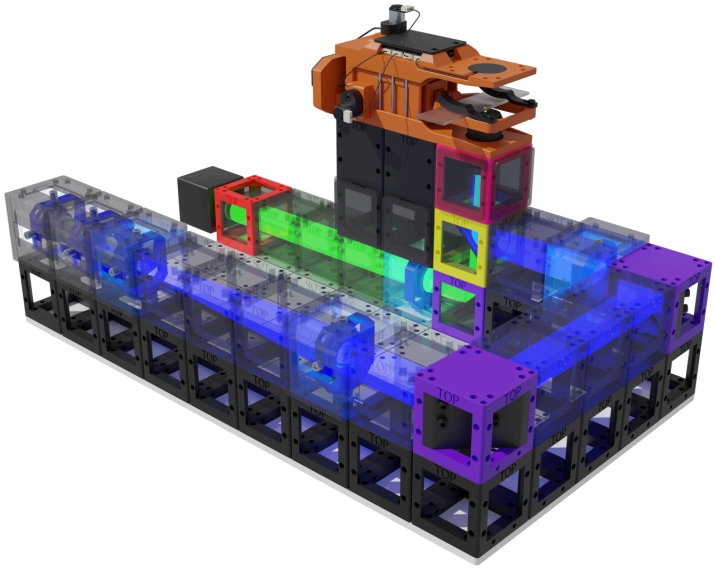
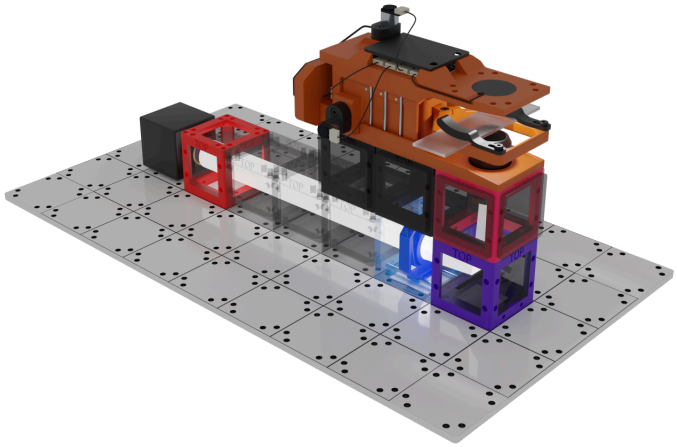
Protocube Functions and Definitions by Colton Hormann

Functions and Definitions

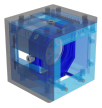
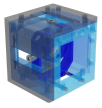
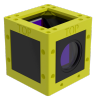
Fluorescence Microscope

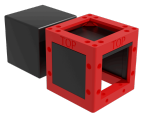
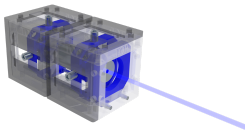
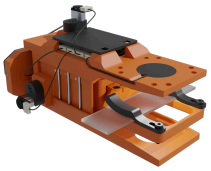
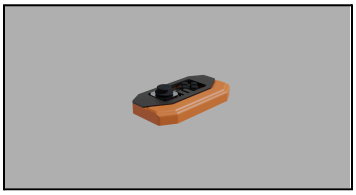
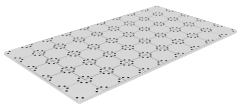


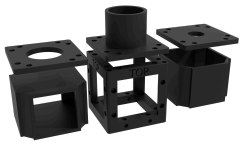
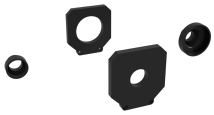
Transmitted Light Microscope



Cubes!

Image	Name	Description
	One Inch Lense Cube	A Light Blue Cube designed to hold a standard 1” optical lens. The lens is fixed in place by an M4 set screw and a press fit cap is included for extra security. The lens is positionally aligned with two M6 set screws and a manual rail adjustment.
	Half Inch Lense Cube	A Light Blue Cube designed to hold a standard 1/2” optical lens. The lens is fixed in place by an M4 set screw and a press fit cap for extra security. Positional alignment is set with two M6 set screws and a manual rail adjustment.
	Horizontal Mirror Cube	A Purple cube designed to fit a rectangular mirror. The mirror’s distance and alignment are determined by three brass thread inserts heat pressed in, along with an M3 Screw contacting magnets on the mirror plate.
	Vertical Mirror Cube	A Purple cube designed to fit a rectangular mirror. The mirror’s distance and alignment are determined by three brass thread inserts heat pressed in, along with an M3 Screw contacting magnets on the mirror plate.
	Dichroic Cube	A Yellow cube designed to hold a standard 1” Dichroic mirror. It is sized for SM1 to hold standard 1” filters via press fit.
	Basic Cube	A Black cube used for support and light occlusion. It prevents unnecessary noise and signal interruption and provides an easy solution to a safely enclosed light path.

	**Camera Cube**	**A Red cube designed to fit a standard size SM1 via a press fit and keep the light path enclosed.**
	**Objective Cube**	**A Pink cube designed to hold a standard microscope objective. It is placed at the appropriate distance on the z-axis for use with a stage.**
	**Light Source Cube**	**A White cube that holds the light source and is adjustable in the same way as the lenses, but with two mounts to align and tilt the light source. It also includes covers to be enclosed once aligned.**
	**XYZ-Stage**	**An xyz-Stage designed to use consumer electronics from Pimoroni and put them together into a functional, position control stage. Currently the encoders are unused, and other attachments for the stage are available.**
	**Stage Electronics**	**The various electronics used to control the XYZ Stage**
	**Gamepad**	**The Gamepad used to control the various functions of the XYZ Stage**
	**Baseplate**	**A laser-cut Base Plate that holds magnets arranged to help align and snap together the cubes.**

	**Accessories**	**An assortment of accessoriesfor the cubes that provide mounting points and light occlusion.**
	**Covers**	**A few different options for extra light occlusion to be inserted into already existing cubes, to save on 3D printing material and time.**
	**Cubes**	**An assortment of various cubes available for different applications with the microscope.**
	**Filters**	**A few adapters designed to hold either an SM1 or SM05 filter from Thorlabs**
	**Tools**	**The tools used in the assembly and alignment of the microscope.**

SAFE USE GUIDANCE - GENERAL

A Class 3R laser is low powered. It normally does not harm eyes during momentary exposure of less than ¼ second. This duration is within the aversion response, when a person turns away and/or blinks to avoid bright light.

Do not deliberately look or stare into the laser beam. Laser protective eyewear is normally unnecessary. A Class 3R laser is neither a skin nor materials burn hazard.

However, a Class 3R laser can be a distraction, glare or flash blindness hazard for pilots and drivers. **NEVER aim any laser towards an aircraft or a vehicle in motion.** This is unsafe and is illegal – you could be arrested and jailed.

ONLY ALLOW USE BY RESPONSIBLE PERSONS!!!

This is NOT a toy. Continuous adult supervision is required for children to safely use Class 3R lasers.

Laser Source	Power (With ND Filter)	Power (Without ND Filter)
Red (640nm)	13.9mW	109.2mW
Blue (405nm)	7.9mW	49.7mW
Green (532nm)	4.3mW	29.5mW

Please refer to the documentation provided by the manufacturer for additional warnings and preventive, protective equipment (PPE) requirements (e.g. laser safety goggles). Always consult your local Laser Safety Officer or Radiation Safety Officer and refer to your laboratory safety documentation for more information.
You can also consult your Laser Safety Standards ANSI Z136 in North America, SUVA 66049.D in Europe, and BS EN 60825-1 in the UK. Additionally, laser safety standards and regulations are covered by IEC norm 60825-1, and LED eye safety standards and regulations are covered by IEC norm 62471 in Europe.
Safety guideline: Hazardous, visible, or invisible radiation from lasers, lamps, and other light sources used for microscopy can cause permanent damage to the retina, skin burns, and fire. Always follow proper laser safety protocols for your equipment and situation.

Choking Hazard!

This product uses small neodymium magnets! Not for use with children under 3!!!

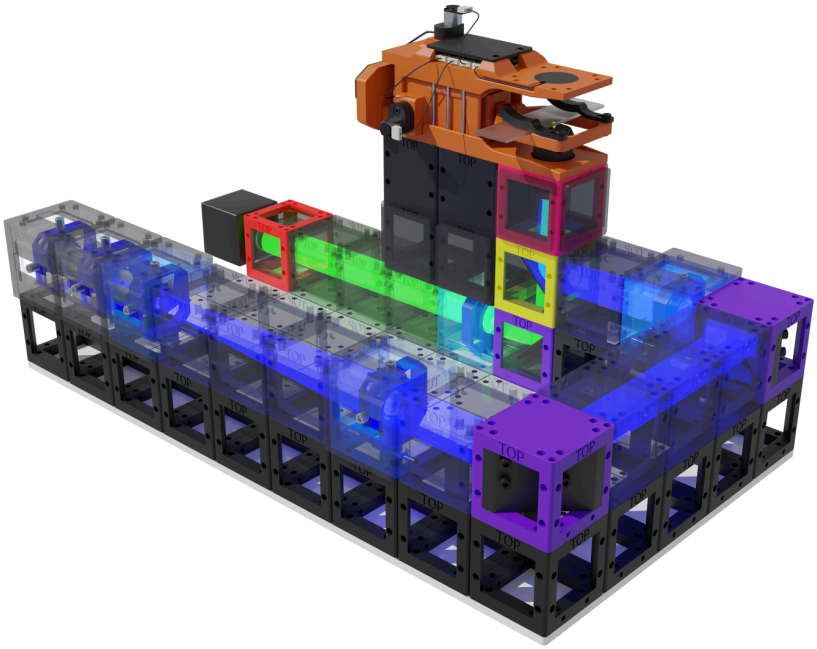
Reminders!

Never touch any lenses or mirrors directly with your hands or fingers!
Never put any part of your body in the beam path, it is dangerous!

Assembly Instructions

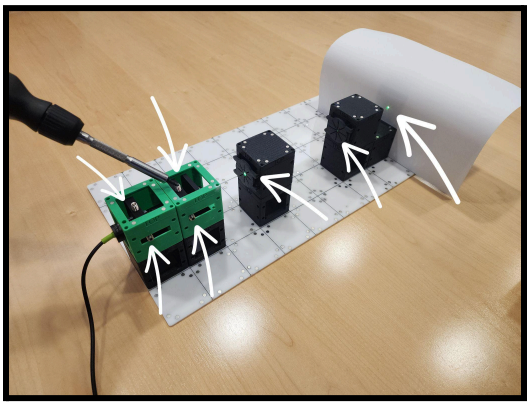
Minimum requirements:

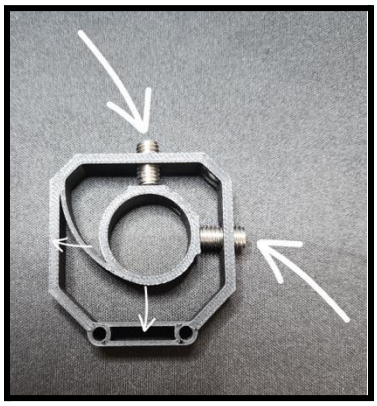
- 24 Closed Horizontal Cubes
- 1 Normal Cube
- 5 Open Outer Covers
- 2 Outer Side Covers
- 1 Open Outer Side Cover
- 2 Horizontal Mirrors
- 1 Vertical Mirror
- 1 Light Source Assembly
- 1 fitting Dichroic Cube
- 1 Half-Inch Cube f30
- 3 One-Inch Cubes (f200+)
- 2 Targets, one or more per type
- 1 Objective Cube
- 1 Camera Cube
- 1 XYZ Stage



Excitation Beam Path:

Laser Alignment

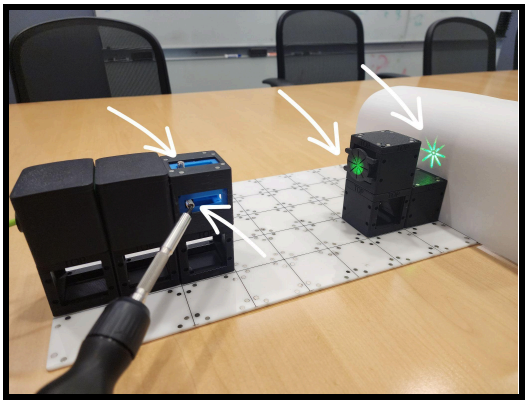


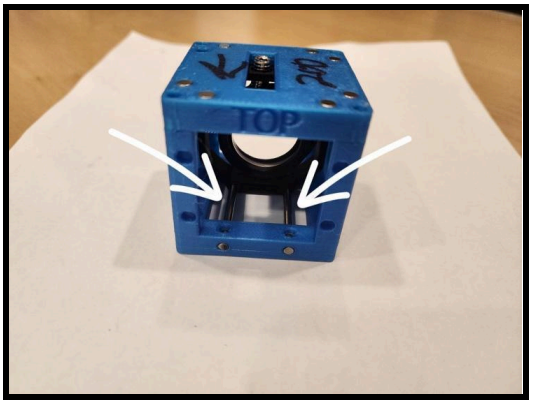


1. Start by placing a laser source on top of two of the black basic cubes, placing them on top of the magnetic base plate. Remove the laser sources covers and loosen the four set screws by unscrewing them a little bit
2. Place two targets in front of the laser like in the image below, making sure to also place them on top of the magnetic base plate at least three cubes apart from each other, and one cube away from the laser.
3. Set up a flat target such as paper on the other end of the targets, to see how the laser needs to be adjusted.
   The set screws push the inner mount horizontally and vertically to allow for precise control of movement.
   Using this, you can make small adjustments, centering and tilting the laser to shine through the center of both targets, swapping them around to double check that it is aligned correctly like above.

Recommendations

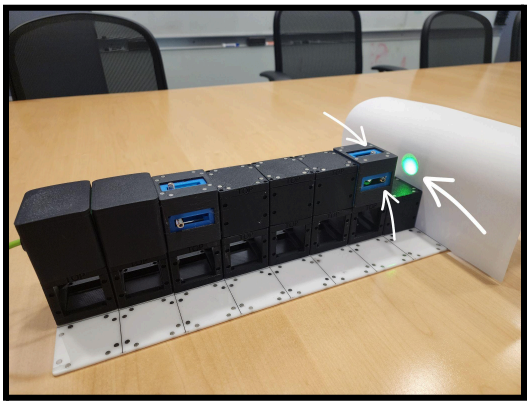
Tap the laser source cube on the top with your screwdriver after every adjustment to make sure the internal mount is seated correctly.
Slightly tilt the cubes, as the magnets don’t set exactly into the same spot and can be adjusted by wiggling the cube a tiny bit.

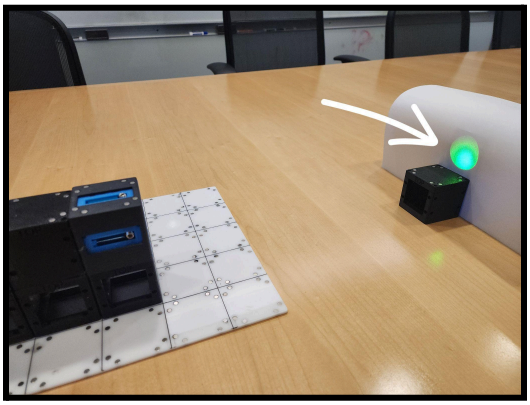




Beam Collimation

1. Start by placing an f30 lens directly in the laser path, loosening the set screws by unscrewing them a little bit.
   Slide the lens back on its rails to be as close to the laser as possible, this is easier done from the bottom of the cube, while picked up.
   Use the set screws to center it as close as possible to the beam path like in the image above. Placing a cube behind the paper with light behind it to act as a target to center to.
2. Next place an f200 lens about 200mm away and center it by using the same method as before with the f30 lens.
3. Slide the f200 lens back and forth along its rails in the cube until it is focusing the beam path to be aligned and parallel, or collimated, and the beam coming out doesn't shrink or grow when moving a flat target such as paper farther away.
   Note: You may have to move the f30 lens back and forth as well to get the best alignment.



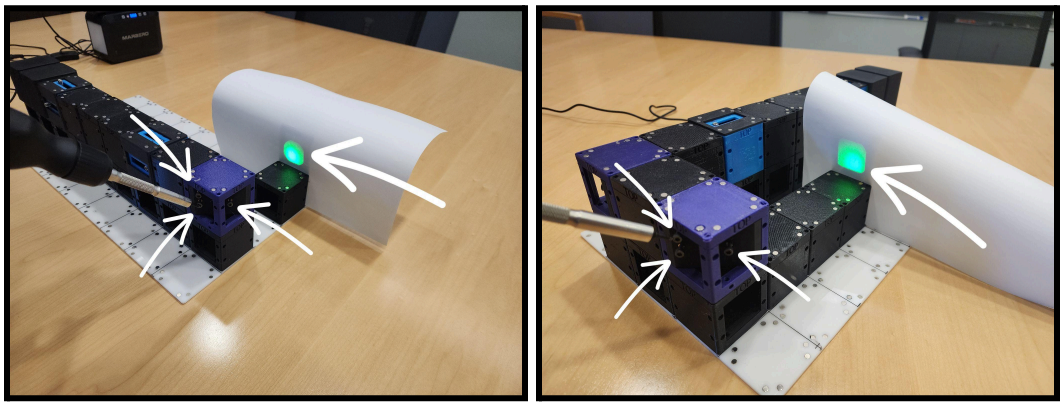


Mirror Alignment

1. If necessary, direct the beam with the horizontal mirrors, making sure it stays in the same plane by adjusting the screws on the back to tilt the mirror and adjust its distance like in the images below.

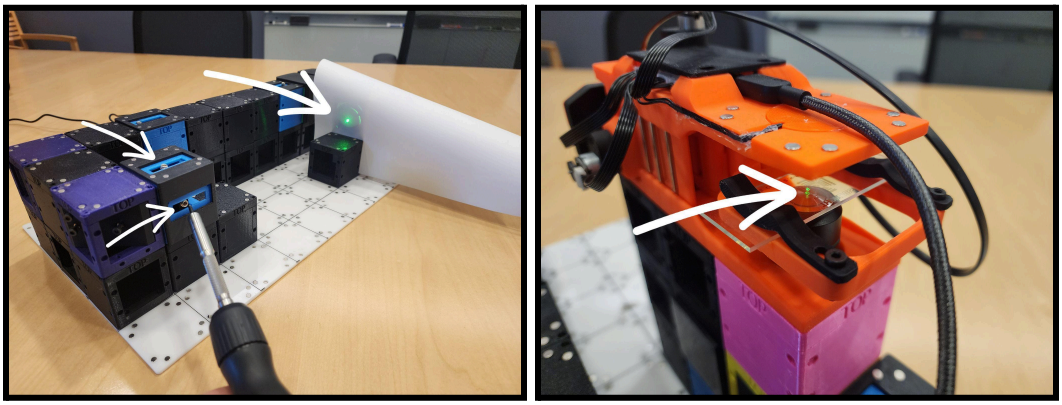
Recommendation

Use the same paper method as that used in the beam collimation steps to ensure that the beam path stays straight such as in Figure XXX.



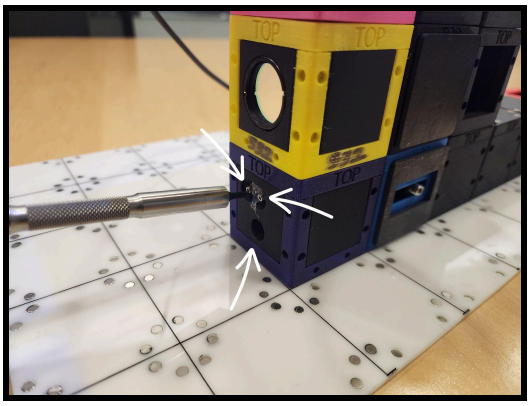
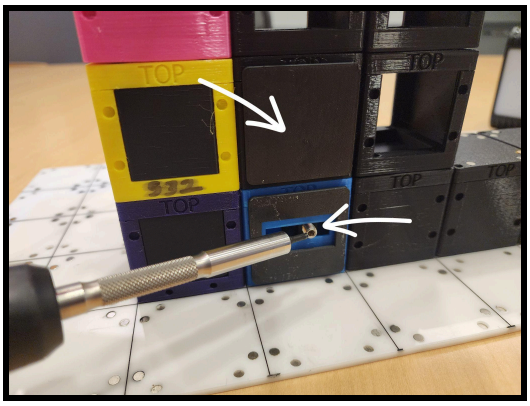
Objective Focusing

1. Direct the beam into an f200 lens, making sure that it is centered with the set screws.
2. Focus the beam into the objective like a cone using the f200 lens, sliding it back and forth so that the beam path comes out on top of the infinity corrected objective parallel.
3. Direct the beam into a correct set of filters and a dichroic mirror in the dichroic cube, and up into the objective.

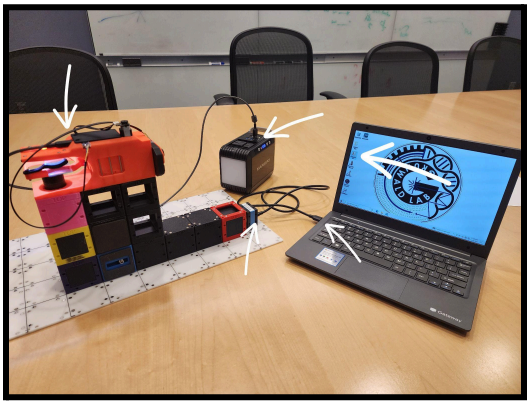
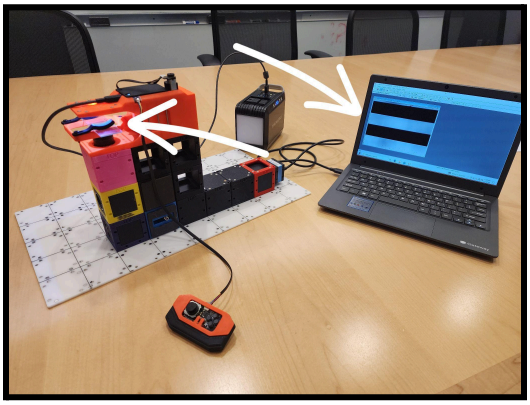


Emission Beam Path:

1. Place the xyz-stage on top of the objective.
2. Let the beam pass through the filter cube and onto a vertical mirror, adjusting the mirror via its screws to keep the beam parallel and direct it forward.
3. Place a f200 lens in front of the beam, making sure it's centered using the set screws.

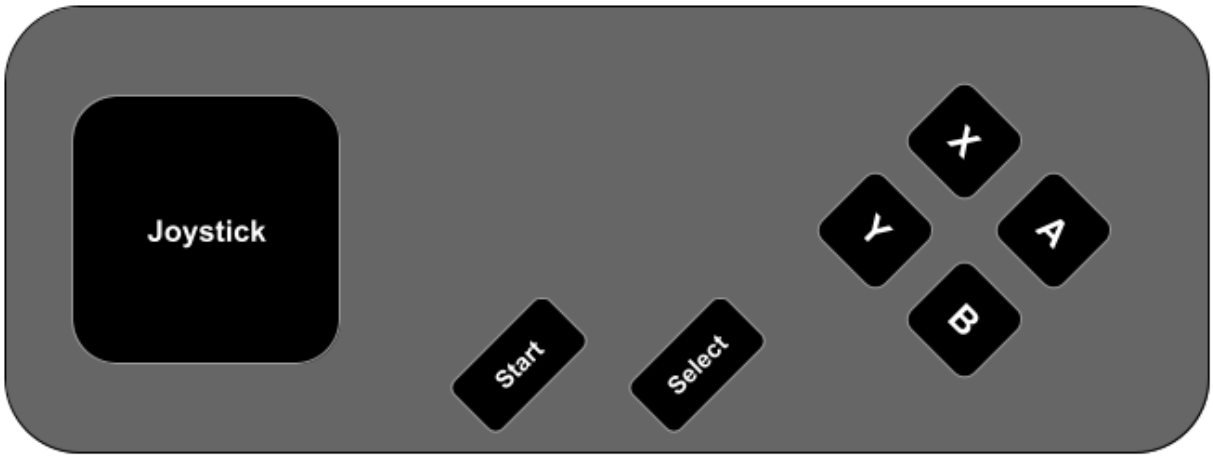



4. Place the camera so the camera sensor is about 200mm from the f200 lens.
5. Plug the xyz stage into a usb power outlet, not a computer.
6. Plug the camera into a computer, and start IC Capture

7. Make sure the path to the camera is enclosed, as any external lighting can overexpose your image.
8. Focus on your sample and have fun!

Gamepad



Joystick

Used to move the X, Y, and Z Axis

(It does not contribute to speed control, push it all the way in one direction at a time to move the stage)

Start

Switch between X/Y, and Z mode on the Joystick

(Hold for one second, Z Mode only uses the vertical axis of the joystick)

Select

Home the Stage.

(You shouldn't need to do this but just in case, hold for one second)

Y	Motor Speed 27.5%
X	Motor Speed 62.5%
A	Motor Speed 100%
B	Toggle LED On/Off