

Laser-Scope: The Affordable Confocal Scanning Laser Microscope for Surface Topography

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Abstract — High-resolution microscopy plays a crucial role in advancing research across fields such as biology, photonics, material science and many others. Among the most powerful imaging techniques is Confocal Laser Scanning Microscopy (CLSM), which enables two-dimensional imaging by eliminating out of focus light and isolating precise optical sections of a sample. However, CLSM systems have a high barrier of entry – cost. To address this limitation, the Laser-Scope project was conceived and developed to deliver an accessible, low-cost CLSM system (< \$1000 USD) that retains key functionality of commercial alternatives while remaining open-source, compact and modular.

Index Terms — Two-dimensional displays, time division synchronous code division multiple access, microscopy, optical design, lasers

I. INTRODUCTION TO LASER-SCOPE

Taking inspiration from the working concepts of confocal laser scanning microscopy (CLSM), Laser-Scope achieves medium to high resolution for images on the micron scale through the specific case of reflection microscopy. Reflection microscopy is used directly in surface topography since light reflections off certain materials can be read easily by a photodiode. This differs from some of the traditional techniques used in microscopy, such as fluorescence microscopy. In biological research, fluorescence microscopy is used to achieve a greater depth of field of an organism by exciting the fluorescence liquid with a laser beam and then reading the excited light through a photodiode [1]. With a complete open-source design, Laser-Scope has to remain simple enough to be replicated, while achieving the same level of precision that can be found in most commercial CLSM systems.

Laser-Scope is comprised of three integrated subsystems: photonics, electrical, and software. The photonics subsystem consists of an array of optical lenses, a 405 nm laser diode, and a high-responsivity photodiode. The photonic system is responsible for directing, focusing, and collecting the light reflected from the sample. The electrical subsystem will enable precise control of power, signal conditioning, stage actuation, and serve as the bridge between the photonic and software subsystems. The software subsystem will act as the interface between the system and the user, managing the scanning process and image reconstruction from raw data.

Utilizing manufacturing techniques that are available to most students and researchers is the biggest advantage of this project that keeps cost low. One of the main parts this project revolved around was the use of an open-source 3D printed microscope stage from OpenFlexure called the Delta Stage. From there, optics, electronics, and software were implemented to achieve desired specifications of micron level resolution with an imaging process speed of less than 15 minutes.

II. FEATURES AND FUNCTIONALITY

The functionality of Laser-Scope can be described by three main parts: stage functions, microscope head functions, and image generation functions.

The microscope stage enables the precise movement of a sample under illumination by the laser. This is achieved by utilizing readily available stepper motors and the open-source OpenFlexure Delta Stage to deliver controlled, high resolution linear motion. Since the scanning process depends on synchronizing the stage's motion with the laser position, this setup enables flexible scanning modes—either point-by-point or line-by-line. Moreover, the stage is engineered for reliability, ensuring consistent performance and repeatability across all scans.

The microscope head houses all of the optical elements in the system including a beam collimator, beam expander, collecting lens, aperture, beam splitter, microscope objective, laser diode, and photodiode. The laser diode emits a beam that is passed through a 1mm aperture to ensure the beam is spherical, then passes through a collimation lens to stop the beam from diverging in the system. Once the beam is collimated it is transmitted into two beam expanding optics to create a beam size with 5x magnification, it is then reflected by the beam splitter into an 20x magnification microscope objective allowing the beam to reach a spot size approximately 1 μm in diameter. The reflected light is then transmitted back through the microscope objective, then the beam splitter, and then into the collecting lens which focuses the beam through a 1 mm

aperture which effectively cuts out any optical noise from the reflected beam. Lastly, the beam is incident onto the active area of a photodiode, the beam signal is then amplified by a transimpedance amplifier, which allows for the change in the intensity of the optical signal to be recognized as a voltage change across the transimpedance amplifier.

To fully understand the position of the optical components in Laser-Scope, a schematic of the optical layout is shown in Fig. 1.

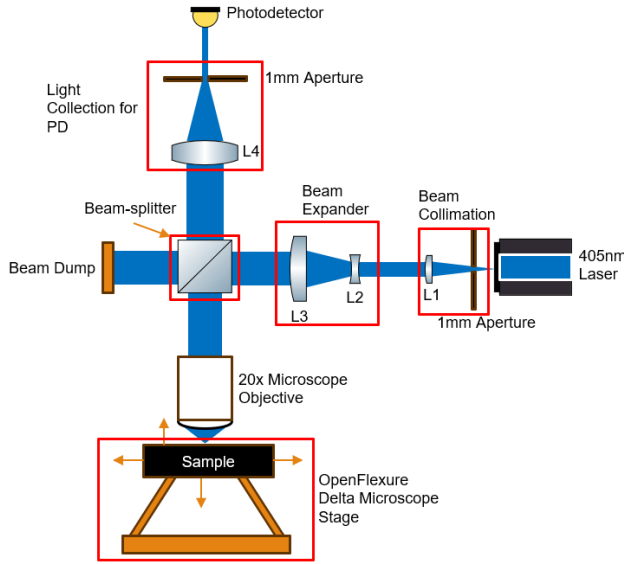


Fig. 1. Optical schematic shows the transmission of the laser beam through the system onto a sample under illumination and back through the system

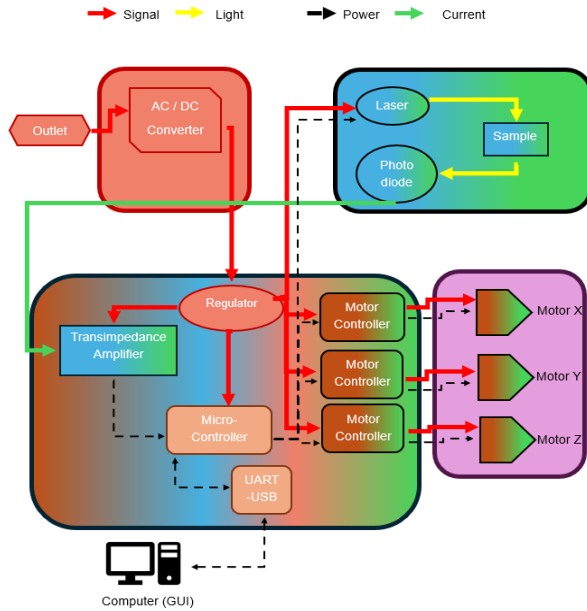


Fig. 2. Overall system block diagram of Laser-Scope

The image generation utilizes the voltage change through the transimpedance amplifier and then digitizes the signal through the microcontroller. The digitized values are read and saved per-point, which is an average of up to 1000 data points per-point, along each line of a 64 x 64-pixel image.

To be understood as a complete system, a block diagram format of the overall system is shown in Fig. 2.

III. SUBSYSTEM COMPONENTS

A. Laser and Laser Driver

The Thorlabs L405P20 laser diode is a compact light source while being suited for microscopy applications. The L405P20 laser diode comes in a standard $\varnothing 5.6\text{mm}$ TO package, with an emission wavelength at 405nm. Divergence of the laser diode has a typical parallel beam divergence of 8.5° and a typical perpendicular beam divergence of 19° . While the diode output is elliptical, it is fixed by transmission through first 1mm aperture at a distance of 23 mm to the diode's surface.

The LP405P20 laser diode can be tuned using the laser driver to output between 5mW and 20mW of optical power. This tunability is important for calculations made for the strength of the signal on the receiving end of the optics array.

To ensure the diode achieves precise wavelength stability and to prevent thermal runaway the LM317 is utilized to regulate its current. The LM317 is configured to a constant-current mode, where 1.25V are kept across a 39Ω resistor connected between the adjust and output terminals. This creates a stable 31mA source for the Thorlabs L405P20 laser diode, ensuring reliable and stable operation even during extended operation.

B. Optics

The first optic that the beam generated by the laser diode encounters is a 1mm diameter circular pinhole. This 3D-printed component takes the diverging beam and resizes and shapes it. This is an important treatment to begin defining the beam into a usable configuration. Next, the beam is incident on the LA1610-A collimating lens, a plano-convex lens which ends the divergence of the beam and gives it a consistent diameter over a long length. This collimated beam is transmitted into a beam expander, which is a two-lens system consisting of biconvex lens LD2060-A and plano-convex lens LA1608-A.

The newly collimated and magnified beam propagates to the next optic in the array, which is the BSW10R beamsplitter, a 50:50 beamsplitter that transmits 50% of

the incident light at the chosen wavelength and reflects the remaining 50% at a 90-degree angle. This optic serves to redirect the beam from the first leg of the microscope into the second leg, which houses the microscope objective. The reflected beam from the beamsplitter propagates to the entrance pupil of this microscope objective, the AmScope PA20X-INF objective. This optic serves to focus beam down into a spot with a size comparable to the desired pixel size of the system, which will be incident on the surface of the object being imaged. The reflections off of the object will be recaptured by the objective, collimated, and directed backwards through the system. The recaptured beam will propagate back to the beamsplitter, where it will experience another 50% loss of optical power.

The transmitted beam is then en-route to the photodetector but will first be focused through the system's collecting lens, the LB1596 biconvex lens. The focused beam is directed through a final 1mm diameter pinhole, and it is this feature that earns Laser-Scope its designation as a 'confocal' microscope. This pinhole is another 3D-printed component, and it functions as a filter for the focusing beam. A significant proportion of any noise or parasitic interference that may be propagating alongside the optical signal will be out of focus with respect to the signal. This means that during convergence, the precise alignment of the intended signal allows it to pass through the pinhole, while the less-focused noise will be blocked and filtered out, increasing the resolution and signal quality of the system. Finally, the beam will be incident upon the photodiode, with a spot size smaller than the 1.1mm x 1.1mm active region of the device.

C. Photodiode and Transimpedance Amplifier

The FD11A Photodiode is an optoelectronic device that converts an optical signal into an electrical one in the form of current. This device has a responsivity of 0.17A/W at the chosen wavelength of 405nm and an active region of 1.21mm². When the object being imaged under the microscope objective is within the objectives focus, the photodiode will measure the strongest signal; inversely, as the object moves away from focus, the strength of the signal will decrease.

The photodetection stage utilizes an OPA380 transimpedance amplifier that converts the photodiode's photocurrent into a measurable voltage. The OPA380 features ultra-low input bias current (0.2pA) and low noise density ($5nV/\sqrt{Hz}$), making it well-suited for high-sensitivity optical measurements. Configured with a feedback resistor of 1M Ω and a parallel feedback capacitor of 33nF, which set the transimpedance gain and stabilize

the frequency response. Additionally, the + input of the OPA380 is biased with 1.65V to enable the ESP32 to easily detect the signal. This configuration provides low noise and adequate bandwidth for the scanning speed of the system while maintaining signal integrity.

D. Microscope Stage

The design for the 3D-printed microscope stage was the OpenFlexure Delta Stage, an open-source schematic available online for easy printing and assembling. Using three stepper motors and a gear assembly, this stage can movement of 9mm in the x-direction, 12mm in the y-direction, and 5mm vertically from its center position. Manipulation of these stepper motors can be used to accurately and repeatably achieve sub-micron motion of the stage, meaning that the system can precisely replicate the 16-micron increments necessary for the measurements being taken.

E. Microcontroller

The microscope's motion and the way we gather data is managed by the Espressif ESP32-WROOM-32 microcontroller. This MCU is a dual-core 32-bit device that works at 240 MHz that allows for precise motor movement and coordination, ADC voltage sampling, and serial communication with the graphical user interface (GUI). The ESP32 utilizes a control loop that receives orders from the GUI sequentially.

The firmware in our project directly controls three 28BYJ-48 stepper motors through ULN2003 driver boards. Each of the motors follows an 8-state half-step using inverse kinematic matrix. This kinematic function allows translation of the stage's trapezoidal ramping. The motion is commanded in cartesian millimeters which will then map to the corresponding X,Y,Z displacement.

Communication between the ESP32 and the computer interface is handled through a serial link operating at 115200 baud. The commands that the GUI sends is through ASCII commands from the GUI. Each command is parsed in real time, allowing for immediate response and no need of interrupts. This format allows for minimal overhead and high responsiveness for both manual and automated stage operation.

Analog data acquisition is integrated within the same control loop inside the firmware loop. On the ESP32's GPIO we selected the GPIO pin 36. This pin captures the photodiode voltage reading and has the capability to be average across large samples.

Data capture in the system using the serial output line with specialized tags like: DATA, PROGRESS, or DONE

to allow for the GUI to record the measurements synchronously along with the stage position.

F. Power Supply

The system's power supply is implemented via a cascaded regulation architecture consisting of the Mean Well LRS-50-12, LM2596-5.0 and LT1085-3.3. The Mean Well LRS-50-12 is an enclosed AC-DC converter that provides a regulated 12V DC output at up to 4.2A from a universal AC input and serves as the primary source for all downstream converters. The LM2596-5.0 buck regulator steps the 12V rail down to a regulated 5V and can source up to 3A with up to 92% efficiency along with over-temperature and over-current protection. This 5V rail feeds the LT1085-3.3 low-dropout linear regulator which will source a clean 3.3V supply for sensitive digital logic and mixed-signal components. Through this architecture low noise, thermal stability and high efficiency can be achieved.

IV. SYSTEM CONCEPTS

A. Scanning Speed

The overall scanning speed of the system is determined by the total scan duration and the number of pixels acquired during image formation. This can be expressed by (1) where t_{total} is the total scan time in minutes and $x_{total\ pixels}$ represents the total number of sampled points in the image.

$$Scan\ Speed = \frac{(t_{total} \times 60 \frac{sec}{min})}{x_{total\ pixels}}. \quad (1)$$

For a 64×64-pixel image (4096 total points) with a total scan time of approximately 12 minutes, the system achieves a per-pixel sampling time of 177ms. This measured time per point establishes the true settling window for the transimpedance amplifier and is the basis from which the feedback resistance and capacitance must be considered.

B. Transimpedance Amplifier

The quality of the signal provided by the transimpedance amplifier directly influences the quality and fidelity of the generated image. Therefore, several factors must be considered to maximize quality.

Bandwidth will determine the amplifier's ability to accurately reproduce changes in the photocurrent over time. The bandwidth of the system can be approximated

by (2) where R_f (1 MΩ) is the feedback resistor and C_f (33nF) is the feedback capacitor

$$f_{bw} = \frac{1}{2\pi R_f C_f}. \quad (2)$$

With this, we achieve a bandwidth of 4.82 Hz. Given a per-pixel sampling time of 177ms this bandwidth ensures that the transimpedance amplifier fully settles before each sample is recorded and as a result the measured signal accurately represents the steady-state photocurrent at each point.

To further ensure that the bandwidth is sufficient the theoretical settling time must also be calculated. The settling time defines how long the amplifier output requires to stabilize within a specified error band following the abrupt change in the input current at each point. This can be approximated by (3) where R_f is the feedback resistor and C_f is the feedback capacitor

$$t_s = 4R_f C_f. \quad (3)$$

With this a settling time of 132ms is achieved which confirms the system's bandwidth is enough to accurately capture the steady-state photocurrent.

A final consideration for the system's transimpedance amplifier is the signal-to-noise ratio (SNR) which quantifies the ratio of the desired signal level to the unwanted noise and will serve as a key indicator for image clarity, this can be calculated via (4), where $V_{signal,RMS}$ corresponds to the mean output voltage measured when the laser is active, and $V_{noise,RMS}$ corresponds to the standard deviation of the amplifier output when the laser is covered.

$$SNR = 20 \log_{10} \left(\frac{V_{signal,RMS}}{V_{noise,RMS}} \right). \quad (4)$$

By sampling 10,000 data points for each condition the calculated SNR at the ADC input was 48.7 dB, which indicates low overall output noise relative to the signal voltage range.

C. Beam Generation and Handling

The entirety of the optics array resides inside the microscope head of Laser-Scope. Utilizing the wavelength of 405nm and a consistent output of the Thorlabs LP405P20 laser diode allows a theoretical resolution of 617.6nm found from (5).

$$R = \frac{1.22 \times \lambda}{2 \times NA}. \quad (5)$$

While Laser-Scope has the ability to reach this resolution in theory, it is almost impossible to achieve this resolution in practice. Due to the amount of factors that can affect the

actual value, Laser-Scope was given an estimated resolution to achieve of $10\mu\text{m}$. This is much closer to what is seen in practice of using this system.

The sensor responsible for detecting the transmitted optical signal is the Thorlabs FD11A photodiode. This device has a responsivity of 0.17 A/W at the chosen wavelength of 405nm and an active region of $1.1\text{mm} \times 1.1\text{mm}$. In order for the transmitted optical signal to be strong enough to overcome the noise floor and provide a significant value it was determined that the laser diode should be driven at $\sim 20\text{mW}$.

Between signal generating and receiving ends of the system are all of the optics designed for handling the beam. After the laser outputs a beam, it is transmitted through a $\varnothing 1\text{mm}$ 3D printed pinhole. This pinhole is a distance of 11mm from the LA1610-A collimation lens, which allows for the beam to expand to $\varnothing 1.4\text{mm}$. The collimation lens keeps the beam at a steady diameter until it reaches the first of two beam expanding optics. With the entrance pupil of the PA20X-INF microscope objective measuring $\varnothing 7.2\text{mm}$, the beam expander needs to achieve a $5\times$ magnification to be approximately the same size as the entrance pupil. The calculated beam size after the beam expander was $\varnothing 7\text{mm}$. Using a Galilean beam expander design shown in Fig. 3, with a length of 60mm between the two lenses found from (6).

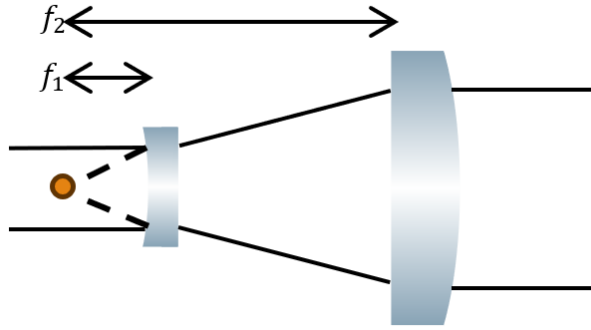


Fig. 3. Simple Galilean beam expander design where f_1 is the focal length of the first lens and f_2 is the focal length of the second lens

$$L = f_2 + f_1. \quad (6)$$

At this point in the optical array, the beam has been shaped, collimated, and magnified to an appropriate size to be used for sampling. A 50/50 beamsplitter, which transmits 50% of light, at the chosen 405nm wavelength, through it and reflects the remaining 50% at a 90-degree

angle, redirects the beam into the entrance pupil of the PA20X-INF microscope objective.

The input beam into the PA20X-INF microscope objective is focused down to a spot size that is less than the pixel size of $15.6\mu\text{m}$. The beam waist diameter found was $\sim 1.27\mu\text{m}$ which is less than the pixel size. This can be seen in the formulas shown by (7) and then plugged into (8), where NA_{eff} is the “used” NA area, EP is the entrance pupil, and D is the diameter of the input beam. The effective NA comes out to 0.3889 , which gives an airy disk diameter (d_{spot}) of $1.27\mu\text{m}$.

$$NA_{eff} = NA\left(\frac{D}{EP}\right). \quad (7)$$

$$d_{spot} = \frac{1.22 \times \lambda}{NA_{eff}}. \quad (8)$$

Once the beam is transmitted and reflected back through the microscope objective, the LB1596 collecting lens focuses the $\varnothing 7\text{mm}$ beam down to $\varnothing 4.42\mu\text{m}$ found from (9) [2]

$$Spot (mm) = \frac{4 \times f(mm) \times \lambda(mm) \times M^2}{\pi \times Beam \text{ Diam. at Lens}(mm)}. \quad (9)$$

By using a second 1mm aperture before the beam completely focuses, the outer edges of the beam can be effectively cut-out of the transmission directly to the photodiode. In doing so, this allows for a less photobleached image (or less saturated in other terms), which can account for a more accurate measurement.

D. Motor Controls

The motion of the OpenFlexure Delta Stage is driven by three 28BYJ-48 stepper motors controlled through the ULN2003 Darlington transistor arrays. Each ULN2003 module provides the required 4 outputs capable of sinking up to 500 mA per channel, allowing for control of the stepper motor coils from the ESP32's IO pins.

Motor motion is achieved by sequentially energizing the four IN channels of the ULN2003 through timed pulses via 4 digital GPIO pins in the ESP32, as such the ULN2003 switches each corresponding transistor to ground and allows current to flow through the motor windings. Direction is determined by the order in which the coil sequence is activated, while speed is controlled by the delay between successive pulses. Utilizing this configuration the stage can achieve accurate and repeatable

position which is required for raster scanning of the desired sample.

V. SOFTWARE DETAIL

A. MCU Programming

The ESP32 microcontroller is programmed using the Arduino IDE, which is a lightweight C++ environment for embedded development. The IDE simplifies firmware deployment, library management and integration, and serial debugging. The code is uploaded to the board via USB using Espressif's esptool bootloader and serial communication is monitored through its built-in console at 115200 baud rate.

The firmware is organized into modular functions that consists of motor control, ADC voltage values, Command interpretation, and various other helper functions. The main loop in the program follows the Arduino structure of "setup()" and "loop()" function. The main initialization routines in the set up portion allow for serial interface configuration, ADC input, and the output pins for all the three stepper motors. After the setup portion the system enters the looping portion where all of the serial commands are received, parsed, and executed in a sequential fashion.

Each of the high-level commands sent by the GUI corresponds to a firmware handler. Some examples of the commands are as follows

- 1) "MRMM dx dy dz": Performs the motion in millimeters in cartesian.
- 2) "GOTO_MM x y z": Moves the stage to the absolute coordinate. Absolute coordinate indicates the center point with respect to the stage boot up in the start
- 3) "SCAN_START": Initiate an automated raster scan (snake like movement left to right) where the system moves point-by-point while sampling ADC voltages.
- 4) "RETURN_ORIGIN": Lifts and returns the stage to its original position.
- 5) "GET_ADC" or "MEAS_AVG": Performs single or averaged voltage measurements for calibration or testing.

The program's motion subsystem converts millimeter-based coordinates to motor half-steps using the system's inverse kinematic transformation matrix. The routine for this is called "move_axes()" where all three motors are synchronized by their displacement magnitudes at every step. Taking this approach allows for smooth motion with no need of concurrency.

The analog data collection routine in the system needs the ESP32's ADC1 channel (GPIO pin 36) to collect photodiode readings. When sending the ADC values to the GUI it gets sent with timing information to track sampling

performance. When we utilize the averaging functionality it combines multiple readings to minimize noise and its results are sent to the host in real time in a formatted serial output. The output in this system is a CSV (comma separated value) file where the GUI can process the file into a grayscale image of the scanned sample.

B. Graphical User Interface (GUI)

The desktop interface is implemented using Java (Swing) and communicates with the ESP32 over a serial link at 11500 baud using the jSerialComm library. The GUI exposes three primary panels: A connection panel, where the user gets to select the port selection, baud configuration, and connect / disconnect, A motion panel, where the user can move the stage to calibrate the position, and a scanning panel, where the user can select the parameters needed for a scan.

The GUI receives lines that are parsed by a lightweight system that can recognize the MCU's structure outputs. Readings from the MCU that are tagged as "DATA" are specialized for data formatting (Image generation purposes, formatted as: position, Raw ADC, mV and V). These tags that the GUI listens for, allow simple debugging and indicators of progress. The GUI mirrors the MCU console for traceability and logs CSV-ready rows to assist in image reconstruction.

The scanning panel writes to the firmware parameters, the user controls the values for the dimensions (1mm by 1mm or 5mm by 5mm), what the step size should be (how far should the stage move to get the next point) and how many samples to take at a specific point. Additionally, the scan panel allows for the user to see the progress of the scan and abort the scan at any point during the scan. After aborting the scan, it will then return to the original point ready to perform the next function on the GUI.

The GUI's architecture is designed to keep it responsive under continuous serial traffic. The GUI's in-memory buffer is used later by image generation to make the grayscale map by using the millivolt values.

C. Delta Stage Movement

The microscope's precision positioning system utilizes a three-motor delta stage, each is driven by a 28BYJ-48 stepper motor through a ULN2003 driver board. The delta geometry of the stage provides compact symmetric motion which is ideal for our micron scale movement. Attached to each stepper motor is a gear with 12 to 24 tooth reduction pair (2:1 ratio).

The ESP32 firmware defines specific 8-state half step sequences for the ULN2003 to enable smooth directional motion. Each motor with the included gear pair has 8192 stage-shaft steps per revolution. The resolution provides

sub-micron displacement capability across the stage's 10-mm soft travel range. To coordinate all three axes, the firmware implements a mathematical inverse kinematics matrix that maps Cartesian displacements. This is the following matrix referenced in the firmware (10).

$$[X, Y, Z]^T = \begin{bmatrix} -\cos(30) & \cos(30) & 0 \\ \cos(60) & \cos(60) & -1 \\ 1 & 1 & 1 \end{bmatrix} [a, b, c]^T. \quad (10)$$

The 3x3 constant matrix is embedded in the firmware and scaled by 8192 steps/rev.

The firmware continuously tracks the current position in both millimeters and motor steps. These are referenced as absolute and relative movement modes. Hardcoded in the firmware prevents the user from exceeding the range that the stage can support. During scans, it is calibrated to not go past a 5mm-by-5mm area, leaving this area will cause the gears to stall and eliminate a motor needed for stage movements.

D. ADC Gathering

When measuring the light intensity from the microscope's photodiode the system uses ESP32's 12-bit ADC1 configured to the GPIO 36. The channel in this pin gives a digital range of 0-4095. The ADC's configuration is performed during initialization by the "adc_init_optimal()" function, which sets the resolution, input attenuation, and performs a calibration read to stabilize the conversion path before the first measurement. When performing a scan the user can set how many ADC readings to receive at a point, allowing to average out the values and reduce noise.

E. Image Generation

The final part of the systems operation would be how the system generates the image. The collected voltage readings are stored in a CSV file and the GUI parses through the entries and stores the data in a 2D array. Each of the recorded values represents the photodiode's analog response to reflected light. The voltages are scaled to a 8-bit grayscale range (0-255) using (11) across each pixel.

$$I(x, y) = 255 * \frac{V(x, y) - V_{min}}{V_{max} - V_{min}}. \quad (11)$$

$I(x, y)$ is the output pixel intensity and the V_{min} and V_{max} are the minimum and maximum recorded voltages across the dataset. The user would be able to select any

CSV file that has been formatted accordingly and process the file into a PNG file.

VI. SYSTEM PERFORMANCE AND CHARACTERIZATION

A. Optoelectronics Feasibility Study

To determine if the photodiode-transimpedance amplifier circuit would provide enough range and stability to produce reliable and repeatable topographical images an evaluation of the output signal was performed. The photodetection response was characterized by measuring the transimpedance amplifier output on a micron-by-micron basis utilizing a translation stage. The resulting voltage response shown in Fig. 4, shows a gaussian-like intensity profile, where the signal strength peaks at focus and symmetrically decreases as the sample moves away from this point. This behavior validates that the system's

optics, photodiode and transimpedance amplifier provide sufficient response in range and function as expected.

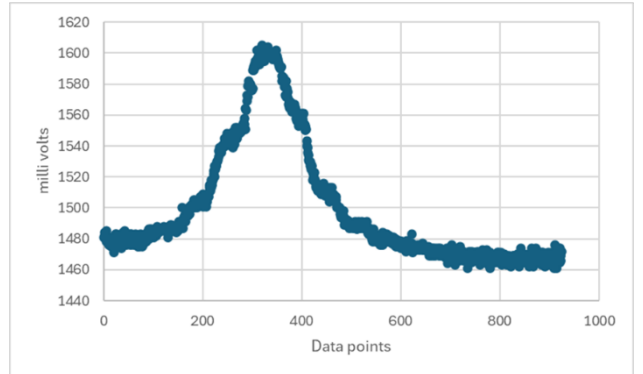


Fig. 4. Transimpedance amplifier gaussian-like response observed, where asymmetries can be attributed to manual movement of the translation stage.

B. Entrance Beam Waist at Focus of Microscope Objective

The generation of the beam and subsequent pinhole filtering, collimation, and expansion should theoretically result in a collimated circular beam of diameter of 7mm entering the microscope objective. In order to confirm this parameter and to test the spot size of the beam waist at focus, the following performance was conducted:

The stage was removed from beneath the microscope head and replaced with a Blackfly S BFS-U3-120S4C-CS 12MP camera whose sensor surface was aligned to 1.79mm out of the focus from the objective, shown in Fig.

5 and the computation of the said image is shown in Fig. 6.

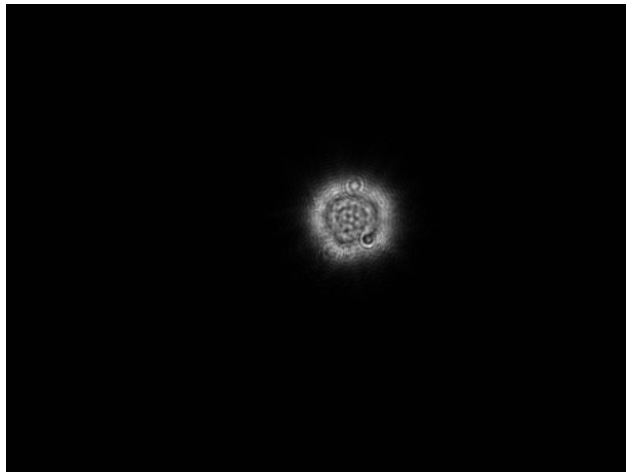


Fig. 5. Image of the output beam at 1.79mm from the focal point, taken on a Blackfly S BFS-U3-120S4C-CS 12MP camera

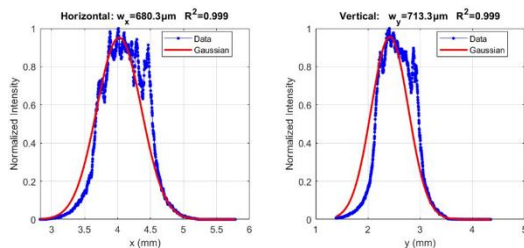


Fig. 6. Beam waist radius at a distance of 1.79mm from the focal point of the PA20X-INF Microscope objective lens

C. Laser Diode Thermal Performance

To evaluate the thermal stability of the Thorlabs L405P20, its drive current was monitored over time while operated under the LM317 constant current configuration as well as the diode's temperature. After 5 minutes of operation, the driver remained capable of feeding the laser diode constantly with no measurable variations and the temperature of the laser diode settled at $\sim 50^\circ\text{C}$ and showed

no signs of overheating or photobleaching. Data acquired from this test can be seen in Fig. 7.

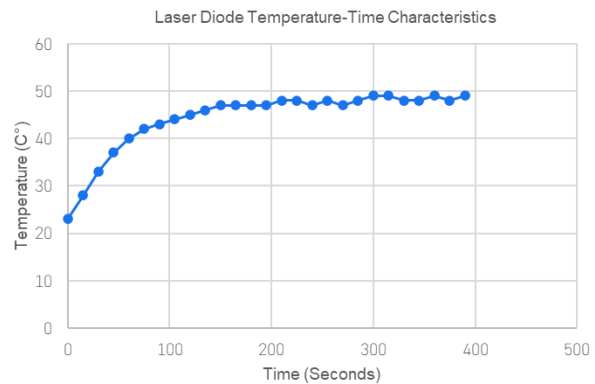


Fig. 7. Temperature vs Time of Thorlabs L405P20 laser diode when driven at 31mA via an LM317 in constant current configuration

VII. CONCLUSION

In conclusion, the CLSM project represents a comprehensive engineering effort spanning analog design, embedded systems, motion control, and system integration. By addressing both the technical requirements and real-world constraints, this design lays a solid foundation for a fully functioning, cost-effective, and scalable confocal microscope platform, with potential applications in research labs, educational environments, and low-cost diagnostic tools.

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