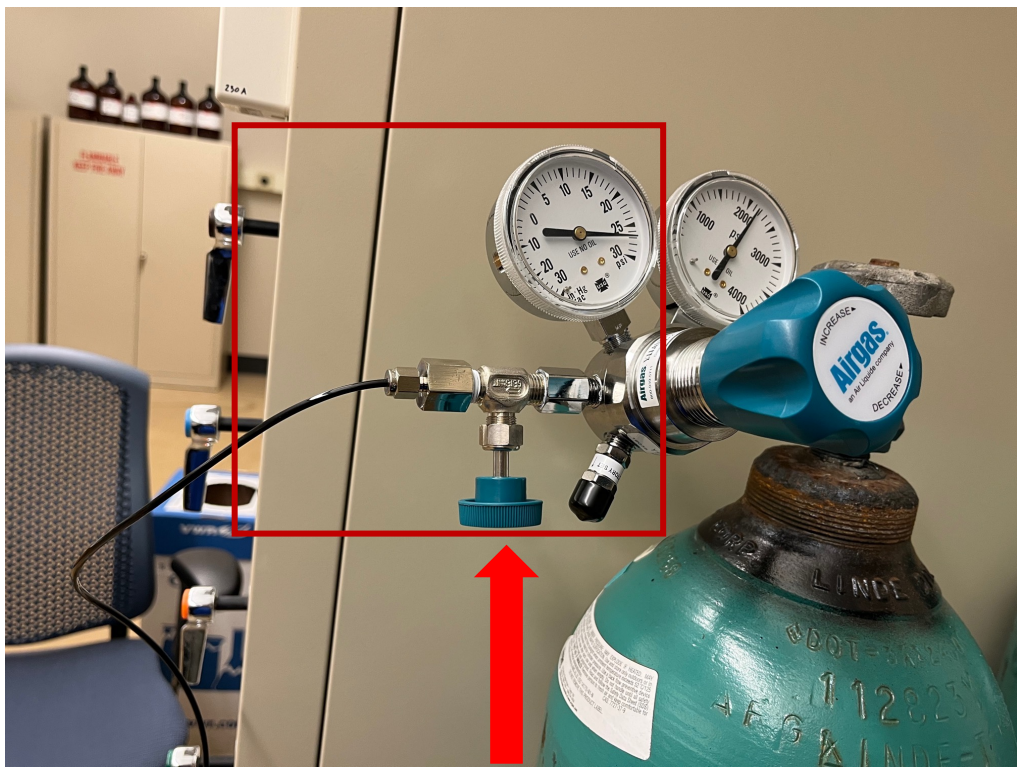
# Tape down the slide

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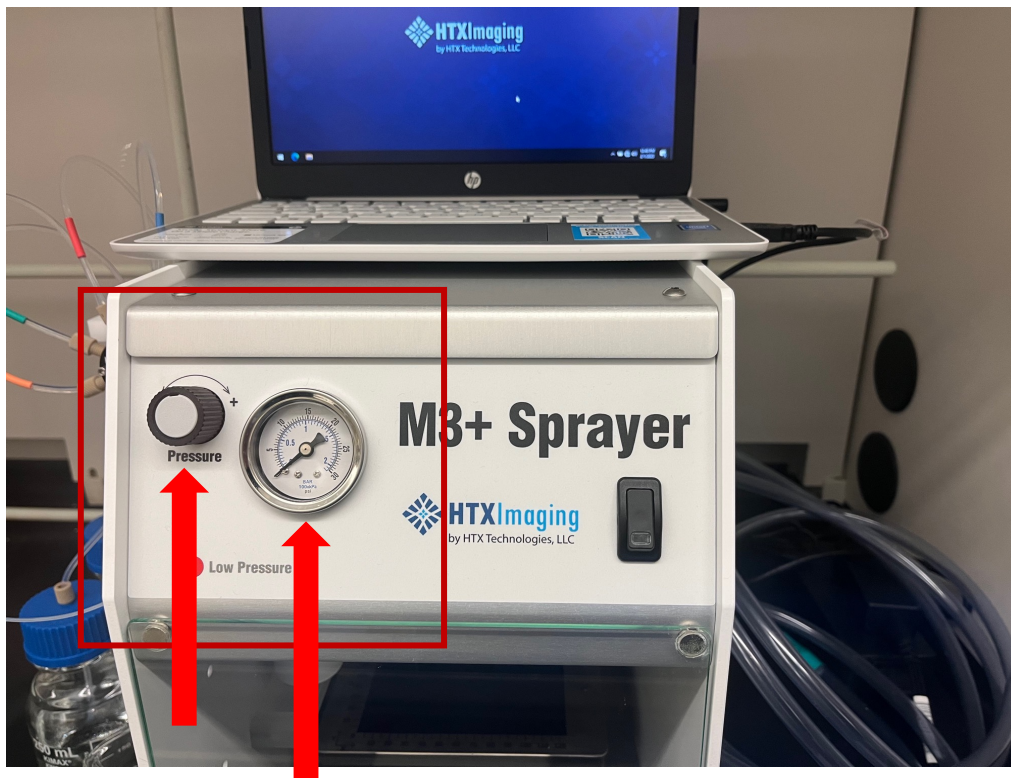


Tape down slide so it will not move during the matrix application

# Nitrogen Gas



Turn bottom nob so that the pressure is between 15 - 20 psi



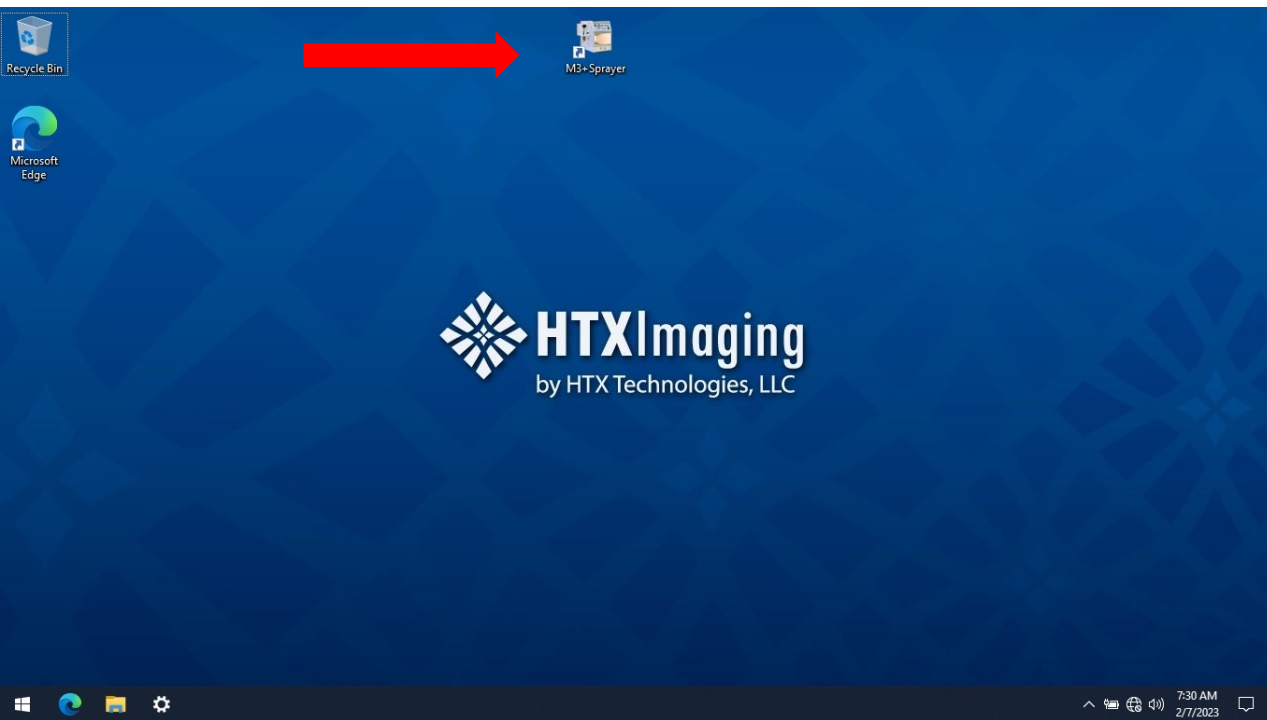
This should be 10 psi, and the pressure can be regulated with the knob to the left of the gauge

# HTXImaging software setup

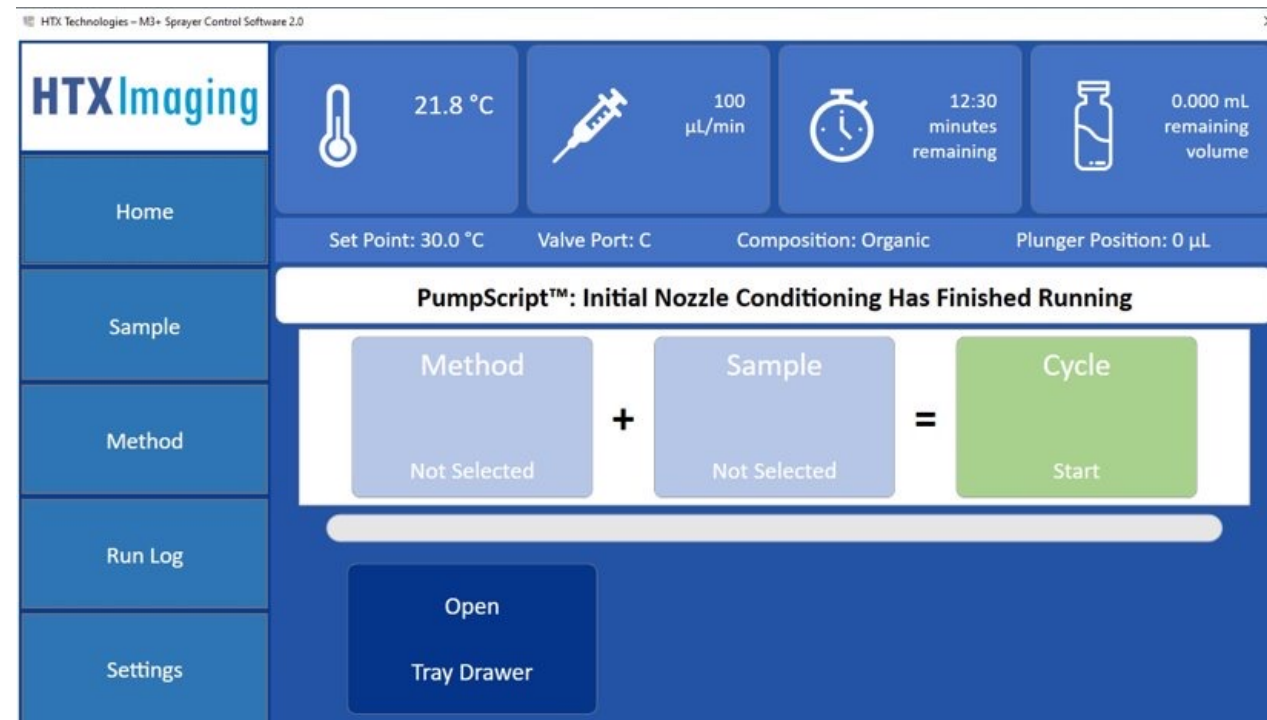




# M3 Sprayer + homepage



Click M3+ sprayer to open software

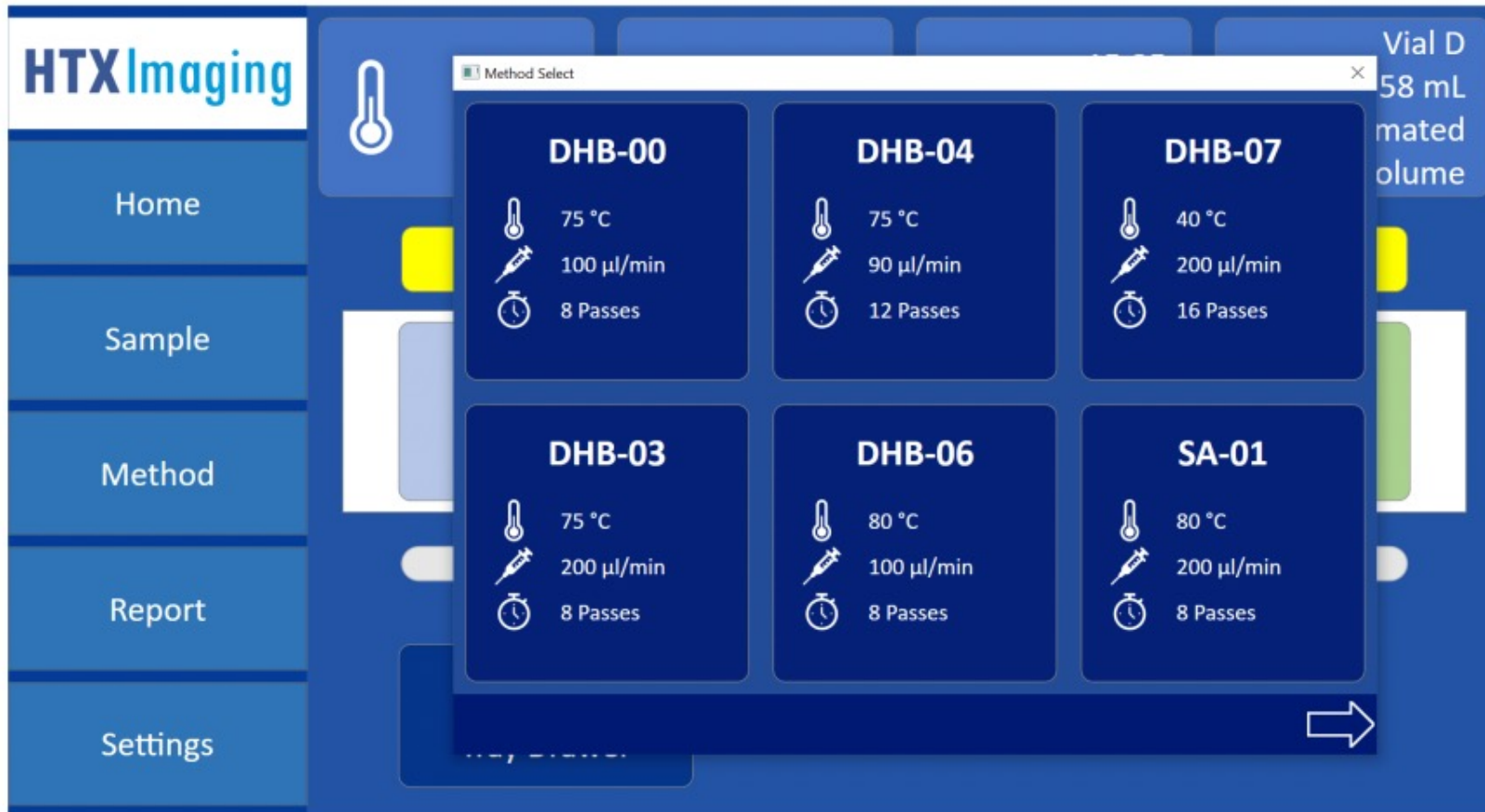


Click in the order shown: Method, sample then cycle. Do not ignore prompt messages on screen



# Step 1

**Step 1.** Select a Method by clicking on the **Method** button to reveal a pop-out of methods in folder. Click on desired method.



Each method has a brief summary of the temperature that will be used, the flow rate and the number of passes that will be executed on the sample.

Note: there are additional parameters that are not visible, such as the nozzle movement speed.

# Step 2

**Step 2.** Select a Sample size by clicking the **Sample** button to bring up the Sample tab. Click on desired template and then adjust as needed by clicking and dragging the red dotted line which represents the spray area. Once sample size is set, click **Confirm Spray Area** button. If selecting a template with no modifications needed, double clicking the template will set the spray area and return to home screen.

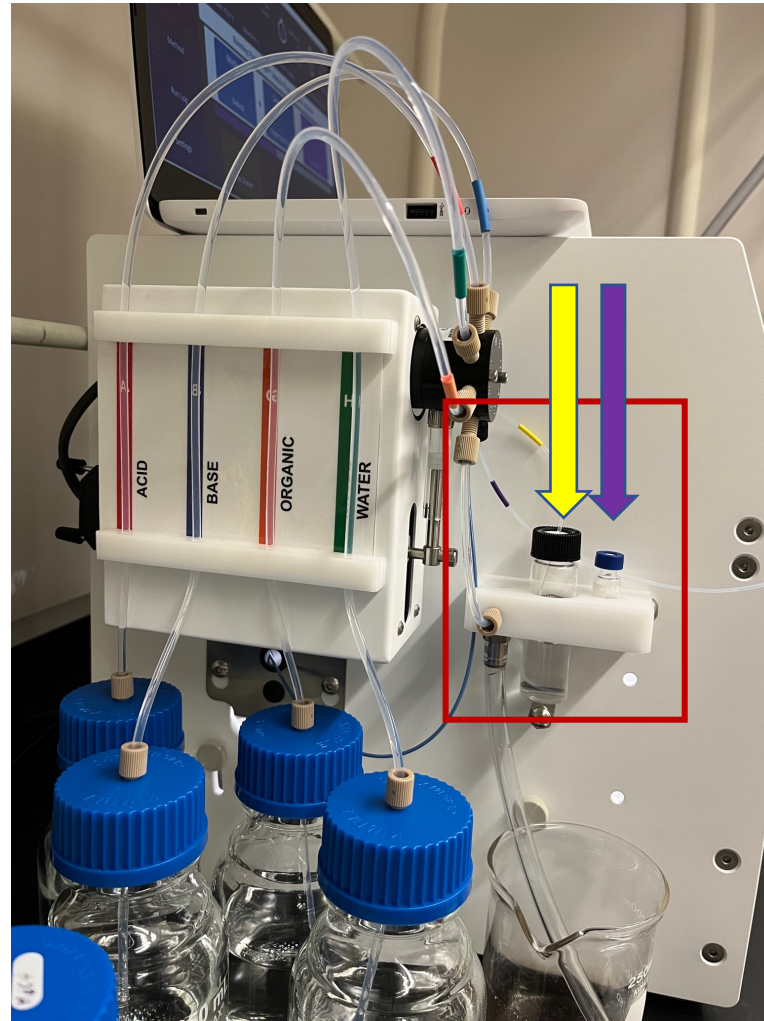
The screenshot shows the HTX Imaging software interface. On the left is a sidebar with navigation buttons: Home, Sample, Method, Run Log, and Settings. The 'Sample' button is selected. In the center, a grid displays a green square representing the slide and a red dotted line representing the spray area. Above the grid, a 'Plate Selection' dialog box shows 'Three Glass Slides' with X Range (mm) from -5 to 80 and Y Range (mm) from -5 to 80. On the right, there are buttons for 'Confirm Spray Area', 'Save Template', and three 'Custom' templates (Custom - Alyson, Custom - 2, Custom - 3).

The green square is your slide, and the red square is the spray area.

You want to leave a little space like the picture to have an even coating

# Step 3

**Step 3.** Load reagent (matrix or enzyme solution) into **Matrix/Enzyme Vial** connected to Port D/E of the pump. Estimated cycle reagent usage is displayed on the Vial button.



The matrix line is yellow which corresponds to port D.

The enzyme line is purple and corresponds to port E.



# Step 4

**Step 4.** Press the **Start** button to begin the spray cycle. This will communicate the method temperature to the unit. A reminder will appear to turn on nitrogen gas for nozzle temperature to increase. Confirm the volume of reagent loaded in vial. The button will remain yellow until the method temperature has been reached.

The screenshot displays the HTX Imaging control interface. On the left is a vertical navigation menu with options: Home, Sample, Method, Run Log, and Settings. The main display area features a top status bar with four panels: a temperature panel showing 30.0 °C, a flow rate panel showing 25 µL/min, a timer panel showing 8:20 minutes remaining, and a vial volume panel showing Vial E with 2.000 mL estimated volume. Below this is a secondary status bar with four panels: Set Point: 45.0 °C, Valve Port: F, Composition: Enzyme Vial, and Plunger Position: 30 µL. The central area shows a white banner with the text 'Cycle Started - Waiting On Temp.' Below this is a visual equation: a light blue box labeled 'Method' with 'PNGaseF' below it, followed by a plus sign, a light blue box labeled 'Sample' with 'Single Glass Slide' below it, followed by an equals sign, and a yellow box labeled 'Cycle' with 'Waiting For Temp.' below it. At the bottom, there are two buttons: 'Open Tray Drawer' on the left and 'Abort Cycle' on the right. The text 'Pass 1 of 15' is centered at the bottom of the interface.



# Gas on + sample volume

+ Sprayer Control Software 2.0

Imaging 21.8 °C 100  $\mu\text{L}/\text{min}$  5:35 minutes Vial 1.000 remaining volume

Plunger Position: 0  $\mu\text{L}$

Attention  
Please Turn On Gas

OK

Cycle

Waiting For Temp.

Open Tray Drawer Abort Cycle

Pass 1 of 8

Imaging - M3+ Sprayer Control Software 2.0

Imaging 21.8 °C 100  $\mu\text{L}/\text{min}$  5:35 minutes Vial 1.000 remaining volume

Plunger Position: 0  $\mu\text{L}$

Attention  
Please Enter Vial Volume. There Must Be At Least 614  $\mu\text{L}$  in Vial

2000  $\mu\text{L}$

OK Cancel

Open Tray Drawer

Home Sample Method Run Log Settings

and Running Cycle Start

# Step 5

**Step 5.** Once the nozzle reaches temperature, the **Continue** button will flash green and play an alert sound. Click Continue. Unit will delay the start of the spray cycle to allow full purge of the delay volume between valve and nozzle tip.

The above temperature must equal the set point temperature or be two degrees lower to continue

The screenshot displays a control interface with several data points and a status bar:

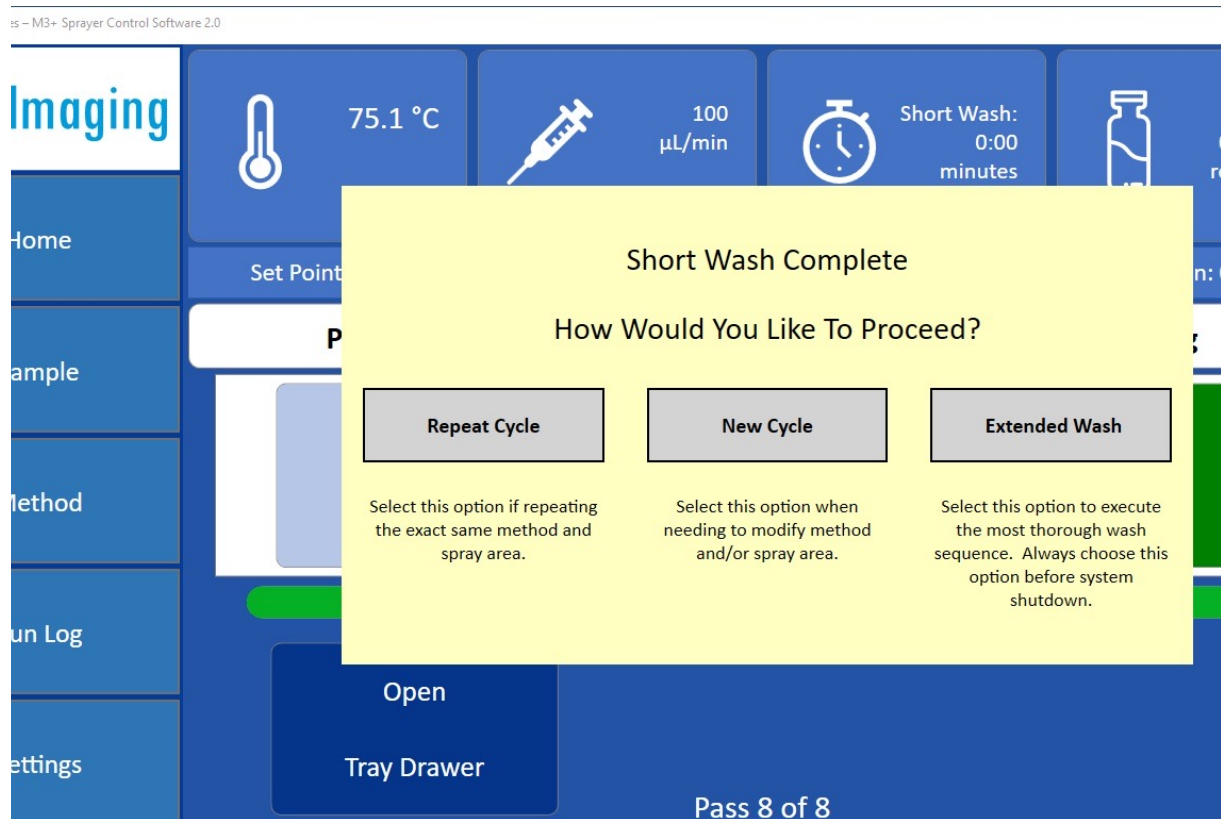
- Temperature:** 74.9 °C (highlighted with a red box). Below it, the Set Point is 75.0 °C.
- Flow Rate:** 100 µL/min.
- Short Wash:** 0:35 minutes remaining.
- Vial D:** 0.440 mL remaining volume.
- Valve Port:** C
- Composition:** Organic
- Plunger Position:** 189 µL

The main status bar reads: **Running PumpScript™: Matrix Flush - Organic Solvent**

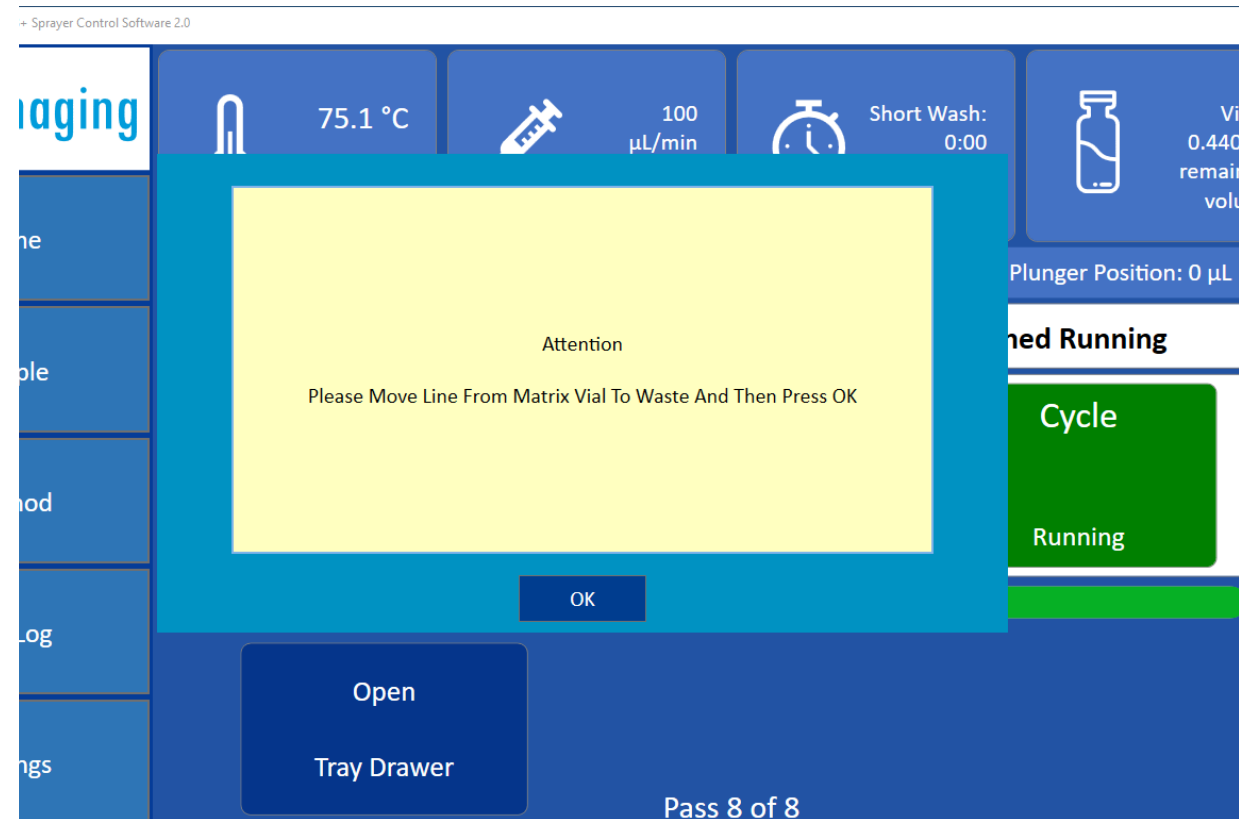
Below the status bar, a visual equation shows: **Method** (DHB-00) + **Sample** (Single Glass Slide) = **Cycle** (Running). The **Cycle** box is highlighted with a red border.

At the bottom, there is an **Open Tray Drawer** button and a progress indicator showing **Pass 8 of 8**.

# Extended Wash



Check one of the three. If finished click extended wash.



Remove matrix line before extended wash

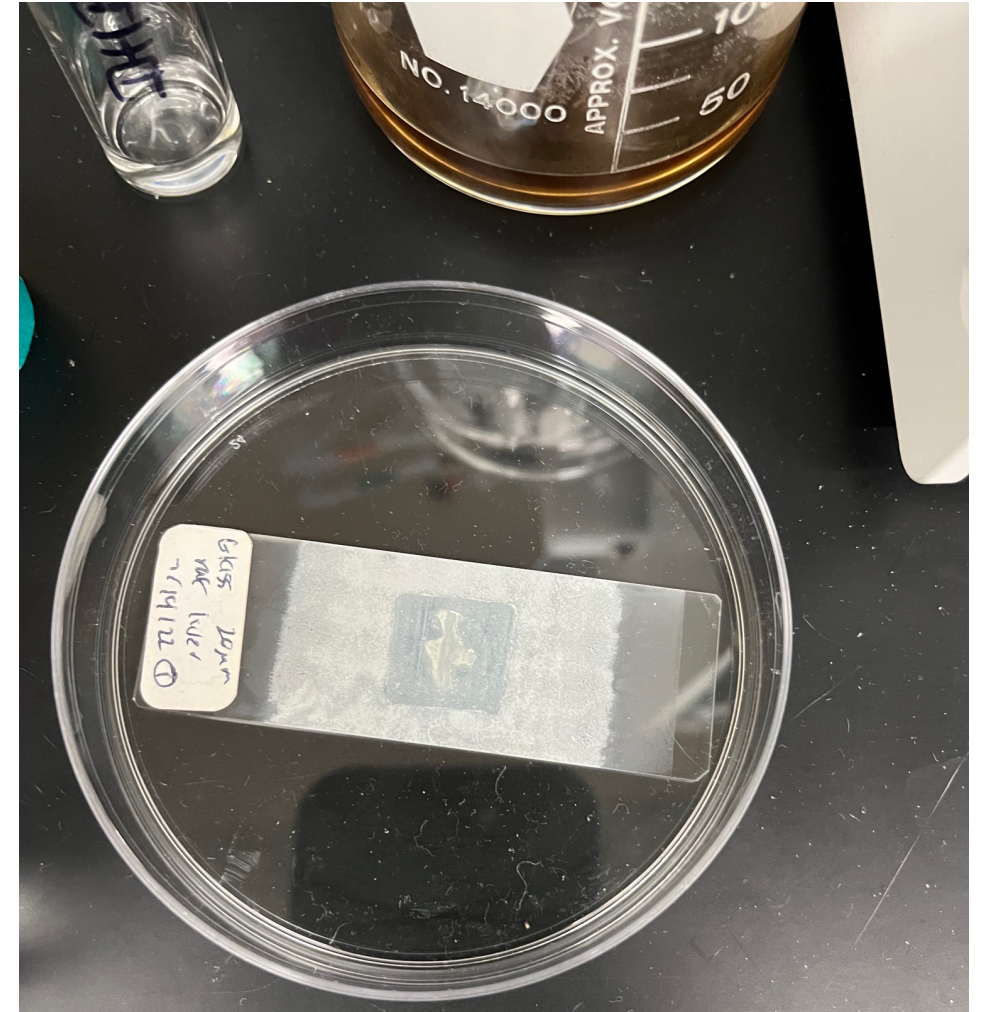
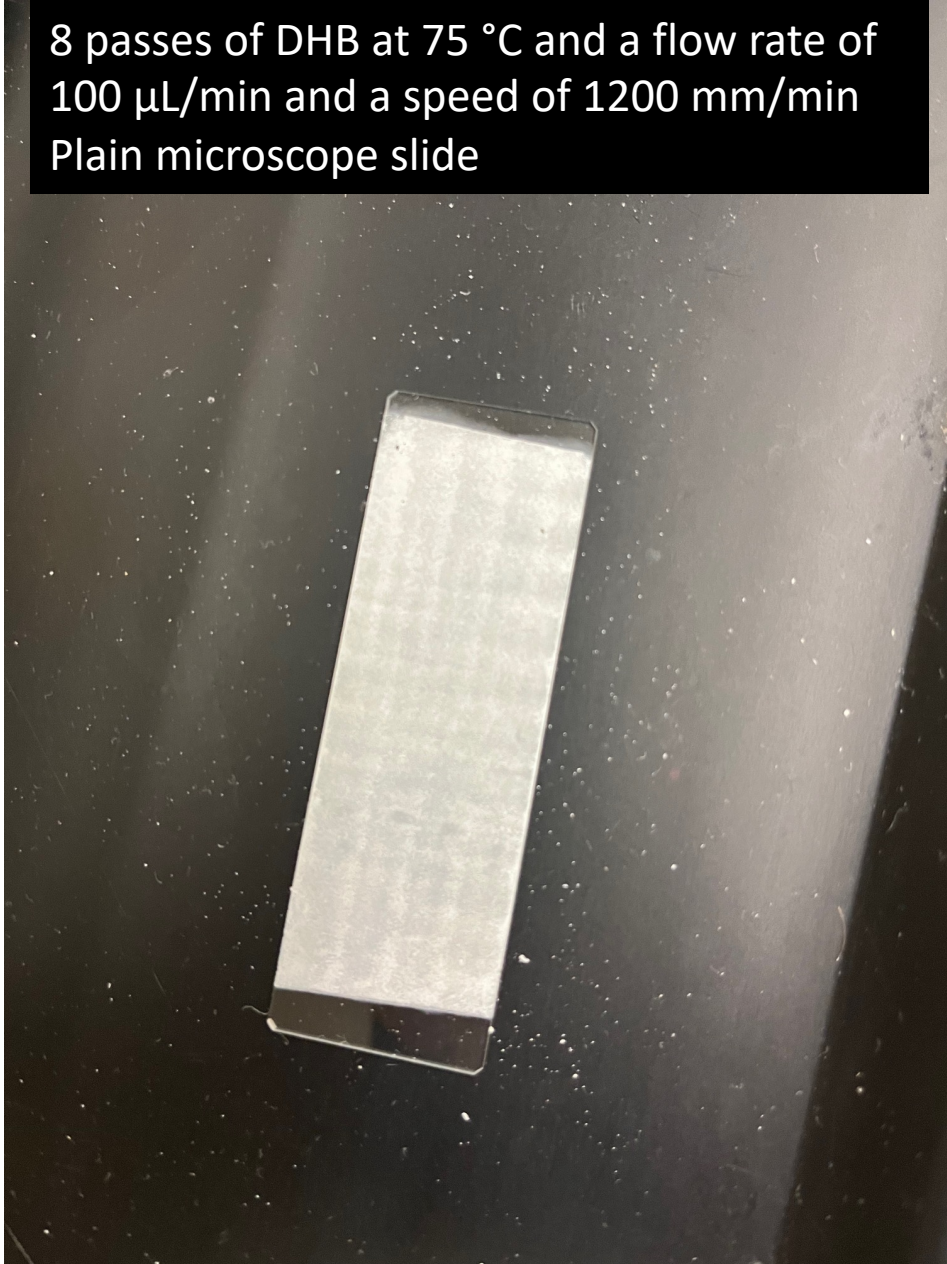
# Examples of matrix application





# Matrix on sample

8 passes of DHB at 75 °C and a flow rate of 100  $\mu$ L/min and a speed of 1200 mm/min  
Plain microscope slide



8 passes of DHB at 75 °C and a flow rate of 100  $\mu$ L/min and a speed of 1200 mm/min  
ITO microscope slide with liver section  
embedded in gelatin