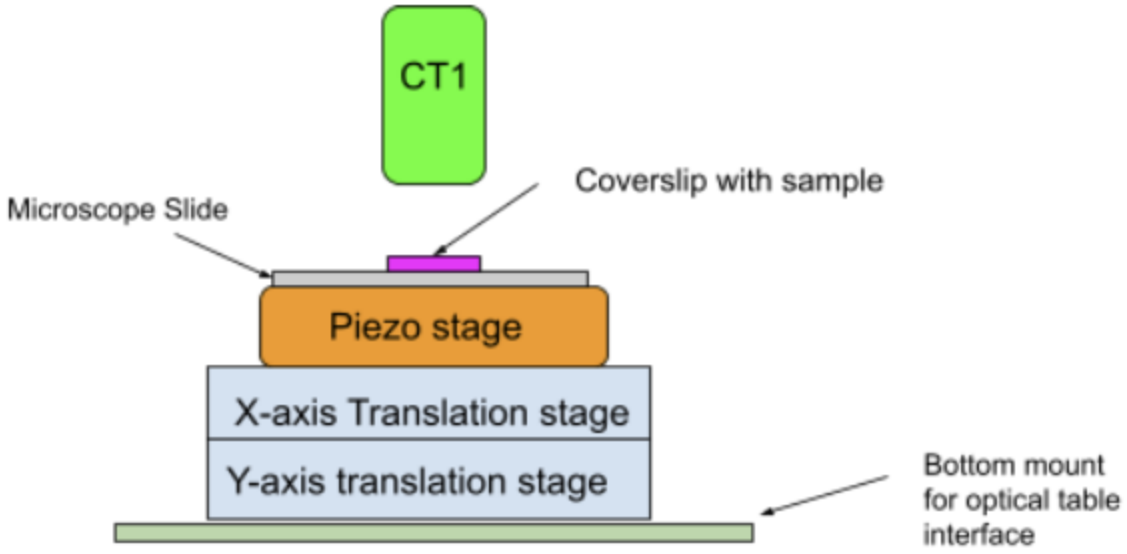
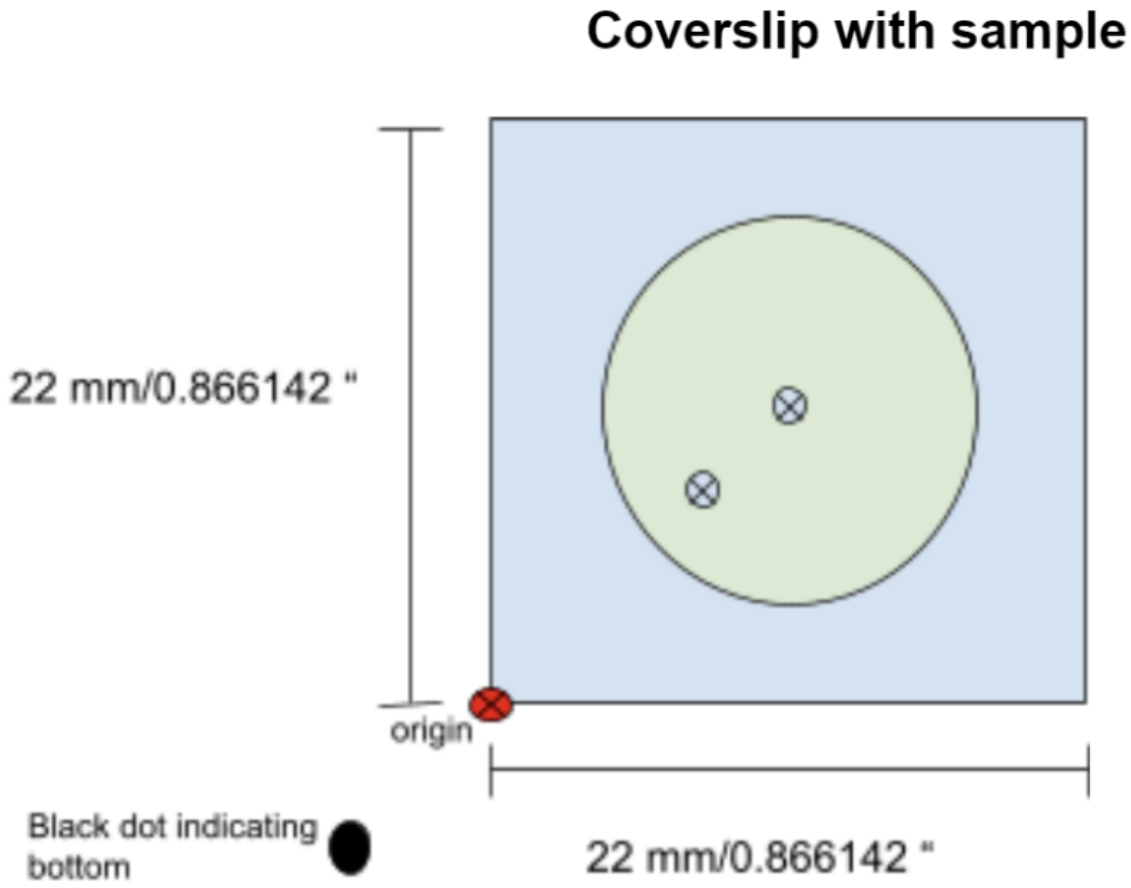
Sample Calibration Procedure

Setup:



- Given a standard microscope slide and coverslip with nanoparticle sample and manual 2-axis translation stage. The piezo was required to mount on top of the translation stages to allow for focal length to be within CT1 range.
- Make sure that a VOA is implemented into your fiber system feeding into the collimator of the microscope.
- Generally speaking, the piezo stage was only used to get the sample within reach of the CT1's focal length, and could be substituted in the case that this was still accomplished.

Coordinate System:



- The bottom left corner of the coverslip (as seen from the CT1 top view) serves as the origin.
- The manual translation stage adjusts with thousandths of an inch precision, which means many turns to get to the center of the coverslip, however, another “square” coordinate: (0.3,0.3) can still guarantee sample penetration from the beam and takes much less work to get there.

Procedure:

1. Use a red laser light in place of 1550 through the collimator to locate the origin point. Ensure the detector is off through this process. An accurate way to accomplish this is to start the red light beam position slightly off of the coverslip and approach one of the edges with the translation stage adjuster. Make sure the red

light is relatively focused. When the red light hits the edge, the light will diffract and “light up” the coverslip. You can back it up and verify the edge as soon as this happens. From the other direction, move the translation sage closer to the bottom left corner position, and approach the edge, stopping at the position that this diffraction instantaneously occurs. Back off the previous direction and re-verify the corner as you approach with the red beam. When the beam is positioned at the very edge of the corner, the diffraction will brighten and show lines of red light through the coverslip. This serves as the (0,0) coordinate.

2. Take note of the measurements on the multimeter of the translation stages and from this position move the square units previously chosen as the sample point. Since (0.3”, 0.3”) was chosen, move an additional 0.3” in each direction into the sample to accomplish this coordinate change from the origin.
3. Replace the red light with the 1550 nm RIO pump fiber into the collimator. Plug the detector into the oscilloscope. Turn the lights off. Turn on and enable the RIO pump at 28 mV power. (This power can be adjusted but must remain constant through data collection for calibration purposes). Turn on the detector.
4. Use the oscilloscope to focus the 1550 nm beam onto the sample at the chosen point. Then attenuate the signal with the VOA until the measurement is reasonably under the saturation point. Once the signal is attenuated, it will not be changed throughout data collection (if the experiment is to show repeatability). Otherwise the powersweep code is more efficient in recording these measurements based on power. Record the output voltage readout on the oscilloscope (average voltage).
5. To show that this is repeatable to a satisfactory percentage of output voltage and verify the calibration point can be found again, we reset the system. To do this, we turn off the detector, disable and disconnect the 1550 nm pump.We then reconnect the red light source, and defocus the CT1. Without changing attenuation, we move the translation stage back to the bottom left corner of the coverslip, slightly off of the coverslip corner. Repeat steps 1 through 4, recording the output measurement each time.

Resulting Data:

# Trial #	Aa Origin point found at(inches)	Target coordinate (inches)	# Output Voltage (V)
1	(0.065, 0.7211)	(0.30 0.30)	3.756
2	(0.064, 0.7208)	(0.30 0.30)	3.845
3	(0.066, 0.7215)	(0.30 0.30)	3.679
4	(0.065, 0.7202)	(0.30 0.30)	3.82

If we take the first measurement as a base, the following measurements can be calculated to be within plus or minus 0.09 V of the base measurement. This is 2.4% error of the base measurement.

Alternatively, the largest difference in voltage found in this trial was 0.166 V, which is at worst, 4.5% error of the measurement.

- NOTE: the data above was taken using attenuation with the irises rather than the VOA but the irises were not adjusted after the first measurement and should be valid for this purpose. The procedure described above should be followed for any further data collection

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More trials can be completed in order to get a more accurate picture of the error and its implications of the repeatability of finding this calibration point.