

Abstract

Background:

The biomedical engineering teaching labs at the University of Wisconsin-Madison have two inverted fluorescent microscopes. Both of these inverted fluorescence microscopes are currently controlled using manual translational control knobs, which do not allow for automated imaging and automated stitching of images. The current commercially available options for motorized hardware for the stages of microscopes are too expensive. The overarching goal of this project is to design, program, and fabricate a lower cost motorized stage to be used for inverted fluorescent microscopes to allow for automated imaging and automated stitching that can be integrated with the Nikon Elements imaging software in the teaching labs.

Impact:

Integrating a motorized stage allows for a range of functions including time-lapse imaging, automated tracking, and image mosaic creation. A fabricated prototype was created to help stabilize the motors responsible for operating the microscope. The use of our prototype will allow automatic movement of the microscope, which once combined with the Nikon Elements software will enable automatic image stitching. This will save students and faculty valuable time and effort.

Problem Definition

- A motorized microscope stage integrated with Nikon software makes collecting imaging data easier and more time efficient, by allowing for automated imaging and stitching.
 - **This semester, it was key to create stabilizing devices to keep the motors balanced during experimentation**
- The Nikon Ti-U in the teaching lab comes equipped with TI-SR Rectangular Mechanical Stages [1] and the Olympus IX71 comes with IX-MVR Mechanical stages [2].
- Ideally, a motorized stage would be used because of its accuracy in movement and its capability for automated imaging.
- Nikon Elements Basic Research: Used for processing, measuring, and analyzing images [3].

Design Specifications

- Stage movement should be controlled by joystick or computer software.
- Stage movements should be within a resolution of **1-10 microns in x and y direction.**
- Structural support system must be put in place for the motor to allow it to frictionlessly move along the rail system while the microscope is being operate
- Should be able to be **easily attached and removed.**
- Should not inhibit the movement of the stage in any direction.
- Remain in budget of **\$100**

Previous Work

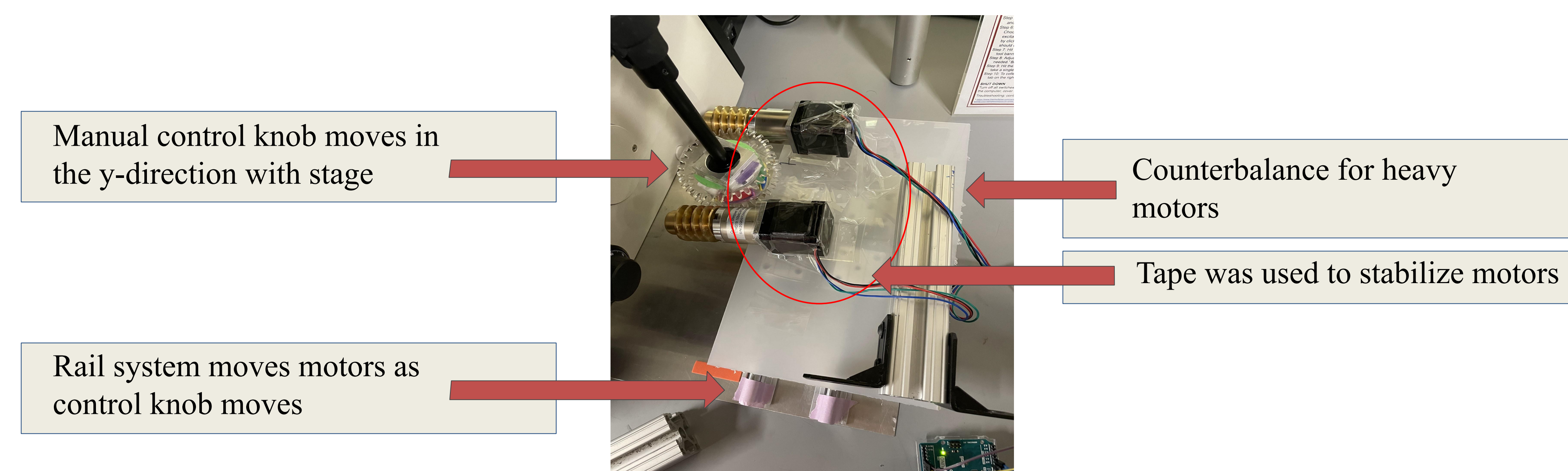


Figure 1. Top view of last year's final design. Stepper motors are connected to a rail system which can slide with the stage in the y-direction. Stepper motors are controlled with an Arduino Uno microcontroller and an Arduino program. The motors are being held down with tape and colored foam is used to give an appropriate amount of separation for the gears.

Final Stabilizer Designs

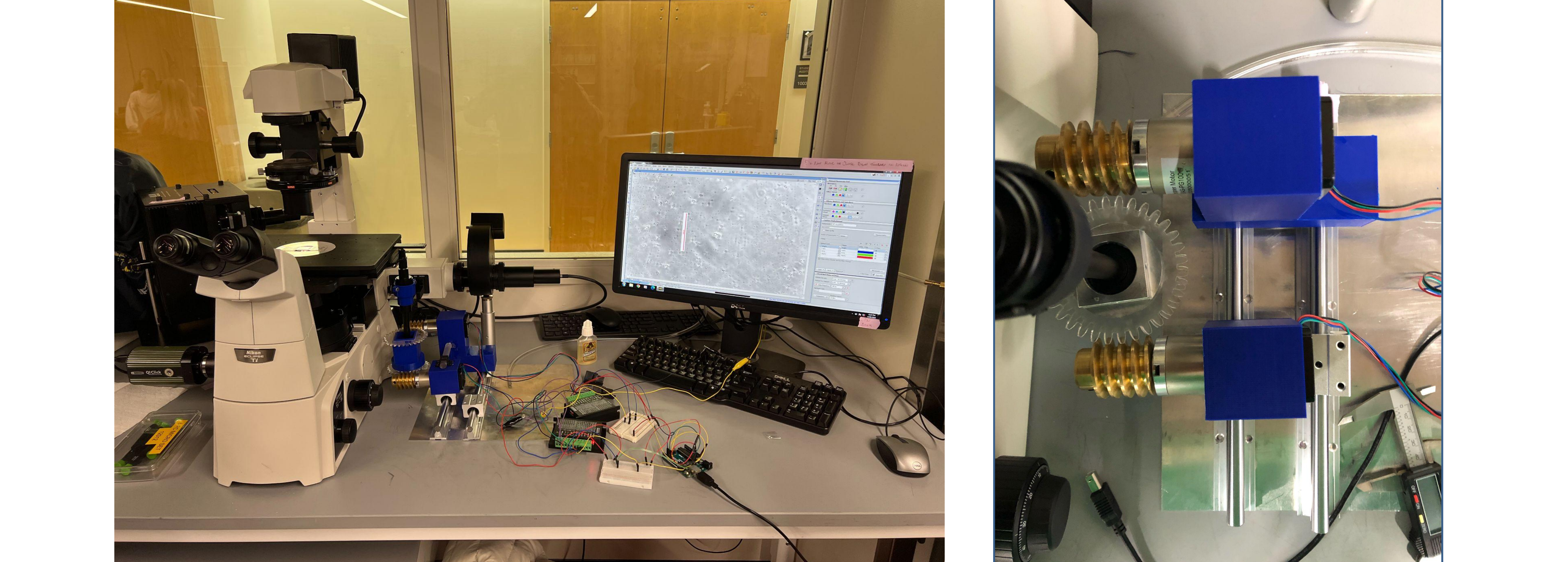
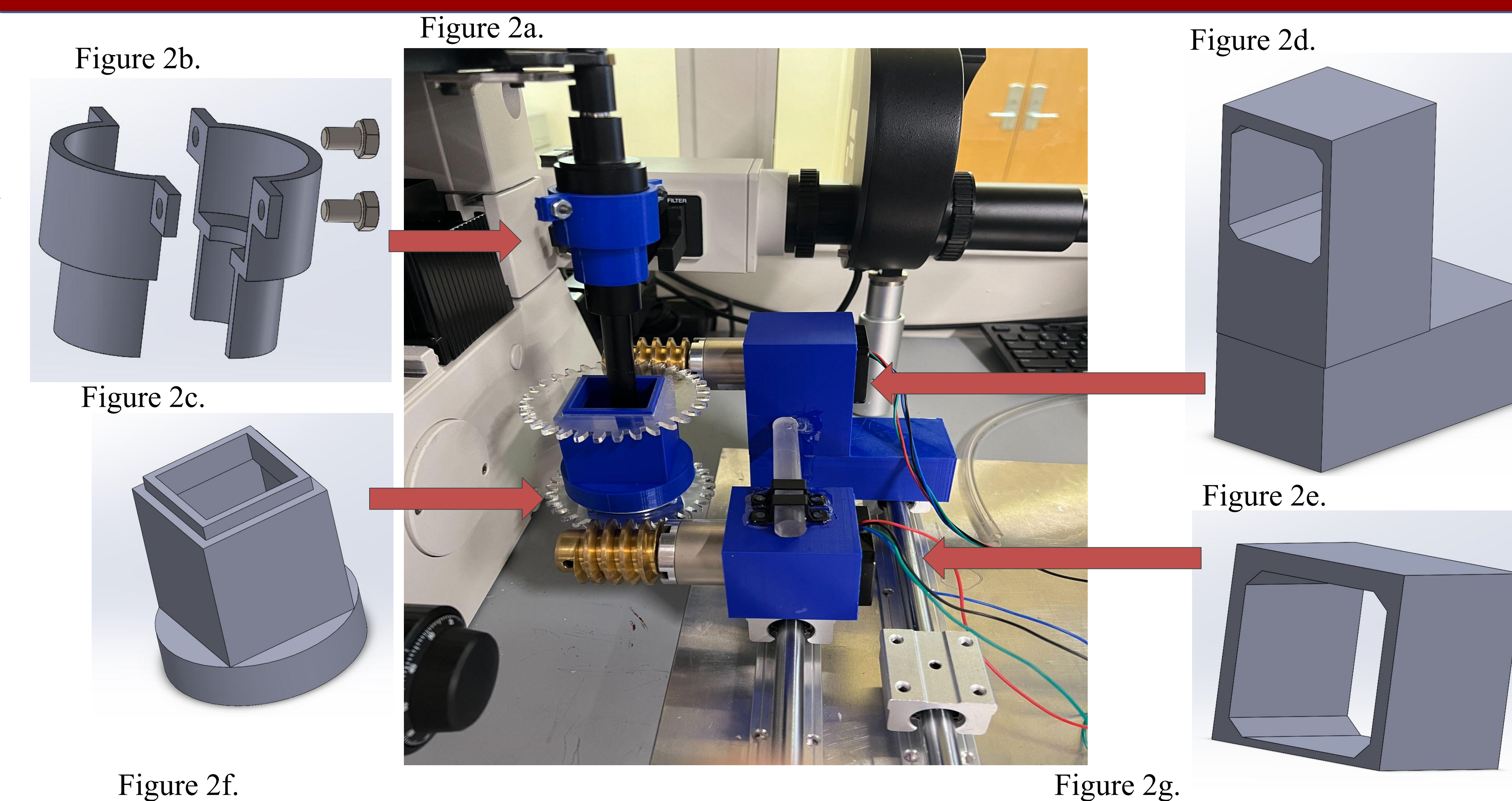


Figure 2. a) Front view of all the gears and the attachments stabilizing the motors and connected to the microscope. Four images showing the SOLIDWORKS designs of all the stabilizers. b) Stabilizer for manual control knob. c) Gear separator to allow each motor to attach to separate gears. d-e) Motor stabilizers. f) Image showing the entire set up with the microscope, attachments, arduino, and projected microscope image all together. g) Image of how the gears are attached to the motors with the stabilizers connected.

Testing Procedure

- Testing for Speed
 - Photograph the sample at position 1
 - Set the motor to a constant speed for 2.5 seconds
 - Photograph the sample at position 2
 - Import images into ImageJ, calculate the distance traveled of one 6 μ m dot
- Testing Distance Traveled Accuracy
 - Set motors to move 100 μ m
 - Measure distance actually traveled of 6 μ m dot using ImageJ
 - Compare predicted vs actual distance travelled

Testing Results

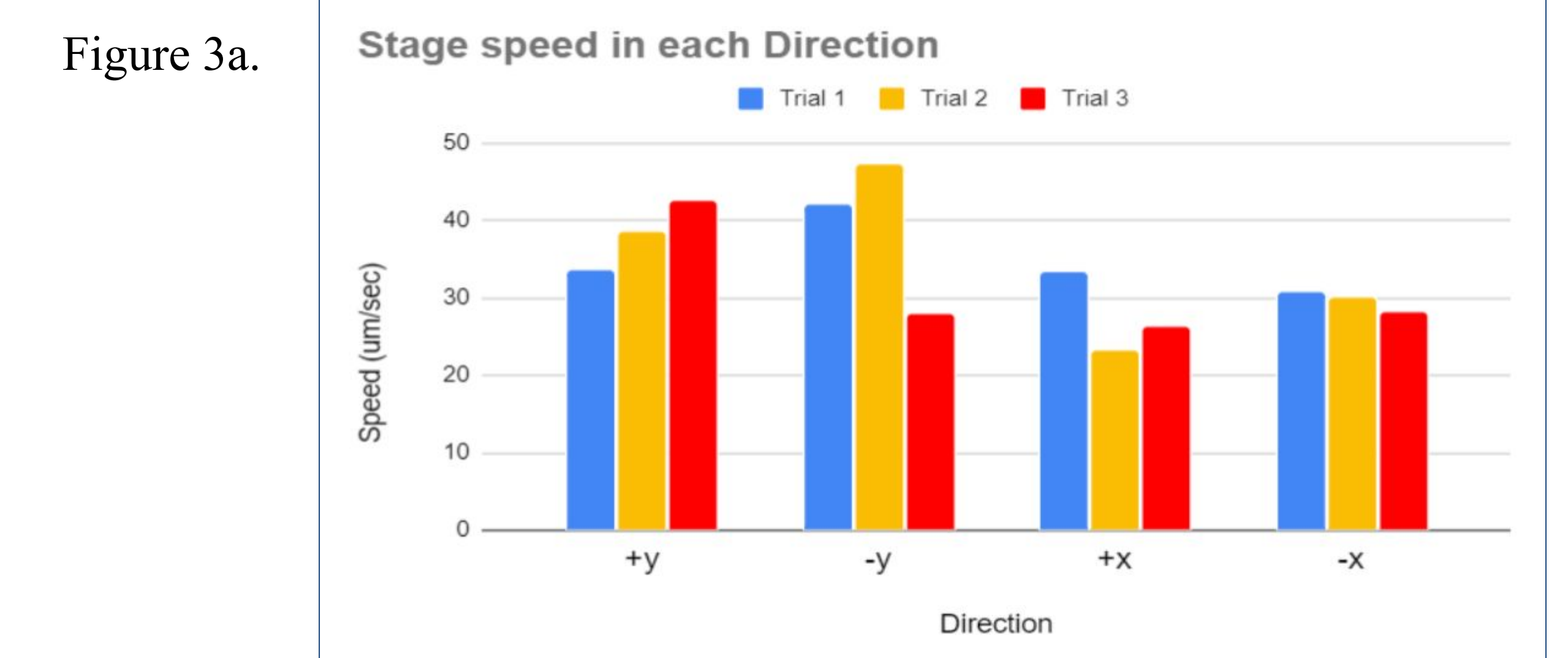


Figure 3b. Accuracy Data

Direction	Mean (μ m)	Standard Deviation (μ m)	Percent Error
+y	103.56	25.86	3.56%
-y	113.85	8.44	13.85%
+x	92.69	16.09	7.31%
-x	95.65	9.02	4.35%

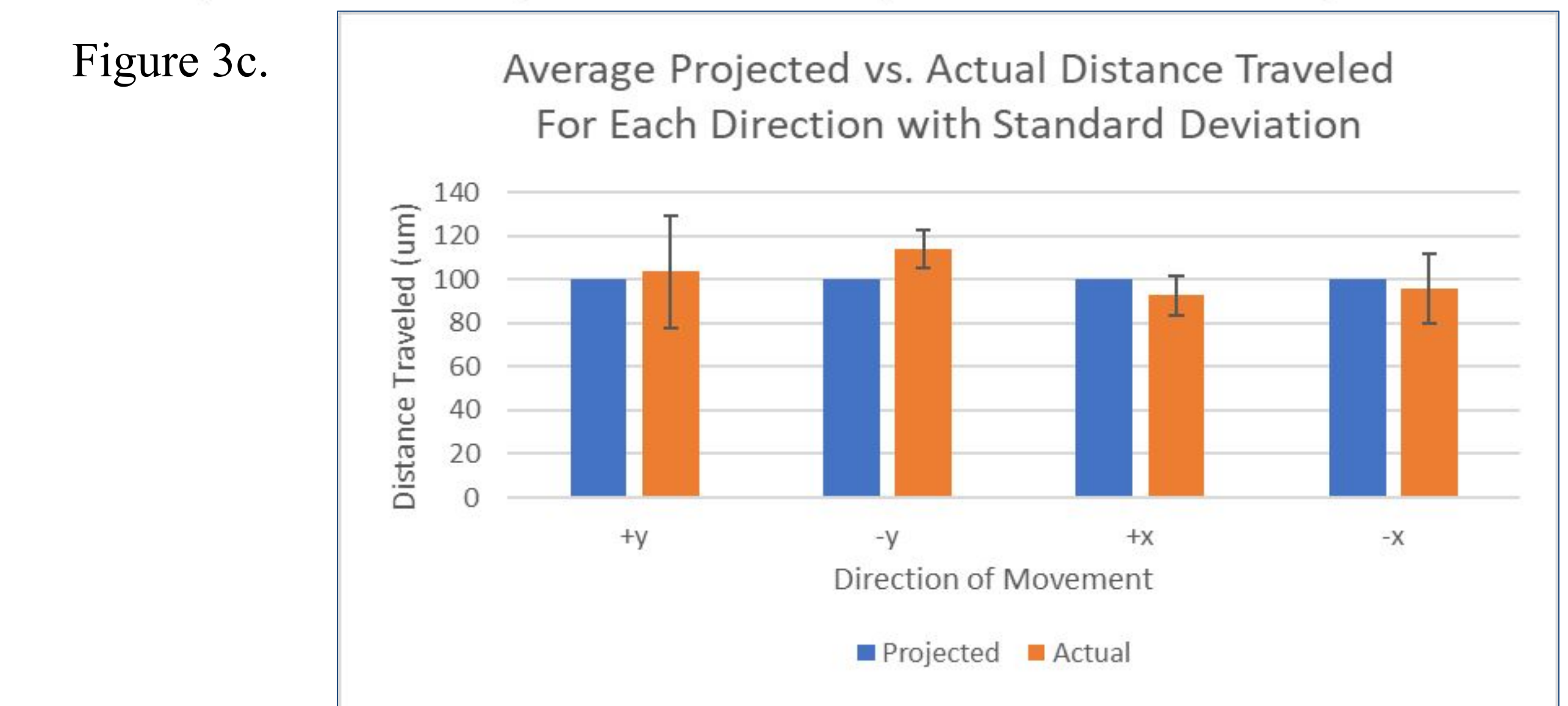


Figure 3. Data table highlights the results two different tests used to analyze the effectiveness of the stabilizing device. The first test (a) was done to find how fast our stage moved with our program, while the second test (b-c) used the average speed from the first test to find how consistently and accurately the stage moved (n=3) given a 100 μ m expected distance.

Future Project Development

- Increase Stepper Motor RPM
 - Improve circuits or code
 - Possibly get new motors
- Integrate motorized microscope stage with Nikon Elements Imaging Software
- Continue integrating the joystick

Conclusion

This semester's focus was stabilizing the motors and increasing the accuracy of the device. The stability due to the 3D printed attachments was greatly improved from the previous semester, and it helped with the overall function. The device produced sufficient results that were similar to the requirements specified in the PDS, however the consistency still needs work. With improvements in the consistency of motor movement, the team believes the device will be ready for integration with Nikon Elements imaging software.

References

- [1] "Nikon Inverted Microscope ECLIPSE Ti-U Ti-UB Ti-S Ti-S/L100 Instructions," *eliceirilab.org*. [Online]. Available: <https://eliceirilab.org/sites/default/files/2016-09/Nikon%20Eclipse%20Ti-U%20Manual.pdf>. [Accessed: 19-Oct-2021].
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- [3] "NIS-elements: NIS-elements basic research," *Nikon Instruments Inc.* [Online]. Available: <https://www.microscope.healthcare.nikon.com/products/software/nis-elements/nis-elements-basic-research>. [Accessed: 11-Oct-2021].