

SiPM Test Bench Overview

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Project Overview

The smartphone-integrated paper-based microfluidic (SiPM) platform includes four primary components: a smartphone with a built-in camera, a green bandpass filter (emission range: 490 - 530 μm), an external lens module, and a blood filtration system using thin paper sections possessing varying pore size ranges to target specific cell types.

The smartphone used in this project is the Samsung Galaxy S20 Ultra, consisting of three separate lenses on its backside. Each lens has its own range of magnification, increasing downward on the back of the phone. The top lens has a range from 0.5x - 1x, the middle lens has a range from 1x ~ 5x (depending on distance from sample), and the bottom lens has a range from ~5x - 100x. For this reason, it may be practical to purchase another external lens set for the bottom-most camera lens. This way, transitioning between the two magnification ranges would only require adjusting the sample on its platform in Figures 3 and 4.

For convenience and simplification, I designed a 3D-printed housing assembly to combine and fix the smartphone with the green filter and external lens (Figure 1). This would allow for more accurate camera positioning, as well as maintaining the external lens location.

To ensure the capability of the proposed design, I tested its ability to focus on samples, representing brightfield microscopy, as well as its ability to detect fluorescent-coated particles that emit green when illuminated with a blue light source (Figure 8).

Throughout this report, I will detail my reasons for my design choices, how the smartphone setup works, and the data I have collected thus far while using the setup on different samples.

Design Approach

The purpose of creating a portable, easy-to-use platform for UTI detection was to eliminate test waiting times and establish a robust point-of-care delivery system for patients. The test bench was designed with three key factors in mind: fix the smartphone, lens, and filter together, create an adjustable XYZ platform for the sample, and make the assembled design convenient for the user (in progress). Keep in mind that this is only a preliminary prototype and is not yet a portable product.

Design Drawings

Below are the drawing views for each component in the SiPM Testbench, as well as the final assembly. All units are in “mm” unless otherwise specified. These were designed on a free-to-use cloud-based design platform called Onshape. If Solidworks is not available due to costs, I would recommend using this software.

Phone Holder + External Lens and Filter

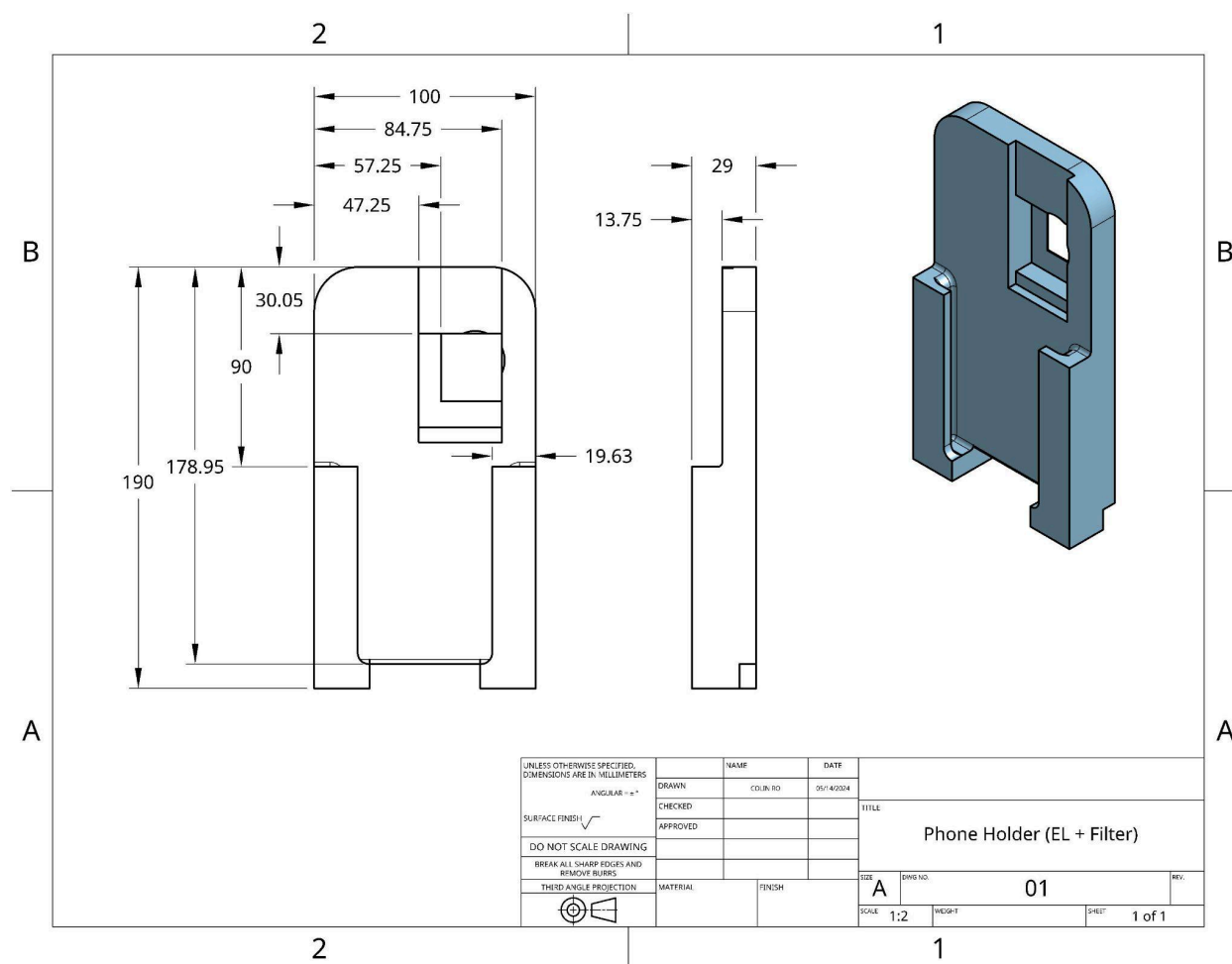


Figure 1. Phone Holder (EL + Filter) Drawing (01)

The phone holder design was created with the intention of securely holding the cell phone while the filter and external lens were fixed. The bottom portion of where the external lens sits maintains a thickness of 0.5 mm. The design is noticeably thin at this point to minimize the distance between the sample and external lens (for precision focus, the distance between the lens and sample must be ≤ 1 mm). A hinge mechanism was attempted but was imperfect due to 3D-printing difficulty.

Battery and Sample Platform Housing

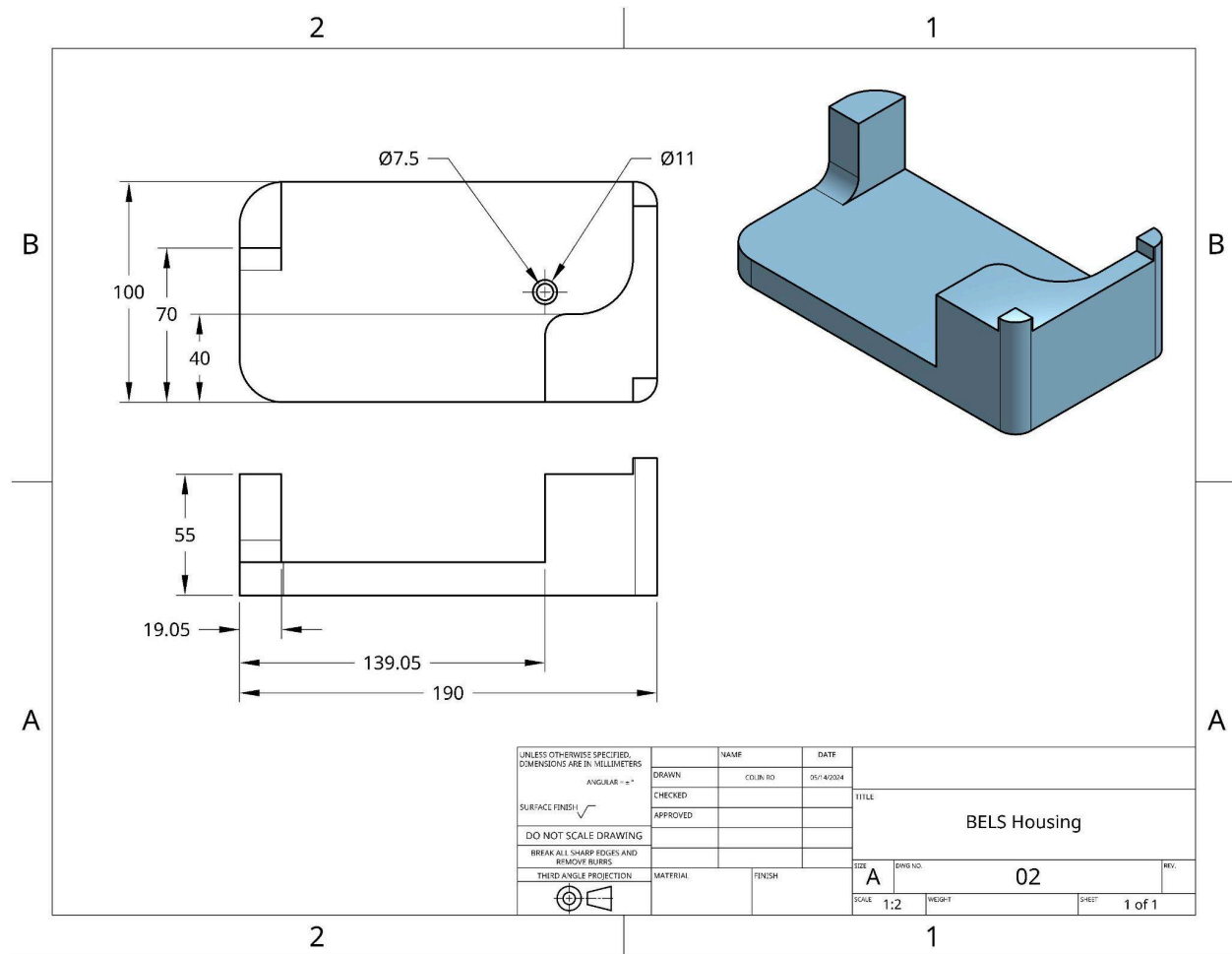


Figure 2. Battery and Sample Platform Housing Drawing (02)

The Battery and Sample Platform features an open-concept design, allowing free range of motion for the XYZ adjustable table that the sample holder attaches to. The hole on the base of the design is meant to support a screw to fasten the structure to a black Thorlabs block.

25mm X 25mm Sample Holder

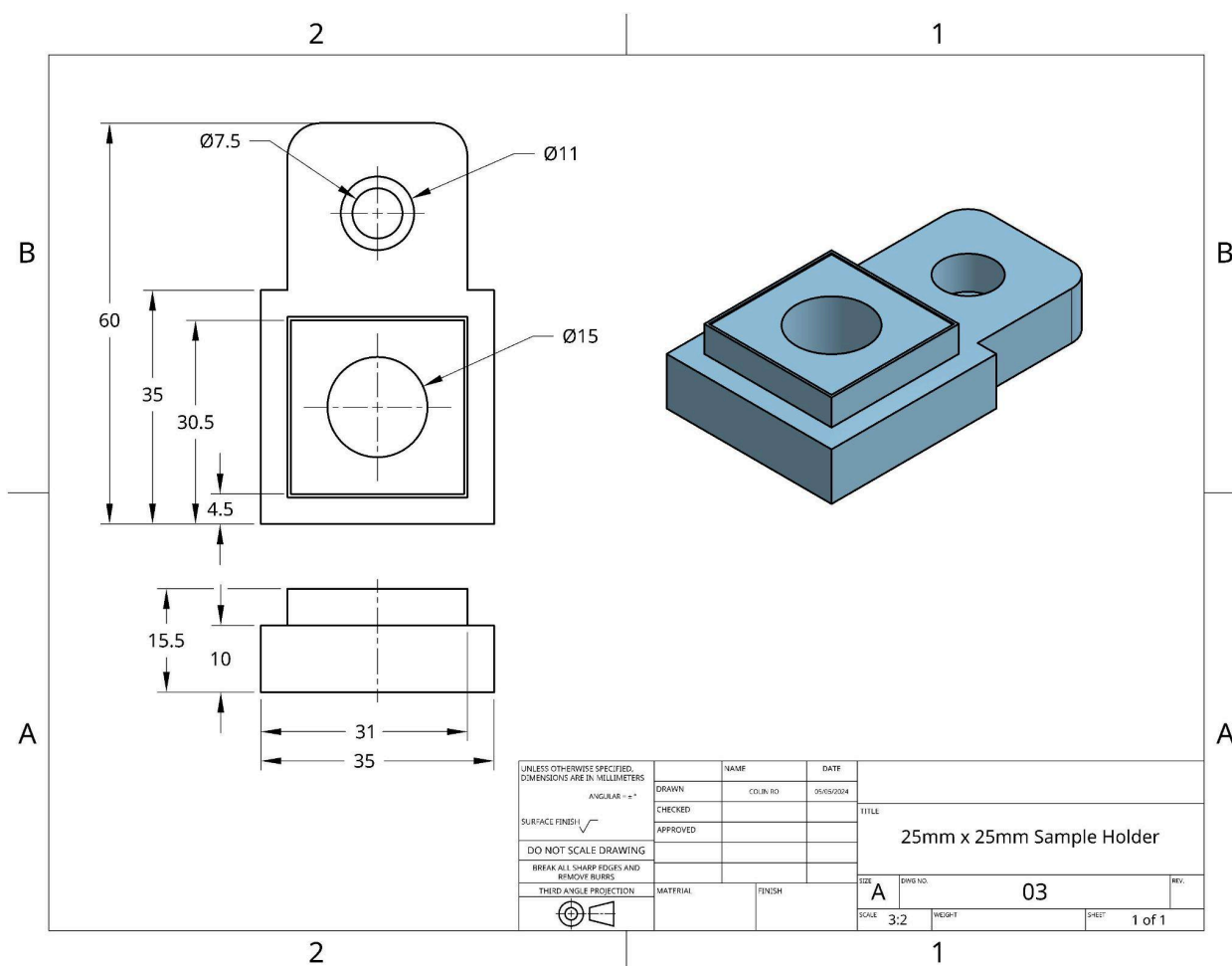


Figure 3. 25mm x 25mm Sample Holder Drawing (03)

After trial and error, a holder dedicated to a sample felt necessary to maintain consistent viewing results. The sample must be within 1mm of the external lens for high picture quality, hence the elevated platform. For Brightfield observation, the 15mm hole in the center of the holder can be used as a light passage to illuminate the sample. For fluorescent particles, it is necessary that the light source be parallel to the sample instead of under it due to fiber shadows in the paper.

Hemocytometer Holder

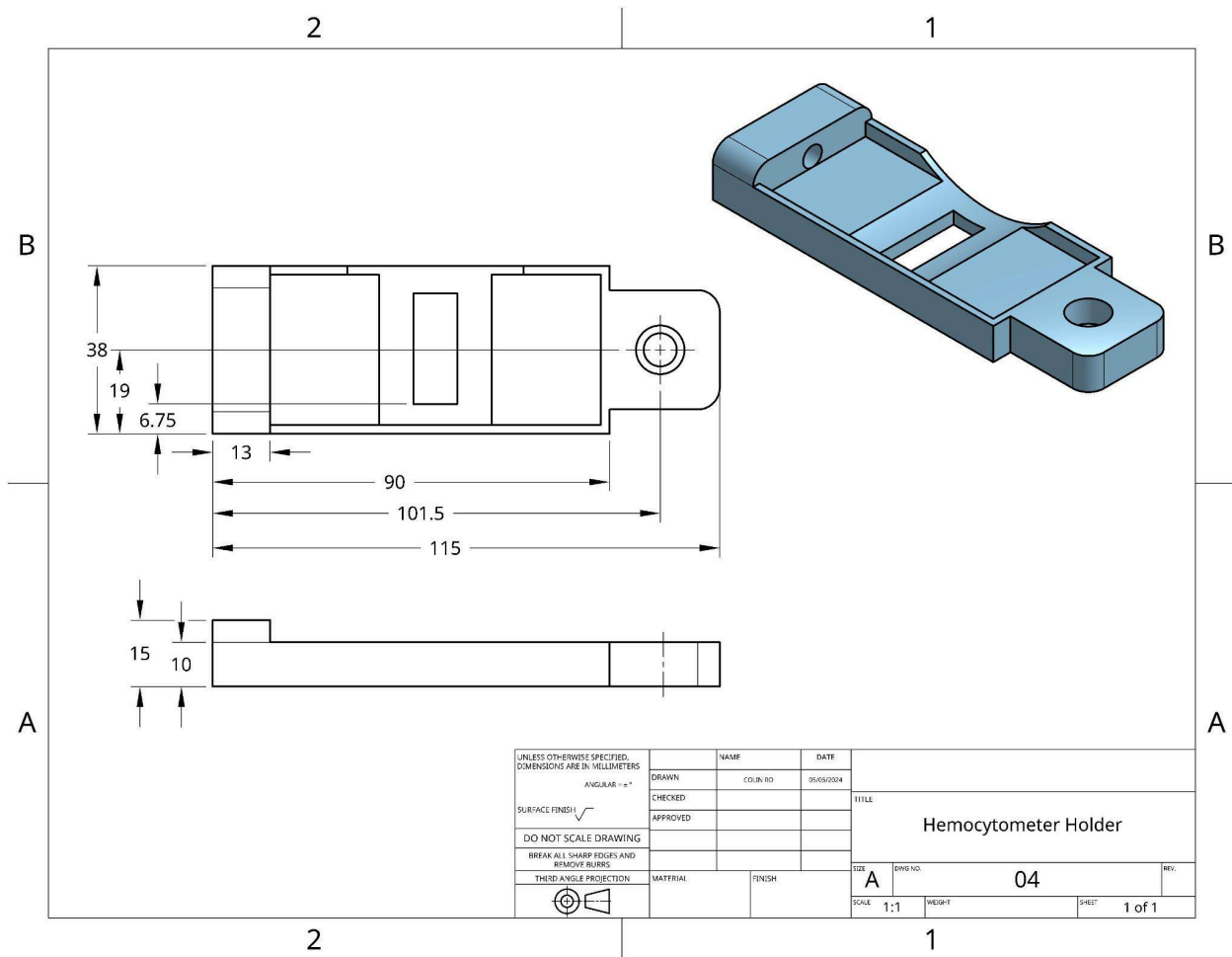


Figure 4. Hemocytometer Holder Drawing (04)

Presently, fluorescent particles have been visible on the SiPM testbench and measurable when using the hemocytometer. Because the hemocytometer has transparent thickness, the blue light from the led is able to travel to the sample more freely, unlike the paper filtration system. When using the filtered paper, the sample must be nearly touching the external lens, consequently not leaving enough room for the blue light to illuminate the particles. This is a current obstacle of the filtration system.

Smartphone Platform Assembled

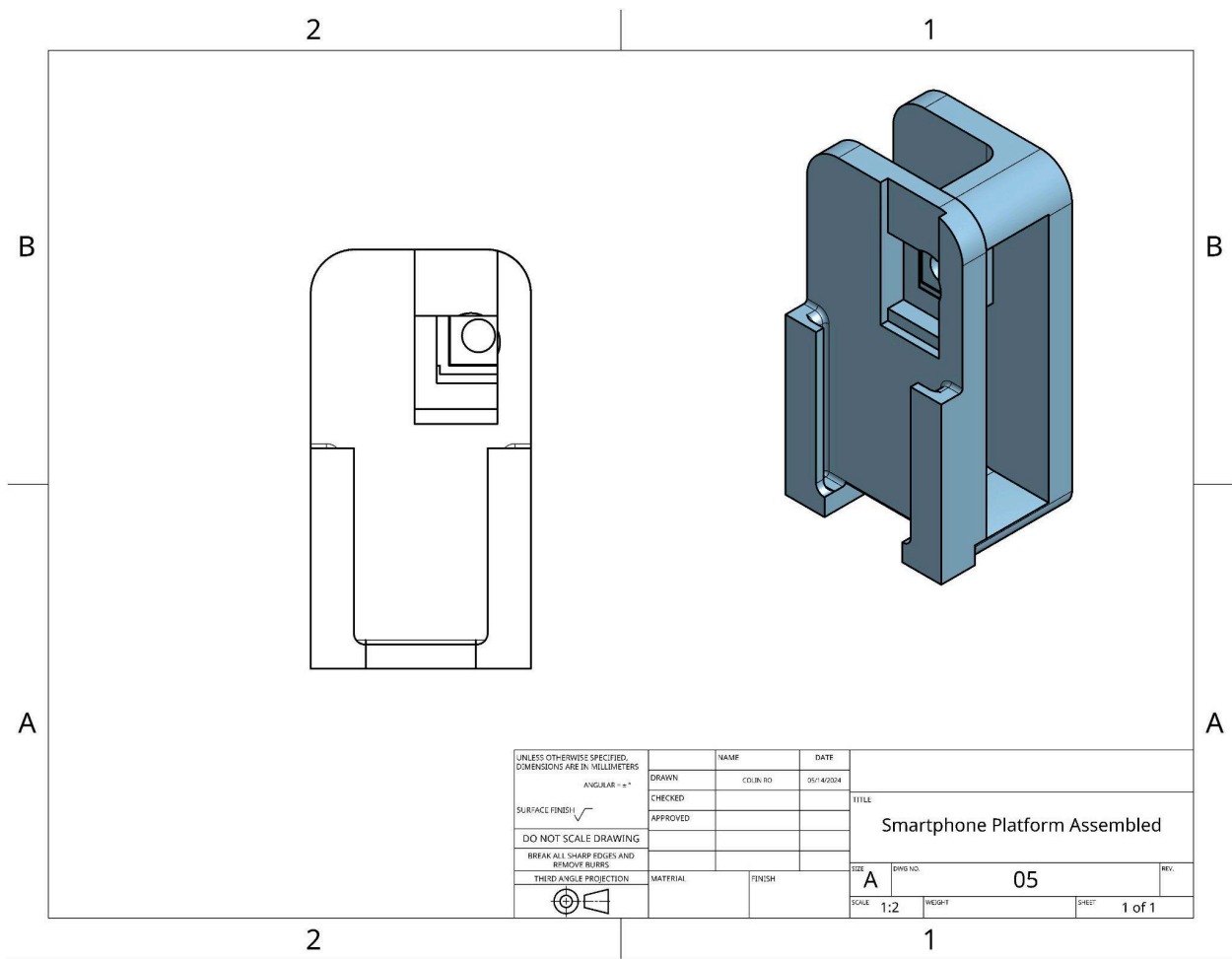


Figure 5. Smartphone Platform Drawing (05)

The full assembly of the SiPM testbench prototype is displayed above. I included an additional cover between the external lens and the smartphone to minimize pass through light.

I would recommend removing the hinge component when 3D printing. When printing, the phone holder requires supports on its underside because of how the hinge protrudes below. The supports are partially permanent and add volume to the underside of the holder, rendering the hemocytometer useless when attempting to elevate it to the external lens's plane.

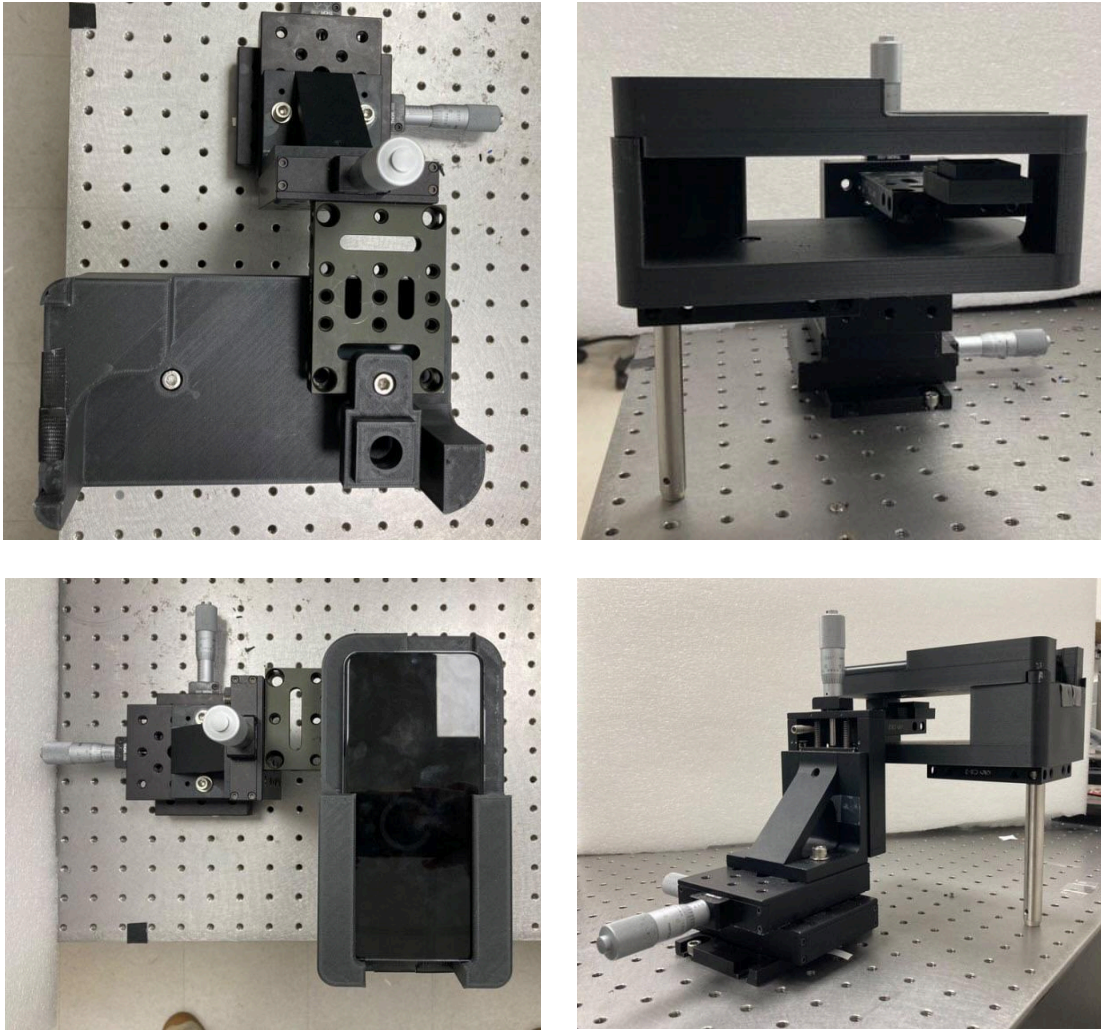


Figure 6. SiPM Test Bench Assembled

The images above display how the test bench is set up. The phone is pressure-fitted into the holder and should not be able to move from its position unless external force is applied to it. In the top left image, a light source (e.g. phone flashlight or ring light in the lab) can be shined from below the sample holder. The Thorlabs XYZ adjustable table can be used to move the sample accordingly.

Component Layout Schematic

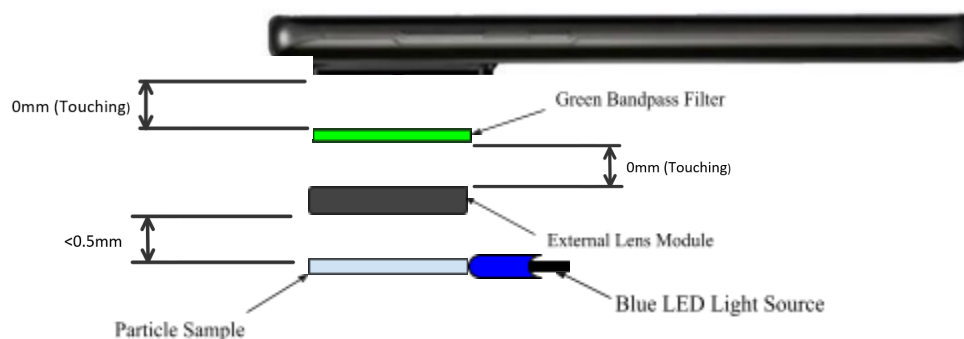


Figure 7. Component Layout Schematic

As seen in the diagram above, all of the components must be neatly stacked together for optimal image quality. You will notice in the SiPM test bench that with all components prepared, the phone's lens is touching the filter, and the filter is touching the external lens module. With the sample loaded, it must be nearly touching the external lens module for it to be visible on the phone screen. Correct spacing between the components is necessary to yield proper image results. When using a fluorescent sample, the blue LED light source must be on the same plane as and as close to the sample platform as possible for maximum particle illumination.

Smartphone Lens Capability

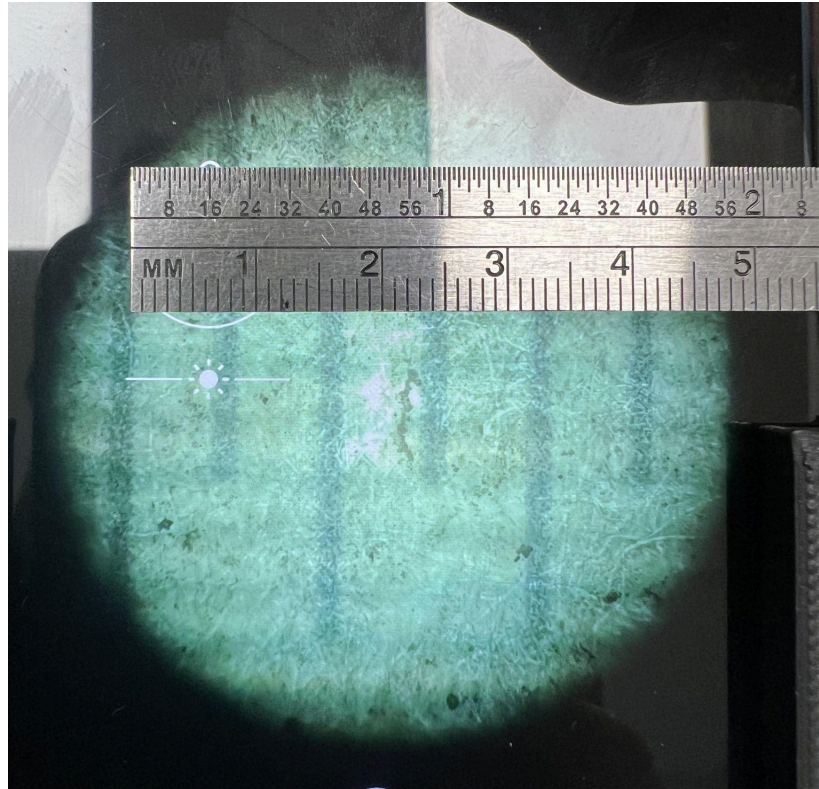


Figure 8. External Lens Zoom Magnification (Phone Camera @ 1x)

When used in tandem with the external lens, the current SIPM Test Bench is capable of an approximate 8.4x magnification ($\frac{42\text{mm on ruler}}{5\text{mm on sample}}$) when the phone's camera is at 1x zoom. Additionally, the phone's zoom function is consistently proportional (e.g. phone camera @ 3x zoom = $\sim 8.4 \times 3x = \sim 25.2x$). When the phone's camera reaches a magnitude too difficult for it to maintain quality focus, it transitions to its bottom-most lens (100x capability). This means that under extremely ideal conditions, the phone camera + external lens could reach a magnification of $\sim 840x$. However, after approximately 10x zoom on the phone's camera, the image becomes noticeably blurry.

Fluorescent Particle Imaging Procedure

When using the hemocytometer as a mode of fluorescent particle visibility, creating accurate dilutions and loading the sample correctly is important to yield consistent results.

Dilution Factor and Particle Concentration

To calculate the particle concentration of the stock fluorescent particle solution, the following formula could be used:

$$N = \frac{6 \cdot S \cdot \rho_L}{\pi \cdot \rho_S \cdot d^3}$$

The stock fluorescent particle solution had a diameter (d) of 10 μ m, a percent weight of solid (S) of 1%, a sphere solution density (ρ_L) of 0.00001g/ μ L, and a solid spheres density (ρ_S) of 1.05*10⁻¹²g/ μ m³. Therefore, the theoretical concentration of the fluorescent particle pure solution was 18189.67281 particles/ μ L. To create a 200x dilution factor, 199 μ L of deionized water was combined with 1 μ L of pure solution (the same way 398 μ L of deionized water can be combined with 2 μ L of pure solution to make the same dilution).

Loading the Sample

To load the sample, use one of the pipettes, ensuring that the volume displayed on its side lists 10 μ L. If not, use the dial on its backside to adjust the volume accordingly. Prepare the hemocytometer by thoroughly cleaning its transparent glass platforms with ethanol and KimTech wipes, and place a square glass cover slide so that it covers both hemocytometer platforms. Using the pipette, pull 10 μ L from the 200x diluted sample. Inject the sample into one platform of the hemocytometer and repeat with the other end.

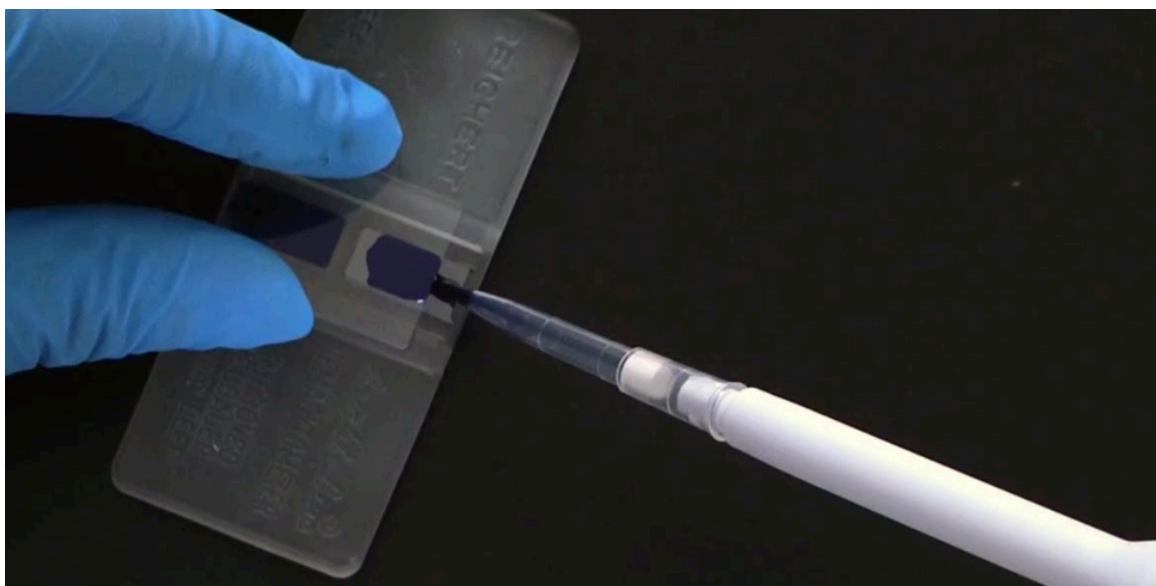
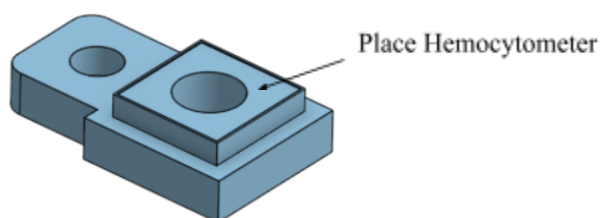


Figure 9. Hemocytometer Preparation

Seeing the Particles

Bring the prepared hemocytometer to the SiPM test bench and place it on the sample platform (Figure 5), lining it up as closely as possible to the external lens. Using the XYZ function of the adjustable platform, line up the hemocytometer with the external lens so that the sample area is fully visible within the phone's field of vision.



Connect the blue LED light source to the provided battery and shine it on the hemocytometer horizontally, ensuring that the room is completely dark. It is important that the LED is on the same plane as the sample and as close to it as possible. Doing this will illuminate the particles as much as possible, allowing for maximum definition.

Collected Data

Fluorescent-coated particles are visible using the SIPM Testbench, as seen below. The image was captured using the hemocytometer placed on top of the sample platform and adjusting it vertically to optimize focus.

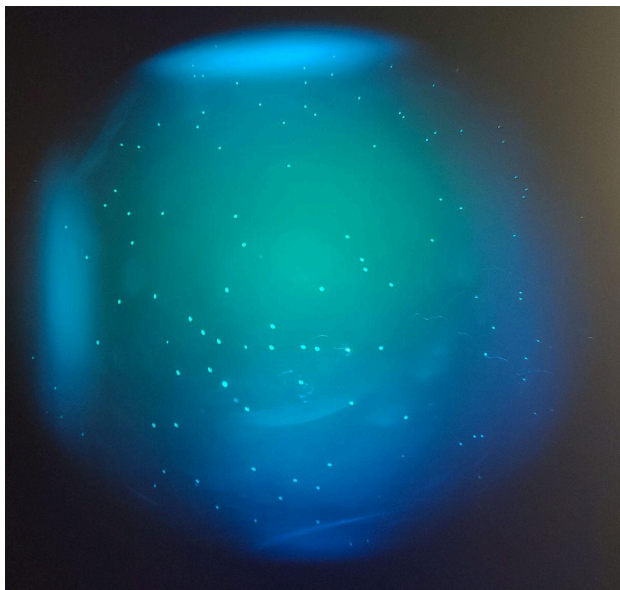


Figure 10. Fluorescent Particle Visibility (1x Camera Zoom)

In Figure 10, the visible area seen through the camera is approximately 6 mm in diameter. This means that the visible volume would be approximately $113.1\mu\text{L}$. In this sample, there were roughly 120 visible particles. In an ideal situation where the diameter of the visible area consistently remains 6 mm, this image yields about 1.06 particles/ μL . 1/9 of a hemocytometer grid has a volume of $0.1\mu\text{L}$. Therefore, Figure 10 yields approximately 10.6 particles for a 1/9 section of the hemocytometer. This result is consistent with the observations using the microscope with its Greenfield function, where the average number of particles per 1/9 of the hemocytometer grid was 10.

Although the fluorescent particles are visible with acceptable definition, this was not a proper representation of Greenfield Microscopy. As seen in the image taken with the microscope below, the user should only see particles over a black background when using this method. This discrepancy would have either been due to light passing through from the LED or from the bandpass filter only being partially effective. Additionally, the image above displays particles that are noticeably more distorted in the middle and reduced in size towards the edge of the visible area. This may have resulted from either how the blue LED light source was directed towards the sample or from the pressure applied to the hemocytometer when in contact with the external lens.

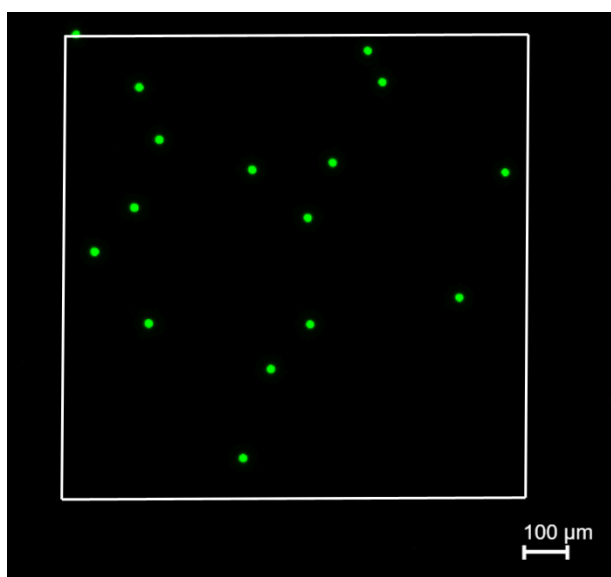


Figure 11. Greenfield Image of Fluorescent Particles on 10x Microscope

The image above using a microscope at 10x zoom displays particles that are roughly equal in size and shape. This consistency makes it easier to identify the particles being tested to make more accurate observations.

One of the challenges in the SIPM Testbench was determining the sample volume. In the microscope image, it is easy to switch between Greenfield and Brightfield microscopy. When switched to Brightfield, the hemocytometer grid is easily identifiable and can be marked. With the SIPM Testbench, the grid on the hemocytometer is difficult to locate, making the sample area much more difficult to determine. Additionally, the camera's focus function may change the visible area on the screen depending on how close or far the sample is. Because there is significant space between the hemocytometer grid and where the particles sit, the sample area changes significantly depending on which one the camera is focused on.

In an attempt to mitigate this issue, I dropped the particles between two glass cover plates and placed them on a 3mm x 3mm grid.



Figure 12. Glass Cover Plates over 3mm x 3mm Grid (1x Camera Zoom)

There are noticeably less fluorescent particles shown in the image above since I used two glass cover plates with unknown volume instead of the hemocytometer. However, I wanted to emphasize that if the grid and particles are nearly on the same plane, they can both be in focus. Granted, the grid should not be visible if the technique was properly executed, but this could serve as a starting point for future experimentation. *For example*, if the grid also had fluorescent coating, it would effectively eliminate this issue since both the grid and particles could be seen with high definition.

Future Challenges

The primary challenge I faced throughout this semester was mitigating light passing through the filter, either directly or from the sides. Though I was unable to eliminate this problem entirely, fluorescent particle visibility is certain. If full removable of pass-through light can be achieved, the background of a Greenfield image using the SiPM platform should be entirely black, and particle visibility will be enhanced significantly.

Furthermore, the images taken on the SiPM platform display particles with distorted sizes and shapes. This is likely due to the sample being pressed up against the external lens for maximum image definition. If a platform that minimizes the pressure applied to the particles can be fabricated, there will likely be much less distortion in the visible particles.