

# FlyBox Assembly Instructions

Brandeis's Rosbash Lab does groundbreaking research on circadian rhythms and neuroscience. This research relies upon data collected by the FlyBox, a box designed to house 96 flies in a stable and replicable environment with means for providing video footage of the flies. The improved FlyBox provides a better working experience for researchers at Brandeis and other labs around the world.



Please consult the Bill of Materials document at

<https://github.com/Rosbash-Lab-FlyBox/FlyBox/tree/main/Directions%20and%20Bill%20of%20Materials> to ensure that you have all required parts.

Have the following tools and supplies on hand:

- Electric screwdriver with M2, M3, M4, small flathead bits
- Vise-grip or slip joint pliers
- Wire cutters
- Wire strippers
- Paper towels
- Tape
- Sandpaper (about 40 grit) or a small hand file
- Soldering iron and supplies
- Boxcutter knife
- Allen wrench

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# Laser Cutting Instructions

The FlyBox laser-cut files can be found at

<https://github.com/Rosbash-Lab-FlyBox/FlyBox/tree/main/fabrication-files/laser-cut-files>.

The FlyBox frame is assembled from 17 laser-cut panels, of which 15 are cut from opaque  $\frac{1}{4}$ " black acrylic and 2 are cut from translucent  $\frac{1}{8}$ " diffuser acrylic. There are 7 cutsheets, each needing a 12"x24" panel. In addition, a lightproofing curtain is cut from an 18"x18" felt sheet and a 1" radius IR filter is cut from IR filtering acrylic.

Material (cutsheets)	Part name (Cutsheet #)
Opaque $\frac{1}{4}$ " Black Acrylic (Six 12"x24" cutsheets)	<input type="checkbox"/> Bottom Panel (1) <input type="checkbox"/> Back Wall (1) <input type="checkbox"/> Outer Top (2) <input type="checkbox"/> Inner Top (2) <input type="checkbox"/> Top Hatch (2) <input type="checkbox"/> Electronics Panel (2) <input type="checkbox"/> Outer Front (3) <input type="checkbox"/> Inner Front (3) <input type="checkbox"/> Front Door (3) <input type="checkbox"/> IR Alignment Panel (4) <input type="checkbox"/> Fly Tray Holder (4) <input type="checkbox"/> Lower Floor (4) <input type="checkbox"/> Upper Floor (5) <input type="checkbox"/> Left Wall (5) <input type="checkbox"/> Right Wall (6) <input type="checkbox"/> Curtain Clamp (6)
Translucent $\frac{1}{8}$ " White Acrylic (One 12"x24" cutsheet)	<input type="checkbox"/> LED Diffuser Panel (7) <input type="checkbox"/> IR Diffuser Panel (7)
Opaque Black Felt (One 18"x18" cutsheet)	<input type="checkbox"/> Lightproofing curtain (8)
IR Filtering Acrylic (One 12"x12" cutsheet)	<input type="checkbox"/> Camera IR filter

## Suggested Laser Settings

The provided cutting files are color-mapped: black lines (#000000) should be completely cut through the material, blue lines (#0000ff) should be lightly vectored and rastered to engrave the part, and red lines (#ff0000) should not be cut at all. The suggested settings listed below apply to an Epilog Helix 60-watt laser cutter and should be verified before being used on other laser cutters.

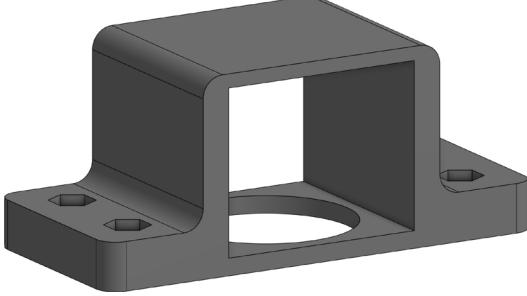
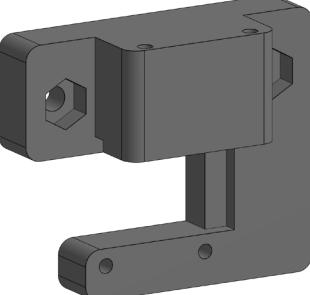
There is a small (1" x 12") test cut file included with this file set that can be used to test that your laser settings are correct. To avoid wasting material, on the black acrylic this should be cut out from cutsheets 3, 4, or 6 that will have excess material on the right side of the sheet.

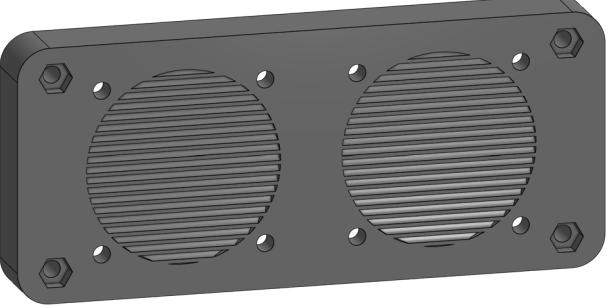
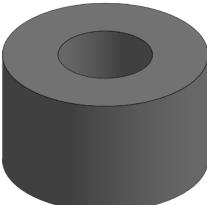
Opaque 1/4" Black Acrylic		Translucent 1/8" White Acrylic		Opaque Black Felt	
	Black lines		Black lines		Black lines
Raster	Speed: 100% Power: 30%	Speed: 100% Power: 30%	Raster	Speed: 100% Power: 30%	Speed: 100% Power: 30%
Vector	Speed: 9% Power: 100% Freq: 5000	Speed: 100% Power: 30% Freq: 5000	Vector	Speed: 20% Power: 100% Freq: 5000	Speed: 100% Power: 30% Freq: 5000

## 3D Printing Instructions

The FlyBox also has a few 3D printed components. 3D-printable STL files are available at  
<https://github.com/Rosbash-Lab-FlyBox/FlyBox/tree/main/fabrication-files/3d-print-files>

These are the parts that need to be printed:

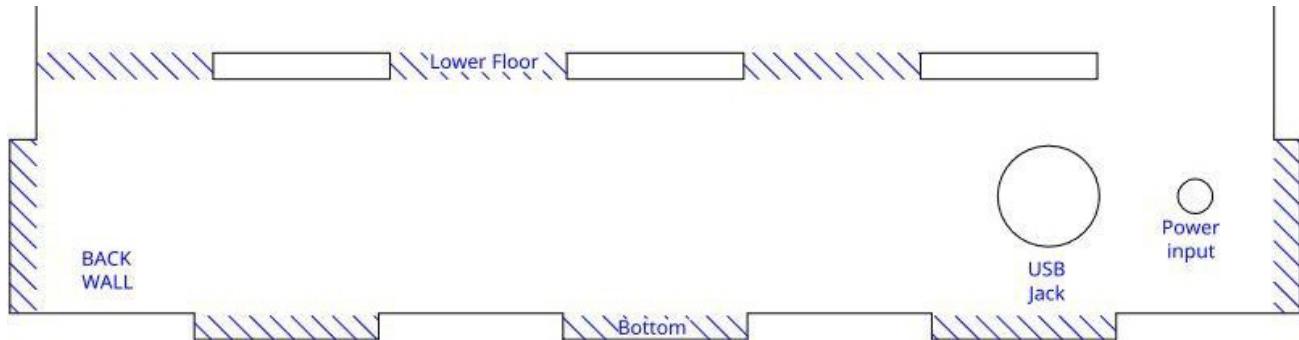
Part Name	Qty	Image	Print Settings
Camera Mount	1		Print with side facing <u>away from</u> camera face down  No supports necessary
SD Card & Rotary Encoder Mount	1		Print with side facing <u>away from</u> camera face down  No supports necessary except possibly in the channel for rotary encoder. Either print with supports or verify the channel is clean after printing.

Fan Baffle	2		Print with side facing camera face down Print with supports
Spacer	14		Print as shown No supports necessary

# FlyBox Frame Assembly Instructions

The laser-cut parts of the FlyBox are labeled with their names and the names of connecting parts. On most parts, the engraved labels face inward to the center of the box, such that they will be hidden when assembly is complete. The name of the part is on the main body of the part in ALL CAPS; lowercase letters indicate where connecting parts attach.

For example, below is a portion of the **Back Wall**, as is labeled at left. The finger joints of the **Lower Floor** and **Bottom** panels fit to the slots and dashed tabs. The **USB Jack** and **Power Input** are attached in the holes at the right.



## Hardware Attachment

Generally, when attaching hardware, the screw heads should be on the outer, unengraved side of panels and the nuts should be on the inside. This will make it easier to replace parts if any component breaks.

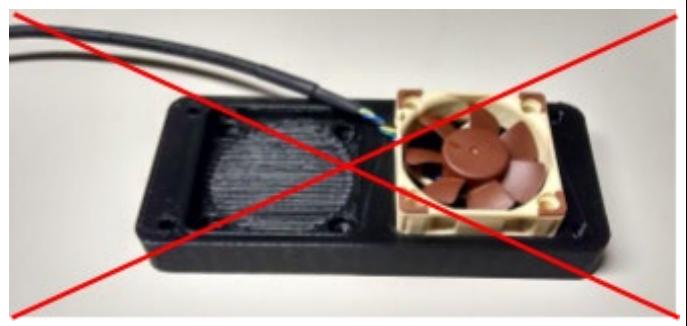
### Right and Left Walls

- 1 Take the four **Fans** and two **Fan Baffles**. On each **Fan Baffle**, attach two **Fans** to the flat faces of the baffle with eight M3x20 screws and M3 nuts (sixteen total).

Fans on each baffle should face the same direction, so they push air in one direction. The fans attached to one baffle should point in the opposite direction to the fans on the other baffle, so that one baffle is pushing air in and one is pulling air out.

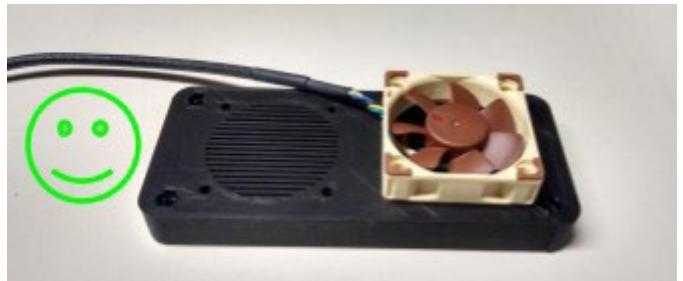
Rotate the fans so the cables on each baffle are pointing the same direction. This will make wiring easier later.

**DS:** It helps to hold 1 nut in place while tightening the screw with the other hand



*Incorrect attachment: Fan is placed inside square recess.*

DS: Now is a good time to peel off the paper coating on all the acrylic pieces. Using a box cutter blade (or something sharp) helps.



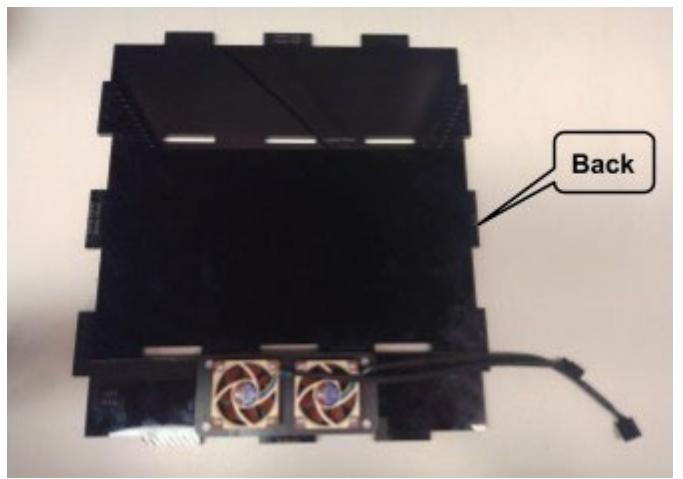
*Correct attachment: Fan is on flat face.*



*Nuts set into recesses on underside*

- 2 Using eight M3x20 screws and M3 nuts, attach one **Fan Baffle Assembly** to the engraved side of the **Right Wall**, and one to the **Left Wall**.

The cables of the fans should point toward the **Back Wall**, as marked on the engraved tabs.



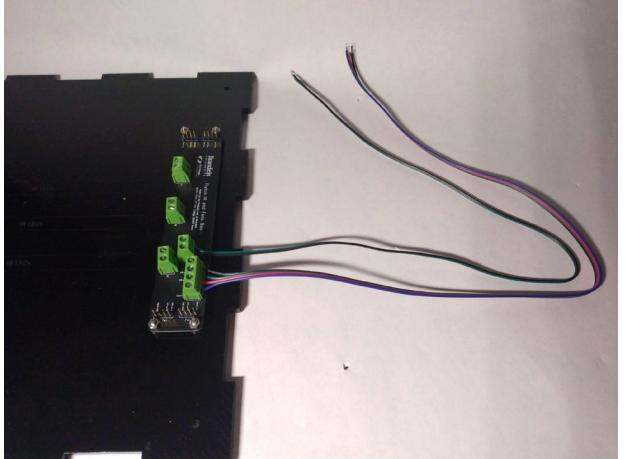
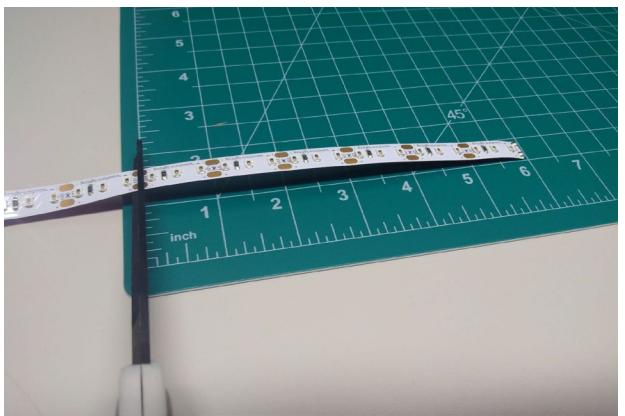
### Bottom Panel (IR and Fans)

- 1 Using four M3x16 screws and M3 nuts, and four **Spacers** attach the **IR and Fans Board** to the **Bottom** in the orientation shown by the engraving

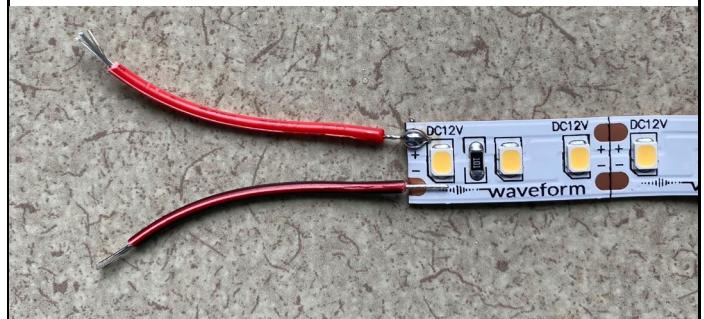
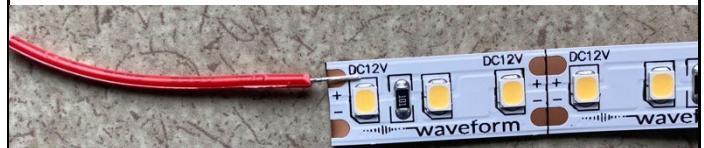
DS: It can be tricky tightening the nuts and screws - you'll have to carefully push the spacers under the board as you tighten

EK: It's easier to attach the bolts and spacers if you put each one on loosely first before tightening them



2	<p>Cut about 24 inches of 4 strand wire and 24 inches of 2 strand wire. Connect the 4-strand wire to the terminals labeled <b>Fans from above</b> and the 2-strand wire to the terminals labeled <b>IR from above</b>. Leave the other ends of the wire loose, they will be routed up the corner channel to the main board later.</p> <p><b>GB:</b> keep note of which wire connects with which screw terminal – this will be needed afterwards</p> <p><b>EK:</b> Use different colors of wire for each of these – it will make it easier to wire correctly later</p>	
3	<p>Cut three 6-inch sections of <b>IR LED strip</b>. Cut on the indicated cut lines with contacts.</p> <p><b>DS:</b> when cutting the pieces, make sure you cut evenly down the middle of the copper areas as you'll need space for soldering later</p>	
4	<p>Carefully solder 1.5" wire onto LED strips with the following instructions:</p> <p><b>EK:</b> cut two sets of wires a little longer to accommodate spacing them further apart for a 2-tray setup</p> <ol style="list-style-type: none"> <li>1. Cut 1.5" of red and black electrical wire and remove the myelin sheath on both ends</li> <li>2. Twist the ends of the wire so it is condensed and not hairy</li> <li>3. Flatten the LED strip horizontally with the (+) metal end away from you and the (-) metal end towards you</li> </ol> <p><b>EK:</b> the strips should be oriented so that the side facing away from you that you will be soldering onto has the (-) end on the left and the (+) end on the right</p> <ol style="list-style-type: none"> <li>4. <b>Orientation matters!</b> If you incorrectly solder the wrong ends, you'll have to solder again. Make sure the copper (+) and (-) ends of the LED strip you solder match up with the (+) and (-) inputs of the screw terminals on the board!!</li> </ol>	

- Do one wire at a time. Place the red exposed wire end on top of the copper (+) end of the LED strip as shown in the image and solder
- A helpful tip in soldering this way is to hold the soldering gun tip and wire closely above and quickly melt the wire to create a droplet, then gently push down the droplet, engulfing the wire and contacting the copper (+) end of the LED strip
- Pull the wire to see if it falls out. If it falls out, simply re solder.
- Repeat for the copper (-) end of the LED strip and test to make sure it does not fall out

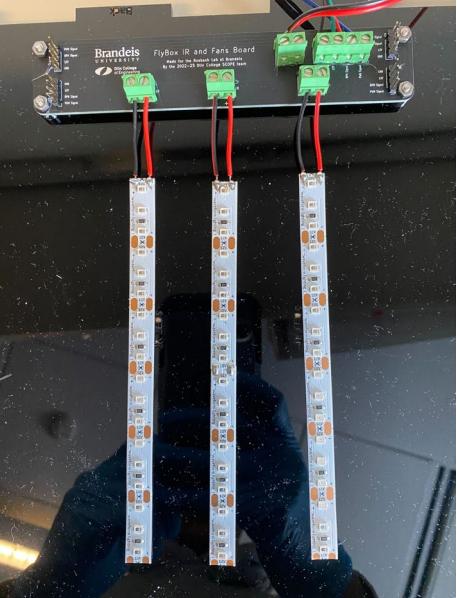
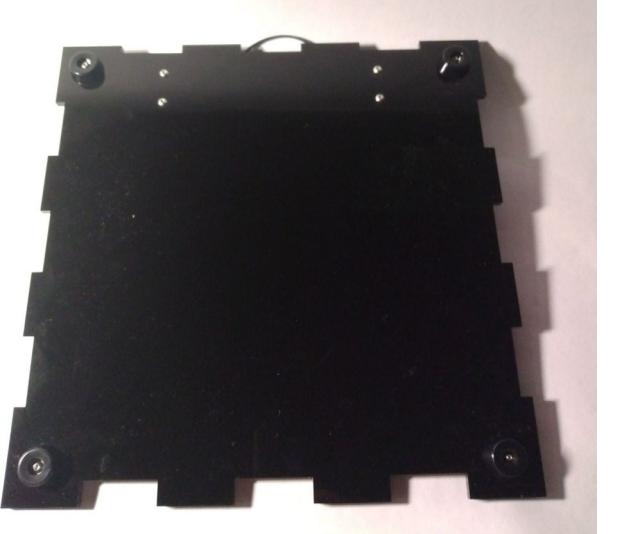


# STOP

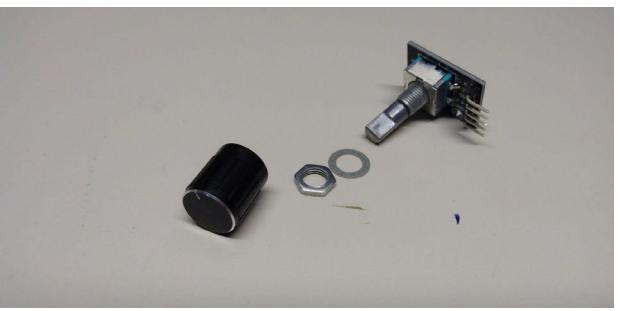
**Check to make sure the orientation is correct.  
Hover the LED strip next to the screw terminal  
and see that the (+) and (-) ends match up**

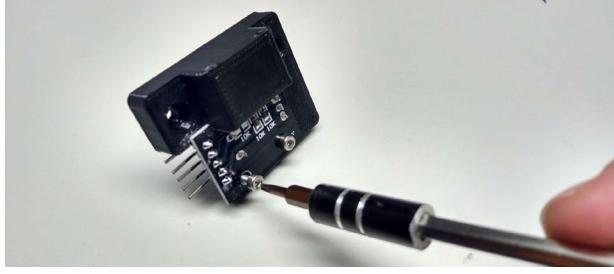
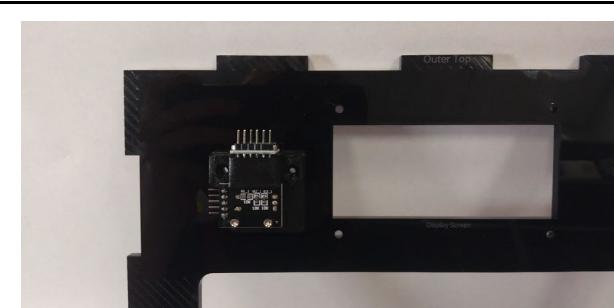
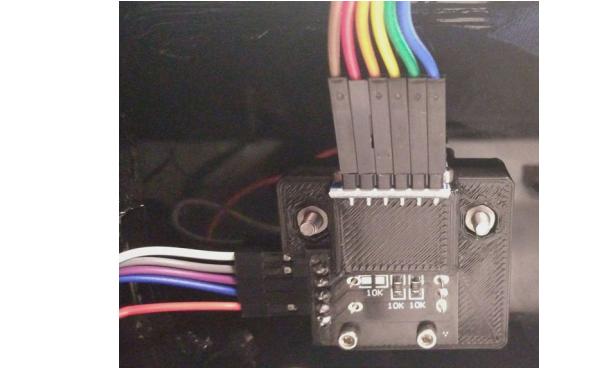
Because this is IR light, we cannot test it until after the box is built.

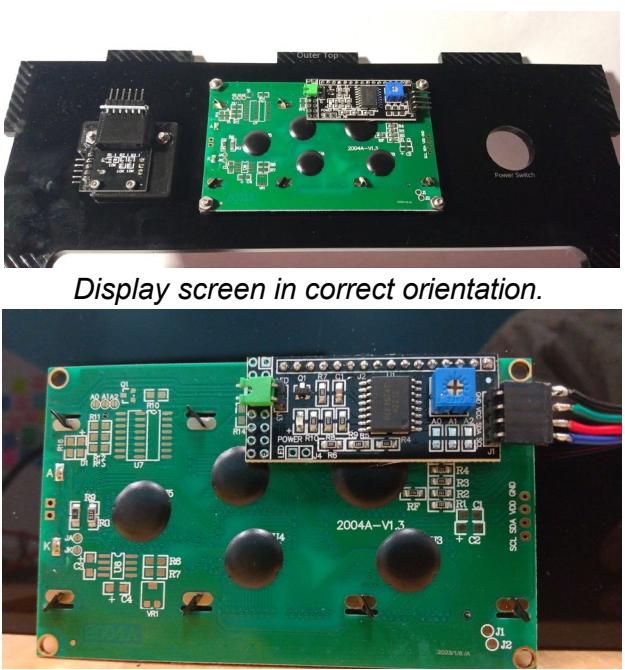
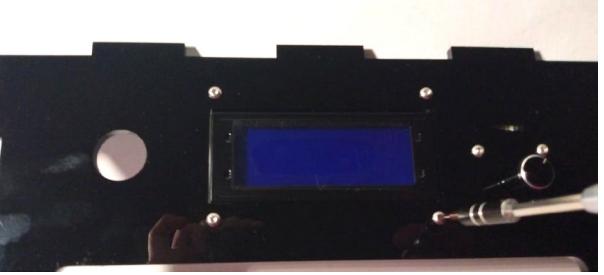
DS + GB steps 1 - 8

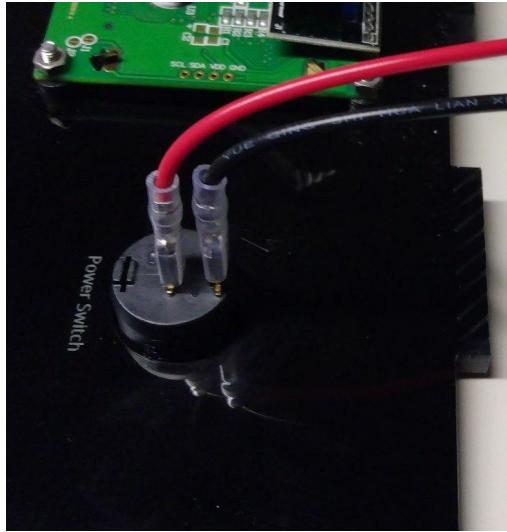
6	<p>Remove the paper backing and apply the three 6" <b>IR Light Strips</b> in the marked rectangular spaces on the <b>Bottom</b>.</p> <p>Connect the wire end of the LED strip connectors to the corresponding screw terminals on the IR Fans Board.</p> <p><b>EK:</b> place the outer two strips further apart than is marked on the board and shown in the picture. This is to accommodate a 2-tray setup</p>	
7	<p>Attach the 4 <b>Feet</b> to the unengraved side of the <b>Bottom</b> using four M3x16 screws and M3 nuts</p> <p><b>DS:</b> The wider part of the <b>Feet</b> should contact the board. The slightly smaller part will contact the ground. The nuts should be tightened being inside the box.</p>	

## Front Wall

1	<p>If the <b>Rotary Encoder</b> came with a knob attached, remove it by gently pulling it until detaches. Set aside the knob and any hardware</p>	
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2	<p>If the <b>SD Card &amp; Rotary Mount</b> was printed with supports, make sure the small channel on the backside is completely clear of support material.</p> <p>Slide the square, silver base of the <b>Rotary Encoder</b> into the large slot on the <b>SD Card &amp; Rotary Mount</b>, with the stem of the encoder extending through the flat face of the mount. It should fit into the slot snugly. Make sure the <b>Rotary Encoder</b> is pressed into the slot as far as it can go. Flip over the assembly and attach using 2 M2x6 screws.</p> <p><b>DS:</b> You may need to apply more pressure than expected. This is intentionally a very tight fit.</p>			
3	<p>Using two M2x6 screws, attach the <b>SD Card Reader</b> to the top square bump out of the <b>SD Card &amp; Rotary Encoder Mount</b>. It should be positioned such that the board sits flat, and the SD card slot is pointing in the same direction as the rotary knob.</p>			
4	<p>Attach the <b>SD Card &amp; Rotary Encoder Mount</b> to the engraved side of the <b>Outer Front</b> using two M3x16 screws and M#3 nuts. The stem of the encoder should protrude through the unengraved face.</p> <p>Replace the knob of the <b>Rotary Encoder</b>.</p> <p>Attach a five-strand female-to-male jumper cable to the <b>Rotary Knob</b> pins and a six strand female-to-male jumper cable to the <b>SD Card Reader</b> pins.</p> <p><b>EK:</b> once the rotary encoder is attached to the front panel, you can put the washers and knob back on</p>			

		
5	<p>Attach the <b>Display Screen</b> to the <b>Outer Front</b> using four M3x16 screws and M3 nuts and four <b>Spacers</b>. The spacers are placed between the LCD screen and the outer front panel. They ensure that the display screen is flush to the front. The pins should point toward the space for the <b>Power Switch</b>.</p> <p>Attach a 4-strand male-to-female jumper cable to the pins.</p>	 <p><i>Display screen in correct orientation.</i></p>
6	<p>Press the <b>Power Switch</b> through from the unengraved face of the <b>Outer Front</b>.</p> <p>Connect the included quick connect wires to the <b>Power Switch</b> by pressing them into the tabs. It does not matter which wire is connected to which tab.</p>	



### Removing the Camera IR Filter

Remove the **Camera** IR filter and base by carefully following this video:

▶ [Removing IR filter from Logitech C920](#)

Important notes:

- If you are capable of soldering small components, you may choose to desolder and later resolder the two leads connected to the lens casing
  - This avoids breaking two of the pins on the lens casing (as described at 13:06)
- Disregard 18:08-19:00, this step is unnecessary
- Do not replace the webcam's base, as shown at 19:00
  - The base does not fit into the FlyBox

### LED Diffuser Panel

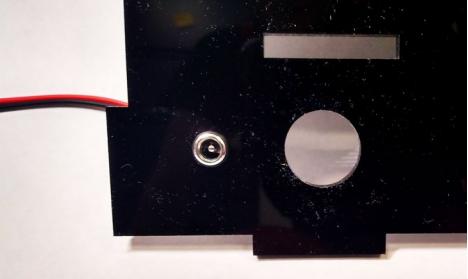
1	<p>On the <b>LED Diffuser Panel</b>, attach the <b>Camera Mount</b> using four M3x12 screws and M3 nuts.</p>	
2	<p>Place the <b>IR Filter</b> into the round space in the <b>Camera Mount</b>. Hold the filter by the edges to avoid smudges that could interfere with the camera's view.</p> <p><b>DS:</b> Not to be confused – this filter will filter everything else out except for IR light. The filter removed from the camera filters out IR light but takes everything else in.</p>	

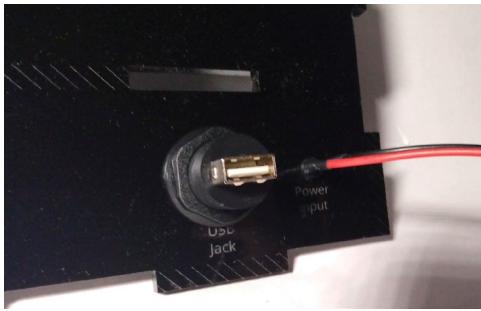
3	<p>Slot the <b>Camera</b> into the <b>Camera Mount</b>, such that the lens is pointing through the hole in the diffuser panel.</p> <p><b>DS:</b> Sometimes putting the camera into the mount can be tight – it should work.</p>	
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### Upper Floor

1	<p>Add four M3x35 standoffs <u>extending upward from the engraved side</u> of the <b>Upper Floor</b> using four M3x12 screws</p>	
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### Back Wall

1	<p>On the <b>Back Wall</b>, attach the <b>Power Input</b> by pressing it in from the unengraved side.</p>	
2	<p>Unscrew the collar from the <b>USB Jack</b>, press it through from the unengraved side, and screw the collar back on to hold it in place.</p>	

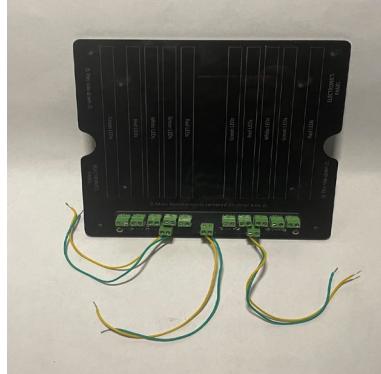


### Bottom Side of Electronics Panel

- 1 On the engraved side of the **Electronics Panel**, attach the **RGW Board** in the orientation shown by the engraving using two M3x16 screws and M3 nuts, and two **Spacers**.



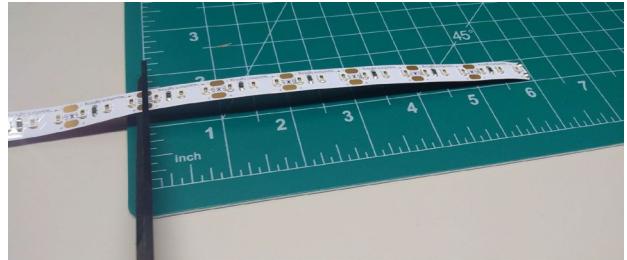
- 2 For the **R from above**, **W from above**, and **G from above** screw terminals, cut about 5 inches of wire (6 pieces total). Connect one end of the wire to the screw terminals on this board and leave the other ends loose for now.



- 3 Cut four 6-inch sections of **red LED strip**, four 6-inch sections of **green LED strip**, and two 6-inch sections of **white LED strip**. Cut on the indicated cut lines with contacts.

Because the red and green strips look identical when unlit, you may want to mark them to avoid confusion.

**DS:** when cutting the pieces, make sure you cut evenly down the middle of the copper areas as you'll need space for soldering later



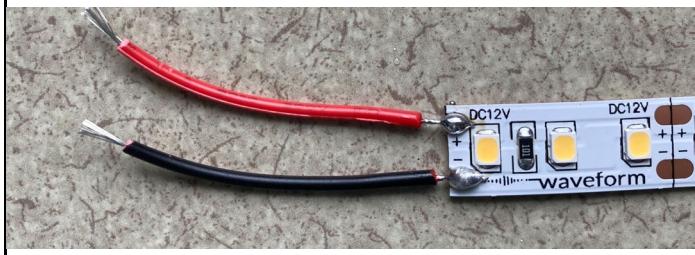
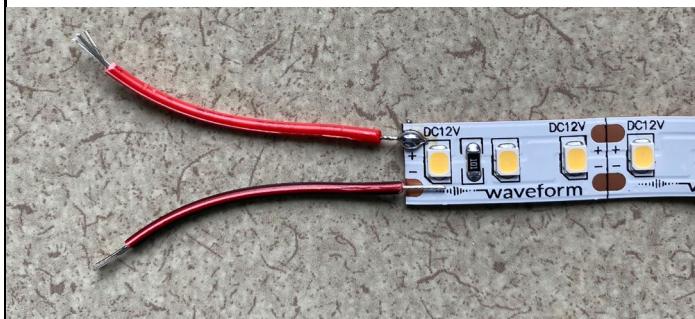
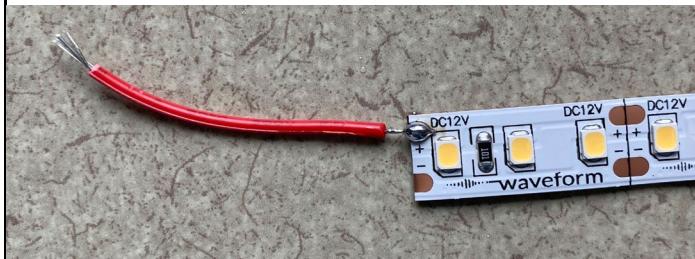
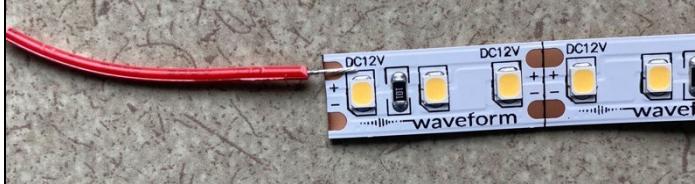
Carefully solder 1.5" wire onto LED strips with the following instructions:

1. Cut 1.5" of red and black electrical wire and remove the myelin sheath on both ends
2. Twist the ends of the wire so it is condensed and not hairy
3. Flatten the LED strip horizontally with the (+) metal end away from you and the (-) metal end towards you
4. **Orientation matters!** If you incorrectly solder the wrong ends, you'll have to solder again. Make sure the copper (+) and (-) ends of the LED strip you solder match up with the (+) and (-) inputs of the screw terminals on the board!!
5. Do one wire at a time. Place the red exposed wire end on top of the copper (+) end of the LED strip as shown in the image and solder
6. A helpful tip in soldering this way is to hold the soldering gun tip and wire closely above and quickly melt the wire to create a droplet, then gently push down the droplet, engulfing the wire and contacting the copper (+) end of the LED strip
7. Pull the wire to see if it falls out. If it falls out, simply re solder.
8. Repeat for the copper (-) end of the LED strip and test to make sure it does not fall out

# STOP

Check to make sure the orientation is correct.  
Hover the LED strip next to the screw terminal and see that the (+) and (-) ends match up

DS + GB steps 1 - 8



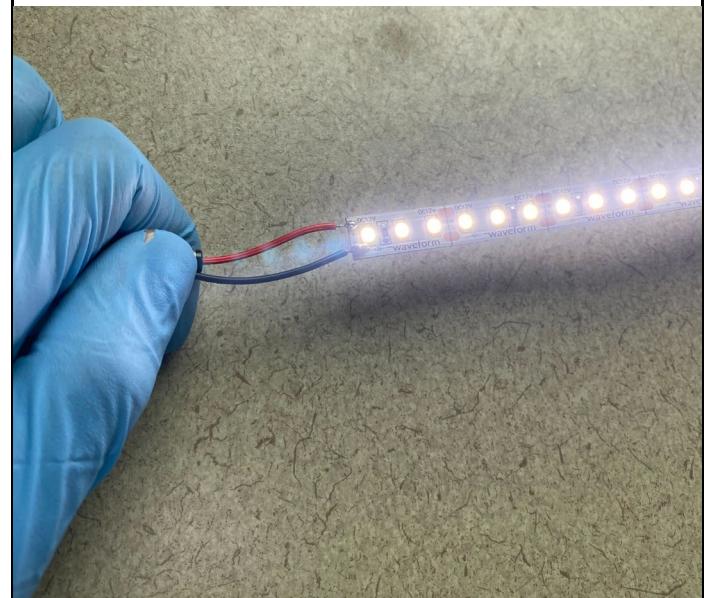
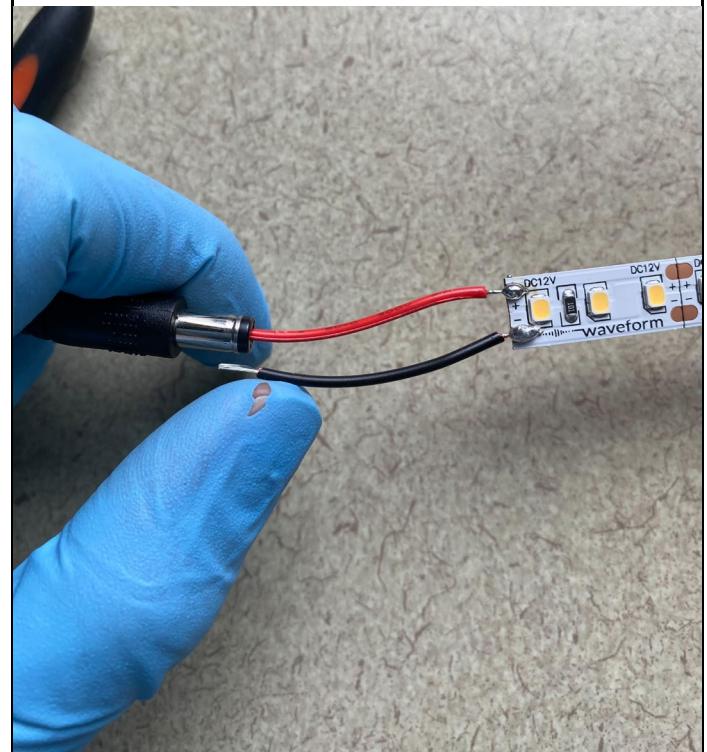
4

Test the LED strip with an AC power cord. Plug the AC power cord and put the red (+) end of the wire inside and touch the black (-) end on the outside.

The light should turn on

The image on the right is just for white light. The red LED strip should emit red light, green LED strip will emit green light.

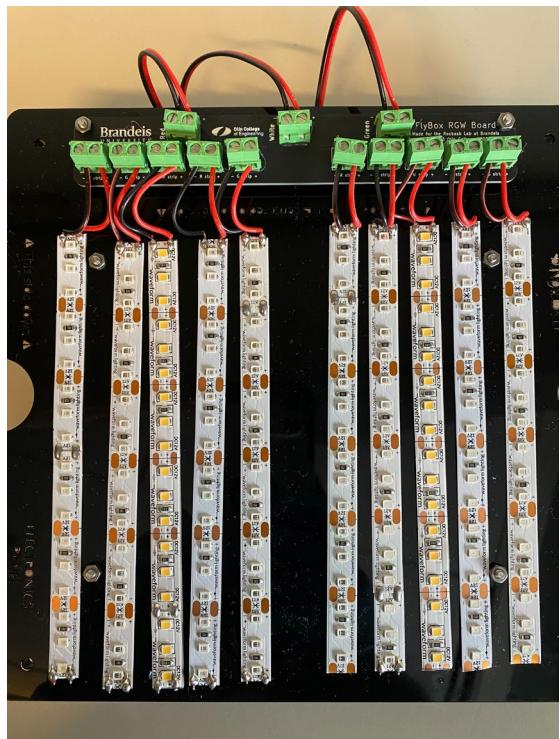
DS + GB



- 5 Insert the ends of the **LED strip connector** into the corresponding screw terminal on the **LED board** making sure to connect the positive wire to the positive side of the screw terminal and vice-versa for the negatives. Remove the tape backing on the **LED Strips** and press them down into position, as indicated by the engraving.

These are a tight fit! It is recommended to start on the outside edges and work your way to the center.

**DS:** Make sure you do not cover the holes around the ends of the board.

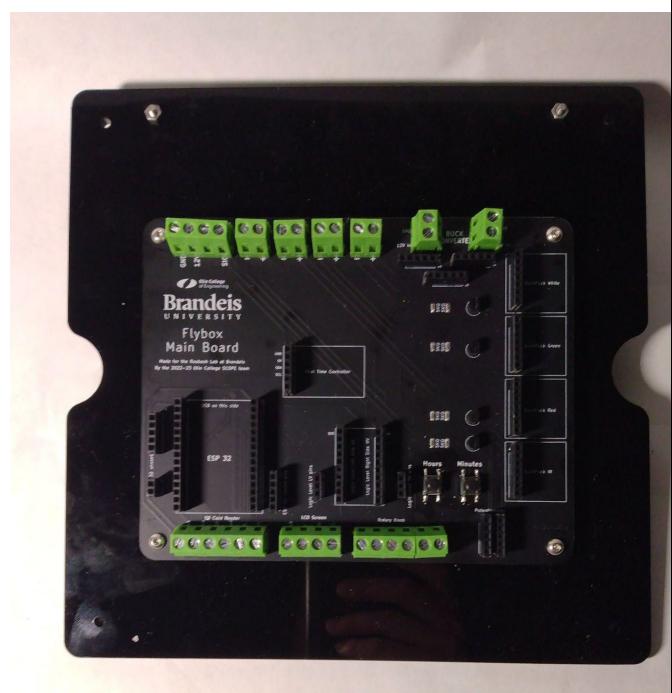


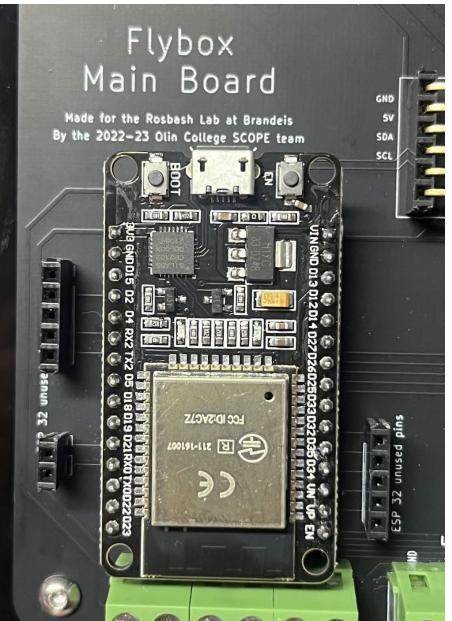
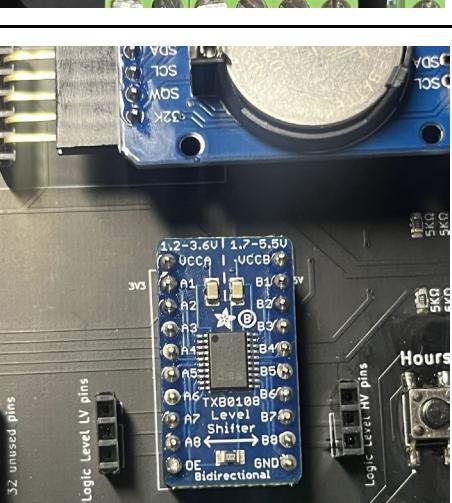
### Top Side of Electronics Panel

- 1 On the un-engraved side of the **Electronics Panel**, attach the **Main Board** using four M3x16 screws and nuts, and four **Spacers**. The main text on the board should face the front door.

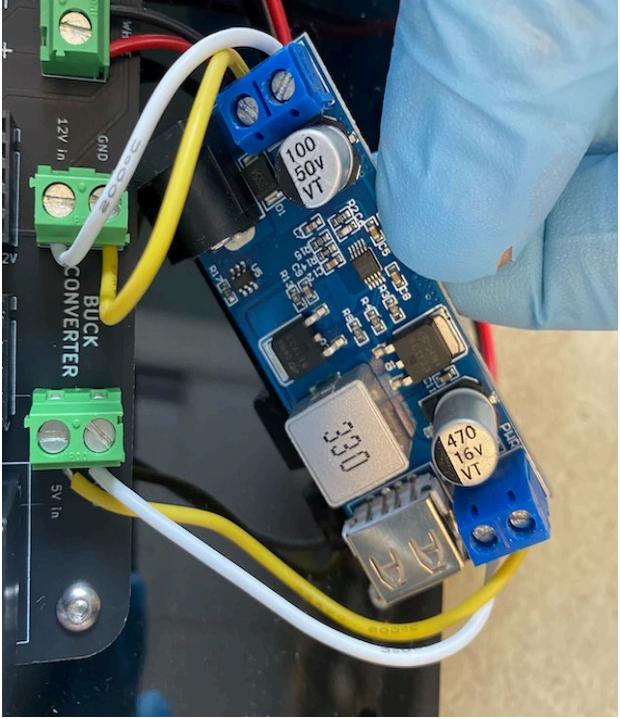
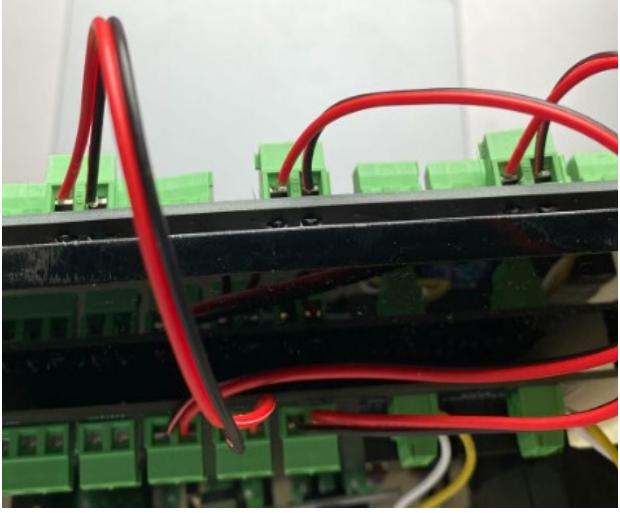
**DS:** Orientation matters! Make sure the Red, White and Green screw terminals on the **RGW Board** are on the same side as the Red, White and Green screw terminals on the **Main Board**.

If the orientation is incorrect, you'll have to flip it.



2	<p>Insert the <b>ESP-WROOM-32</b> component into the corresponding area on the <b>Main Board</b> (lower left), plugging the male pins on the component into the female pins on the main board, with the micro-USB slot facing upward.</p>	
3	<p>Solder the <b>Logic Level</b> component and insert the <b>Logic Level</b> component into the corresponding area on the <b>Main Board</b> (lower middle), plugging the male pins on the component into the female pins on the main board. Make sure the 3V3 and 5V labels on the main board match up with their corresponding labels on the Logic Level component.</p> <p><b>DS:</b> When soldering, keep one extra metal thing so the metal ends do not fall out and push in the metal ends onto a soft material, so it does not move.</p>	

4	<p>Insert each of the four <b>BuckPuck</b> components into the corresponding area on the <b>Main Board</b> (right side), plugging the male pins on the component into the female pins on the main board.</p> <p><b>DS:</b> If you find yourself using the last BuckPucks, order more immediately as these parts have a long lead time to order</p>	
5	<p>First, insert the <b>Button Cell Battery</b> into the <b>Real Time Controller</b> component. Then, insert the <b>Real Time Controller</b> male pins into the female end of the <b>90 Degree Header Pins</b>, and connect the male pins into the <b>Real Time Controller</b> area indicated on the <b>Main Board</b> (middle left).</p>	
6	<p>Insert the <b>Audio Potentiometer</b> component into the corresponding area on the <b>Main Board</b> (lower right), plugging the male pins on the component into the female pins on the main board. The pins will be to the left of the knob, as indicated in the photo.</p> <p><b>DS + GB:</b> This step is difficult, and you will not be able to fit the metal ends inside completely. This is okay.</p> <p><b>EK:</b> This piece does not fit very tightly or securely. You may want to wait until the box is assembled and the board is in place to attach it as it will likely fall out.</p>	

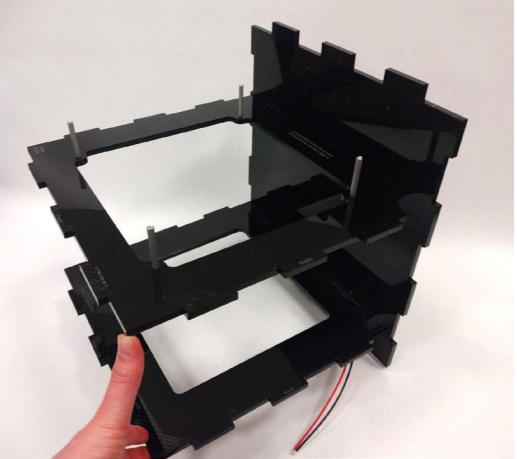
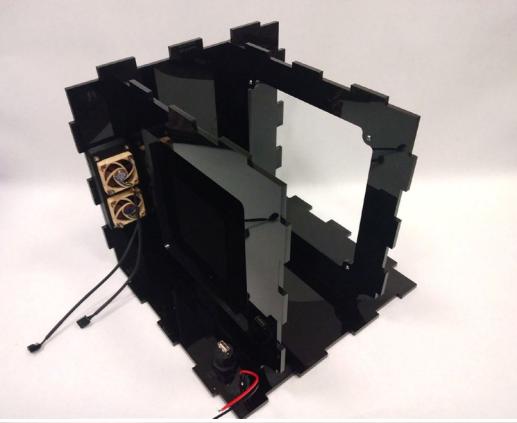
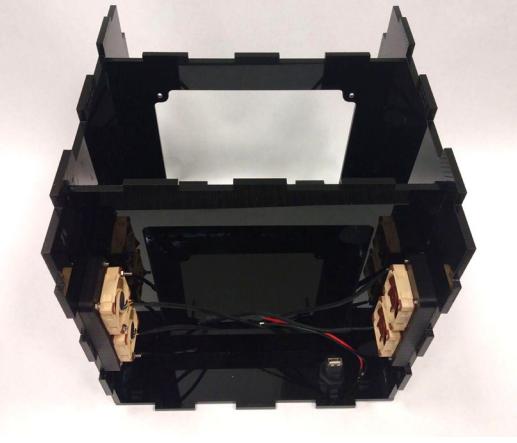
7	<p>Place the <b>Buck Converter</b> above the Main Board. Terminals are labeled on the back of the buck converter</p> <p>Connect the VIN+ screw terminal on the Buck Converter to the 12V in on the Main Board</p> <p>Connect the VIN- screw terminal on the Buck Converter to the GND on the Main Board</p> <p>Connect the 5V screw terminal on the Buck Converter to the 5V in on the Main Board</p> <p>Connect the GND screw terminal on the Buck Converter to the GND on the Main Board</p> <p><b>NOTE:</b> <u>Be very careful when wiring this component.</u> Wiring this component incorrectly may cause significant damage to the box and to any laptop connected to it. Make sure the 12V and the 5V sides are facing the correct direction.</p>	
8	Connect the <b>Red screw terminal</b> in the top left corner to the corresponding wires from the <b>LED Board</b> .	
9	Connect the <b>Green screw terminal</b> in the top left corner to the corresponding wires from the <b>LED Board</b> .	
10	Connect the <b>White screw terminal</b> in the top left corner to the corresponding wires from the <b>LED Board</b> .	

## Box Structure Assembly

As noted above, the laser-cut FlyBox panels are marked with the name of the part in ALL CAPS, and attaching parts in lower case. On most panels, the engraved side faces into the inside of the box.

Striped hatching patterns indicate areas where glue should be applied when joining panels. A row of striped finger-joint tabs or a striped panel face will have the name of the attaching panel in lower case. A “dry fit” without glue can be done first to understand how the box is assembled without permanently fixing the pieces.

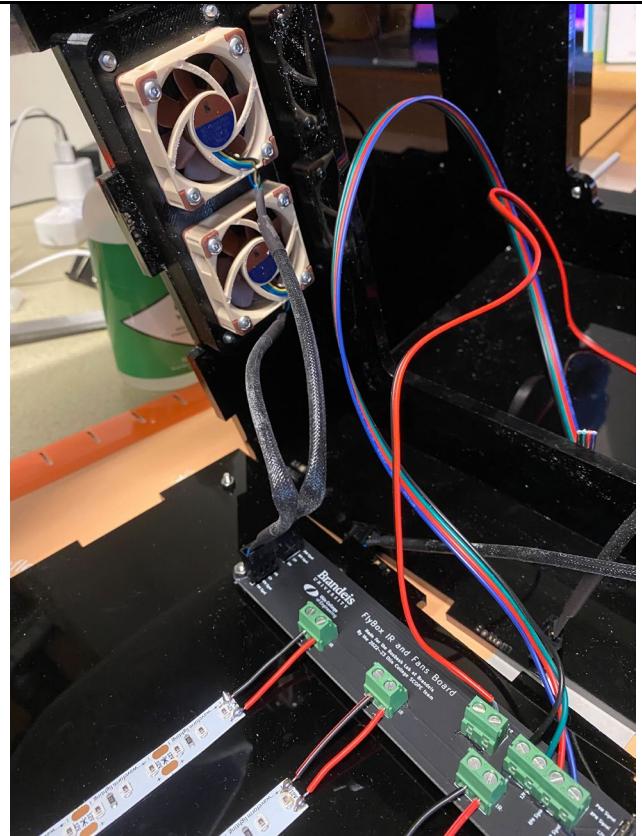
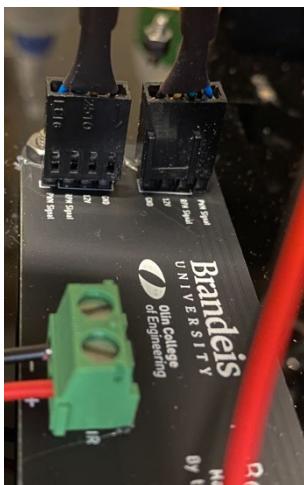
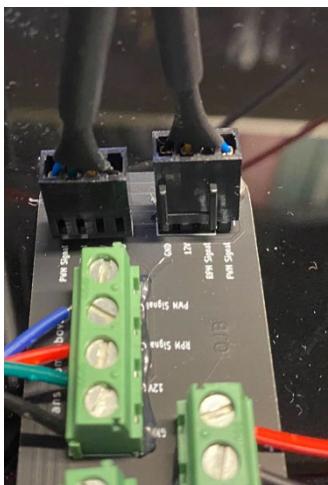
EK: Pieces should fit securely, but you should not have to wiggle them or use a lot of force to fit them together. If pieces are too tight, use a file to shave down the tabs until the pieces fit together easily. File a little at a time and check the fit until they are at the correct tightness.

<p>1 Glue <b>Upper Floor</b> and <b>Lower Floor</b> into the <b>Back Wall</b> panel, referring to markings on each panel for alignment. The rounded corner cut-outs in the <b>Upper</b> and <b>Lower Floor</b> should be facing toward the right side of the <b>Back Wall</b>.</p> <p><b>EK:</b> DO NOT GLUE ANYTHING TO THE BACK PANEL!!!!!! Keeping it removable makes troubleshooting and repairing the flybox a much easier and less frustrating experience. It will be secured in place with gaff tape once the box is checked over after assembly.</p> <p><b>DS:</b> Before gluing on the acrylic pieces, fit them in together without glue first to make sure they connect smoothly without squeaky noises</p> <p><b>DS:</b> Please wear gloves – acrylic glove is very strong and can burn your skin</p> <p><b>DS:</b> You may want to put covering (newspaper, old paper, etc.) down as the glue can be messy</p>	
<p>2 Glue on the <b>Left Wall</b> and <b>Right Wall</b>, with the fans facing in.</p> <p><b>EK:</b> to avoid getting glue on the back panel, do not put any glue on the last cm or so of the walls/floor closest to the back panel.</p> <p><b>DS:</b> Sometimes the boards may not fit in completely – you may have to grind or shave down the connecting ends or try using different pieces</p> <p><b>DS:</b> If you need to shave down any pieces, do so bit by bit. Shave then try, shave then try, etc.</p>	 

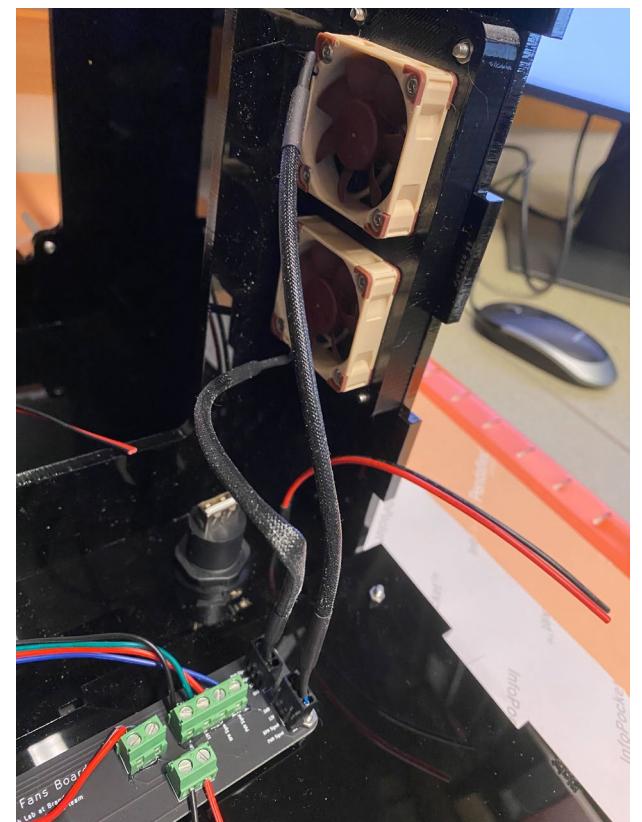
3

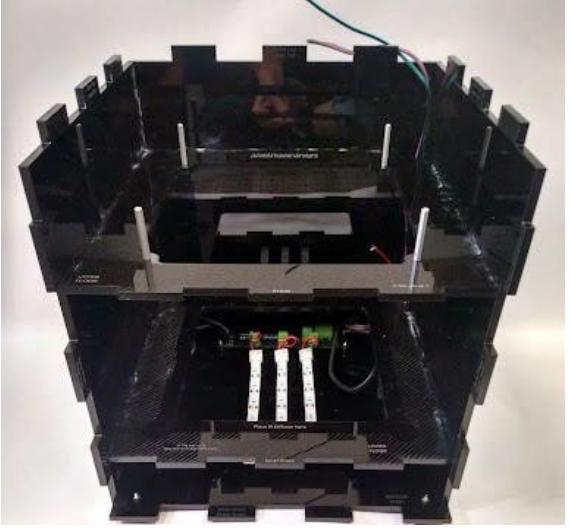
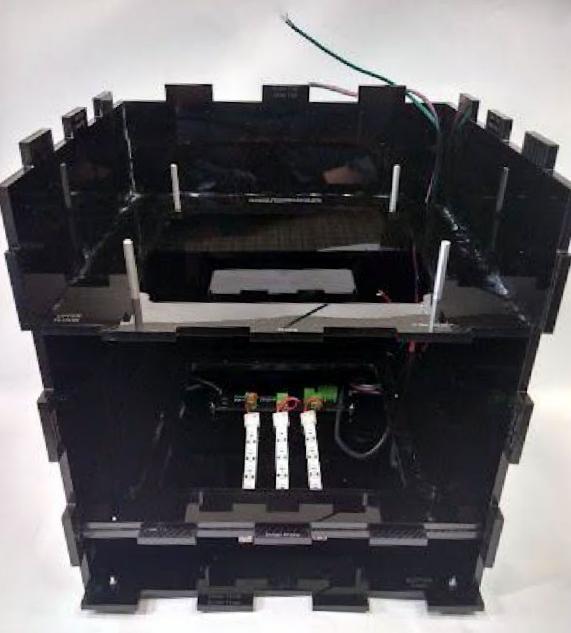
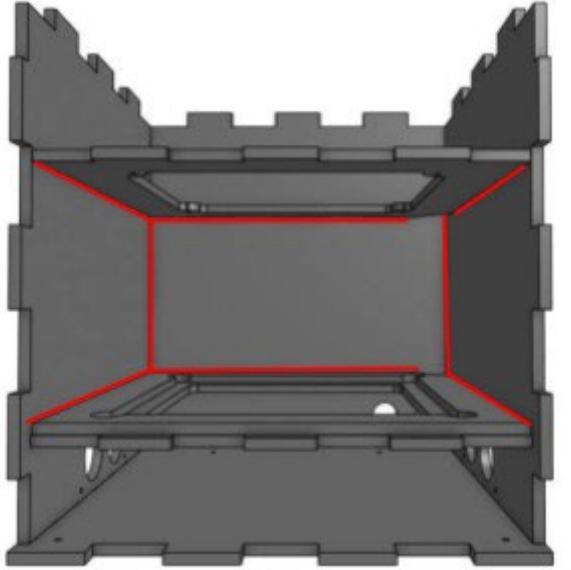
DS + GB: Take the **Bottom**, plug in each **Fan** to the male header pins on the board. The orientation matters for each fan! Please look carefully at the images on the right to see which fan goes to which male header pins.

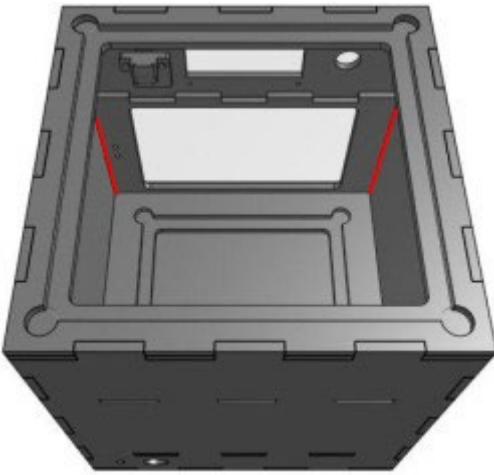
Please see below to see the orientation of the fan female receptor. If the wires are not connected properly the fans will not work.

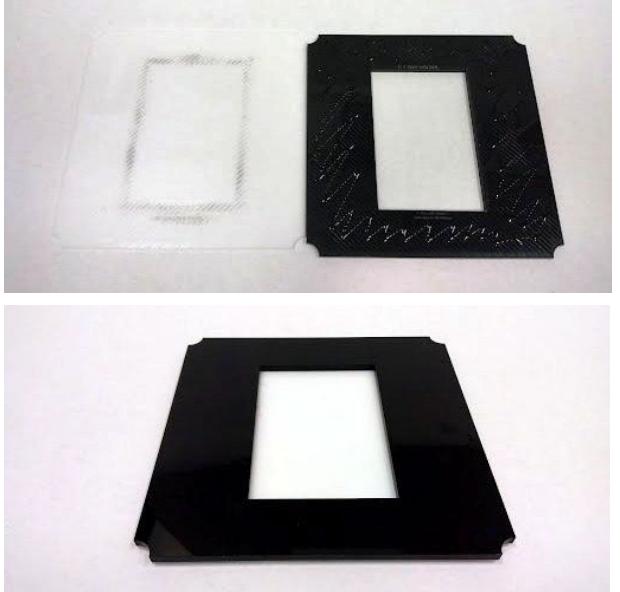


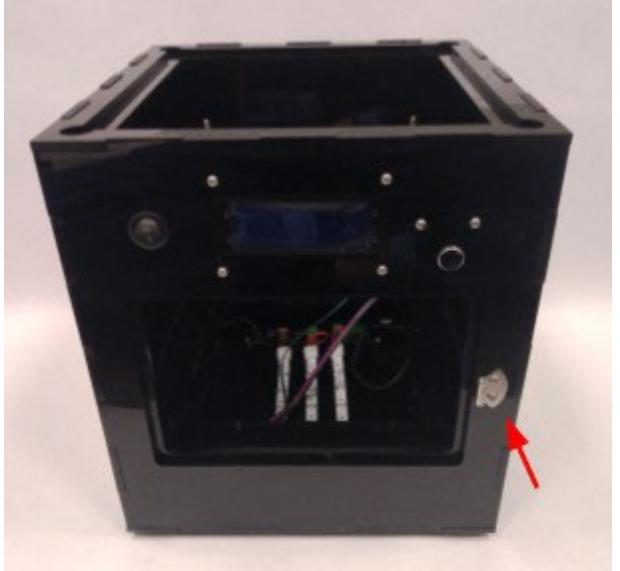
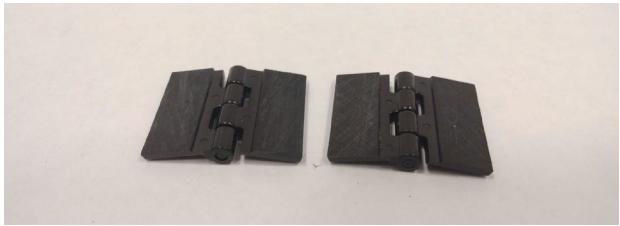
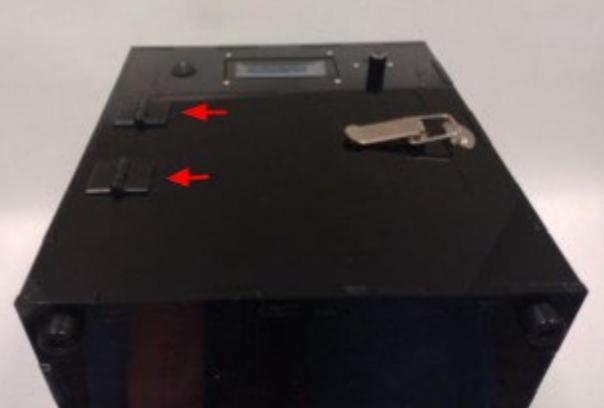
EK: you can see the colored wires on the fans right before they go into the plug. Plug the blue side into the pin that says PVW signal, and the black side into the pin that says GND.



4	<p>Glue on the <b>Bottom</b>, with the IR lights and board facing in. The 20-inch wires of the <b>Fans from above</b> and <b>IR from above</b> screw terminals can be routed upward through the round channel in the back right corner.</p> <p><b>EK:</b> remember not to glue anything to the back panel</p> <p><b>DS:</b> Before gluing down the <b>Bottom</b>, push the wires (the 4-strand wire and 2-strand wire) behind the fan wiring. Not the end of the world if you do not, will make it a lot easier after the <b>Bottom</b> is glued in.</p>	
5	<p>Apply glue to the striped surface of the <b>Lower Floor</b>. Glue on the <b>IR Alignment Panel</b>, striped face to striped face. The round corner cutouts on the back right of each panel should align.</p> <p><b>EK:</b> the pieces no longer have stripes. Glue text side down.</p>	
6	<p>Silicone can be applied to the inside corners of the central chamber, as shown by the highlighted edges to the right. Corners can be tested for light-tightness by shining a flashlight over them and checking that light doesn't go through.</p> <p><b>DS:</b> The silicone tube can be hard to press, expect some resistance and a forearm workout</p> <p><b>EK:</b> Instead of silicone, you can apply gaff tape over the joints on the outside of the flybox. It is easier to get a lightproof seal, and you don't have to deal with the sticky, smelly silicone.</p>	

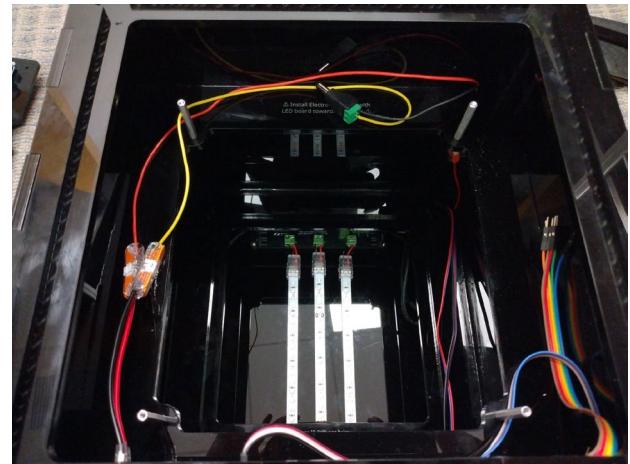
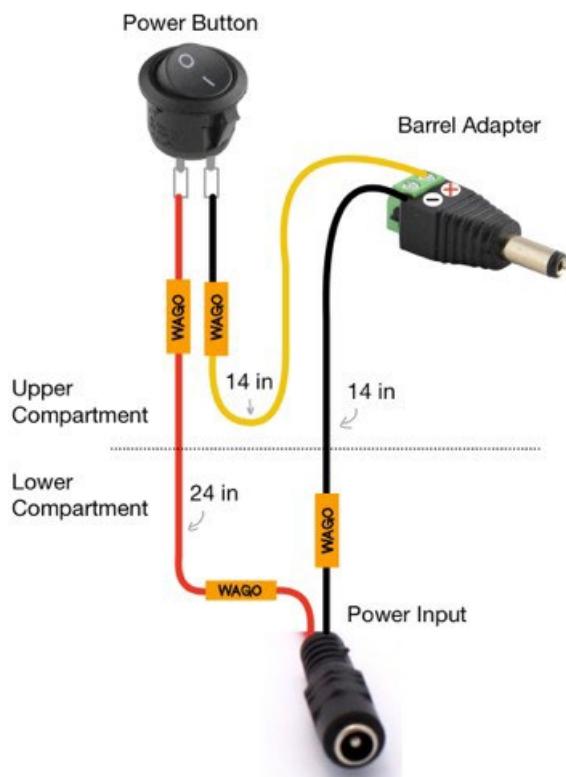
7	<p>Glue on the <b>Inner Top</b> and <b>Inner Front</b>. These panels should have the striped sides facing outward.</p> <p><b>GB:</b> Tape applied on the inside and outside can be used instead of silicone</p>	
8	<p>Glue on the <b>Outer Top</b> and <b>Outer Front</b></p> <p><b>DS:</b> The order does not really matter</p> <p><b>DS:</b> When gluing in the <b>Outer Front</b>, be careful with guiding the exposed wires into the box as they may get unplugged. If they get unplugged, a long skinny nose plier helps to re-plug connections.</p>	
9	<p>Reaching in from the top, apply silicone to the corners of the center chamber where the <b>Inner Front</b> meets the <b>Left Wall</b> and <b>Right Wall</b>, as shown by the highlighted edges to the right.</p> <p><b>EK:</b> This silicone has also been replaced with tape</p>	 <p><i>The Upper Floor has been hidden in this image for clarity</i></p>

10	<p>Glue the <b>Fly Tray Holder</b> to the <b>IR Diffuser Panel</b> and sandwich it together tightly.</p> <p>Let glue dry! Blue tape can be used to secure parts in place as they dry.</p>	
12	<p>Using 2 M4x12 screws and nuts, screw in the larger latch piece on the unengraved side of the <b>Front Door</b>, such that the side with the spring mechanism is closer to the edge.</p> <p><b>DS:</b> For this you will need a slightly larger sized Allen wrench as the electric screwdriver's adapter is too small!</p> <p><b>DS:</b> Watch the orientation of the latches!</p>	

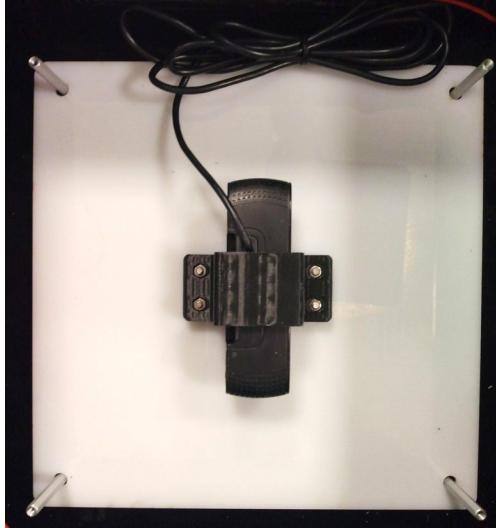
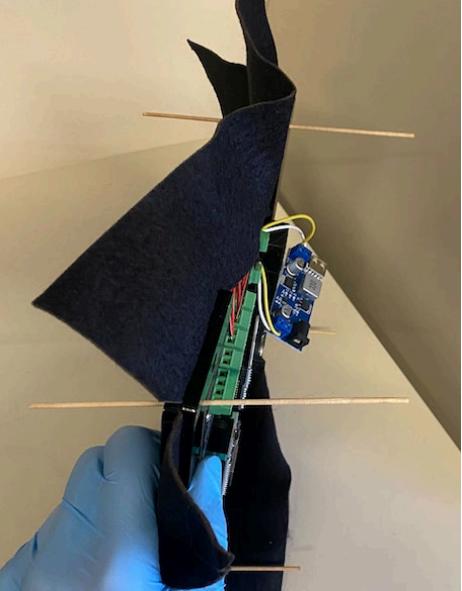
13	<p>Using 2 M4x20 screws and nuts, screw in the smaller latch piece on the right side of the <b>Outer Front</b>, such that the hook-shaped side is nearest the front door opening.</p> <p><b>DS:</b> Watch the orientation to make sure it is correct</p>	
14	<p>Rough up the inner faces of both hinges using sandpaper to increase adhesion surface area.</p>	
15	<p>Gently press the <b>Front Door</b> into place on the front of the box with the latch on the right side. Ensuring each hinge is vertical and aligned over the gap, glue them to the left side of the <b>Front Door</b> and <b>Outer Front</b>.</p>	
16	<p>Let glue dry! Blue tape can be used to secure parts in place as they dry. Do not open the <b>Front Door</b> until the glue has had time to set fully.</p>	

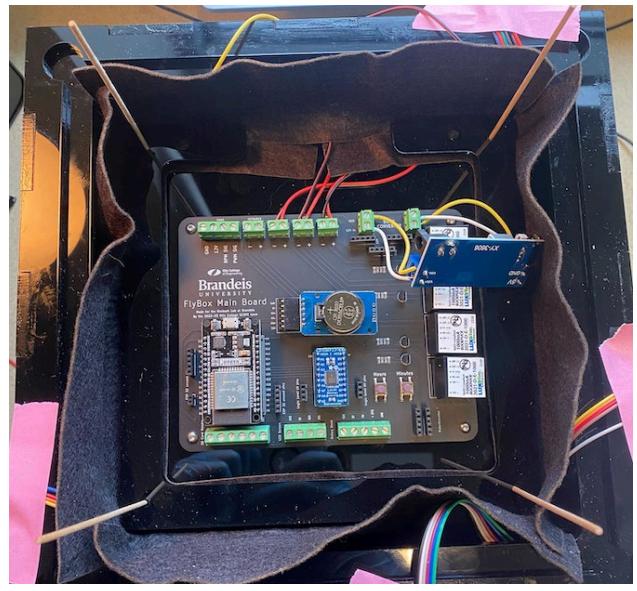
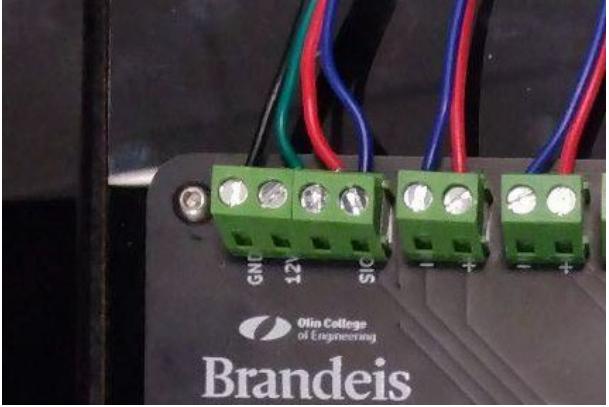
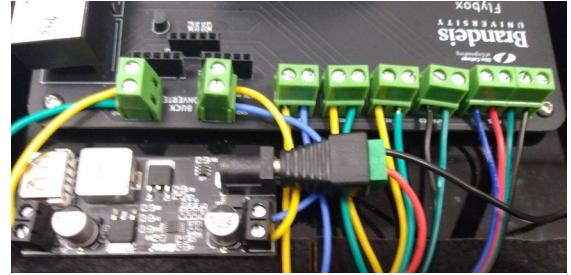
## Final Wiring

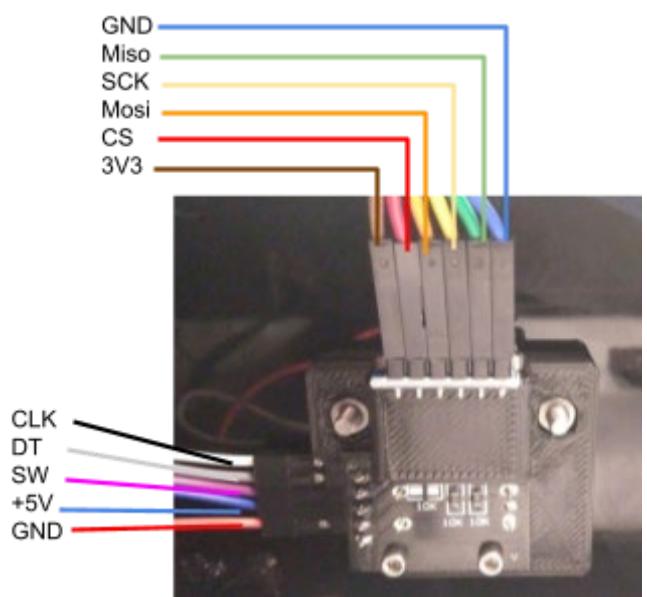
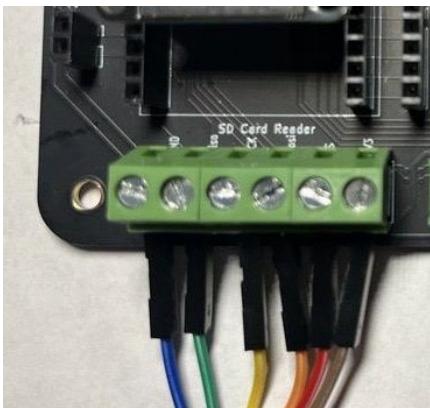
- 1 Attach a **WAGO inline wire connector** to each end of the **Power input**.  
 Attach a **WAGO inline wire connector** to each end of the **Power Switch** wires.  
 Cut a 24-inch length of red wire. Connect it to the red wires of the **Power Input** and **Power Switch** using the **WAGO inline wire connectors**.  
 Cut a 14-inch length of yellow wire. Connect one end to the black wire of the **Power Switch** using the **WAGO inline wire connector**. Connect the other end to the + side of the **Male Barrel Adapter**.  
 Cut a 14-inch length of black wire. Connect one end to the - side of the **Male Barrel Adapter**. Connect the other side to the black wire of the **Power Input** using the **WAGO inline wire connector**
- EK:** add an extra 6 inches to the red and black wires so that the removable back panel can be set down without pulling on the wires.
- DS:** Do not forget to weave the wires through the opening on the far-right corner and upward
- It may be dark and hard to see inside the fly box at this stage – you can grab lights used for microscopes or any other light source



Top view showing WAGO connectors attached to the Power Switch and Male Barrel Adaptor

2	<p>Take the <b>LED Diffuser Panel</b> and attached <b>Camera</b>. Run the camera cable down the back right corner channel and plug it into the inside side (USB A port) of the <b>USB Jack</b>.</p>	
3	<p>Slot the <b>LED Diffuser Panel</b> and attached <b>Camera</b> over the standoffs on the <b>Upper Floor</b>. The <b>Camera</b> lens should point into the center chamber and the cable of the <b>Camera</b> should extend toward the <b>Back Wall</b>.</p> <p><b>DS:</b> There will be a lot of wires sticking outward from all places – make sure the <b>LED Diffuser Panel</b> does not hit or sit on any of them when placing downward through the standoffs. Carefully move the wires out of the way. Use nose head pliers or tape.</p>	
4	<p>Place the felt <b>Light proofing Curtain</b> on top of the <b>Electronics Panel</b> and the <b>Curtain Clamp</b> on top of that. The wires from the <b>LED Board</b> should run through the slit in the <b>Light proofing Curtain</b> and the gap in the <b>Curtain Clamp</b>.</p> <p>Insert each wooden stick Completely through the <b>Curtain Clamp</b>, <b>Light proofing Curtain</b> and <b>Electronics Panel</b>. It helps to hold it sideways</p> <p>Once all 4 wooden sticks are through, you should be able to hold the amalgamation of the 3 parts like a sandwich.</p> <p><b>Method by DS</b></p>	

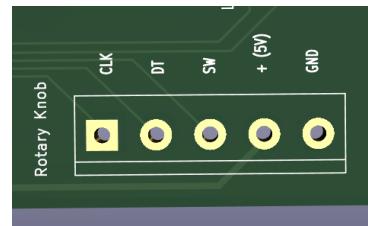
5	<p>Carefully place the <b>Electronic Panel</b> “sandwich” on top of the 4 standoffs and insert each wooden stick completely into the standoff hole. It may take a while to connect the wooden stick with the standoff hole, but you should feel it go in.</p> <p>Do this one at time: Take out a wooden stick and connect a M3x16 screw through all the holes.</p> <p>Repeat until all 4 holes are connected.</p> <p>Orientation matters! Everything should be positioned such that the screw terminals in the <b>Main Board</b> for the <b>SD Card Reader</b>, <b>LCD Screen</b>, and <b>Rotary Knob</b> are at the front.</p> <p><b>Method by DS</b></p>	
6	<p>The Fans and IR wires (pulled through the corner channel earlier) can be connected to the <b>IR</b> and <b>Fans</b> screw terminals on the <b>Main Board</b>. Take care to connect the correct wires</p>	
7	<p>Plug the <b>Male Barrel Adapter</b> into the <b>Buck Converter</b>.</p>	 <p><i>Barrel Adapter plugged into the Buck Converter</i></p>

8	<p>Connect the pins from the <b>Display</b> component in the front panel to the display screw terminal portion on the main electronics board, matching the labels on the board to the display, using the male-to-female jumper wires.</p>	
9	<p>Using male-to-female jumper wires attached earlier, connect the <b>SD Card Reader</b> pins to the SD card screw terminals on the <b>Main Board</b>, matching the labels on the board to the display.</p> <p>Using male-to-female jumper wires attached earlier, connect the <b>Rotary Encoder</b> pins to the Rotary Knob screw terminals on the <b>Main Board</b>, matching the labels on the board to the display.</p>	 <p><i>SD card reader and rotary encoder pins with label</i></p> 

*SD card reader terminal on main board*



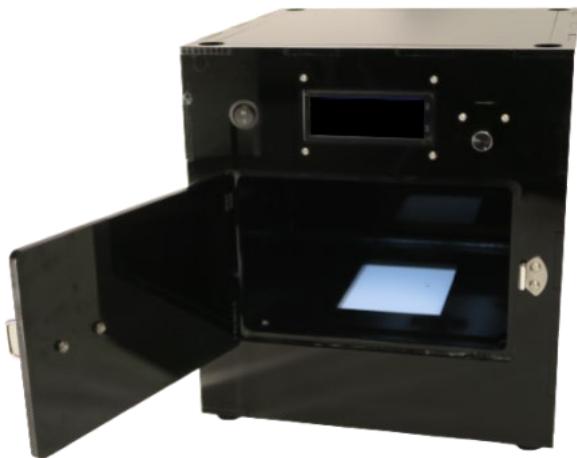
*Rotary knob terminal on main electronics board*



*Labeled pins for rotary knob on main electronics board*

Remove any protective film from the **Display Screen**. The **IR Diffuser Panel** (with the **Fly Tray Holder** glued on top) can be set inside the box short edge to the front, loosely fitting in the **Alignment Panel**. Tuck the **Light proofing Curtain** down around the edges of the **Electronics Panel**. The **Top Hatch** can be placed in the space on top of the box, flush with the **Outer Top** panel.

Congratulations! You have completed assembly of the FlyBox. Now on to loading firmware!



# Firmware Loading

Before you can use the box, you will need to install its firmware. You should only ever need to install the firmware on the box once, unless further updates are made.

## Setting up the Arduino IDE

You will need to set up an Arduino IDE. You can download and install the Arduino IDE by following the following setup guide:

<https://docs.arduino.cc/software/ide-v2/tutorials/getting-started/ide-v2-downloading-and-installing>

## Setting up the Arduino IDE for ESP32 Microcontroller

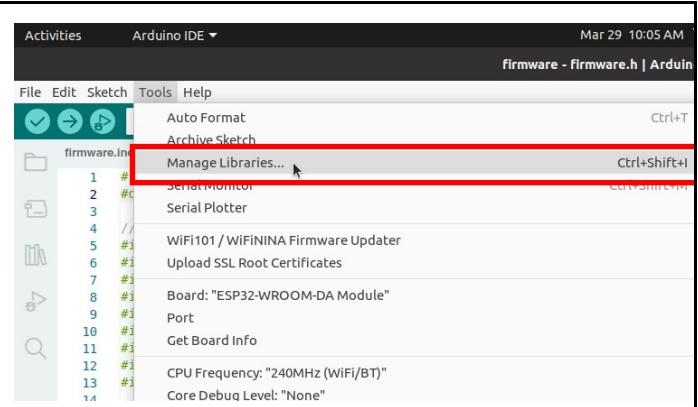
By default, the Arduino IDE doesn't support the microcontroller in the FlyBox. To add compatibility with the FlyBox, follow the following guide:

<https://randomnerdtutorials.com/installing-the-esp32-board-in-arduino-ide-windows-instructions/>

## Installing the necessary libraries in Arduino

There are a few external libraries that you will need to install in Arduino.

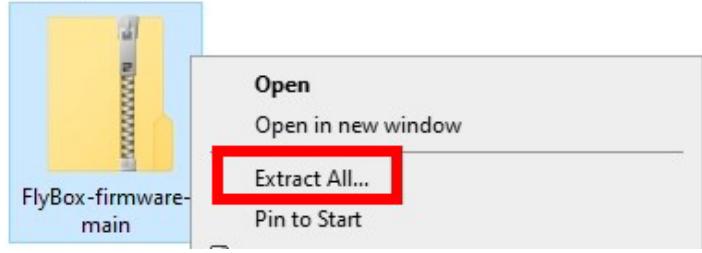
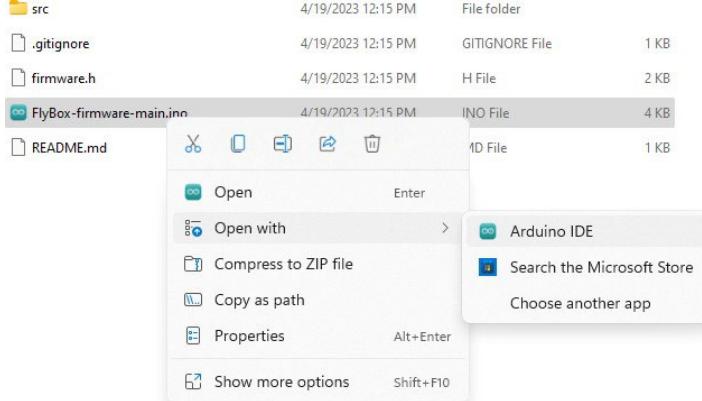
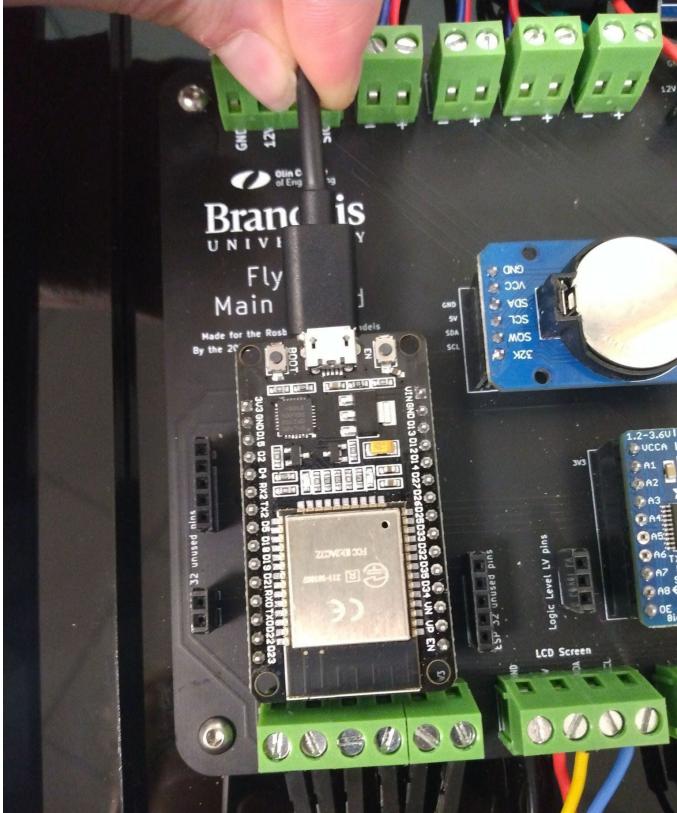
- 1 Go to "Tools" in the top menu bar, then under the drop down menu, select "Manage Libraries..."

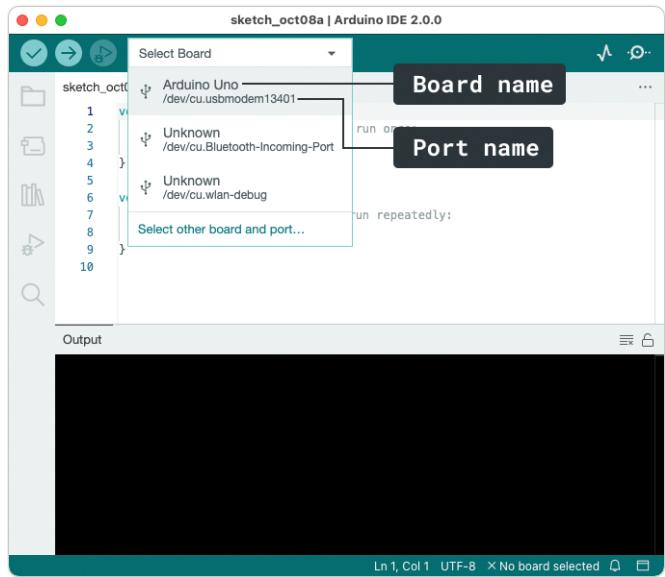
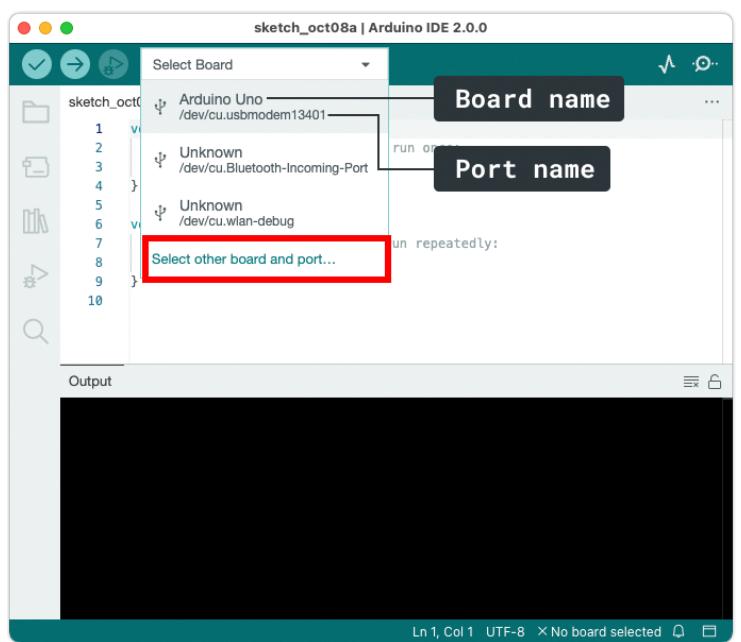


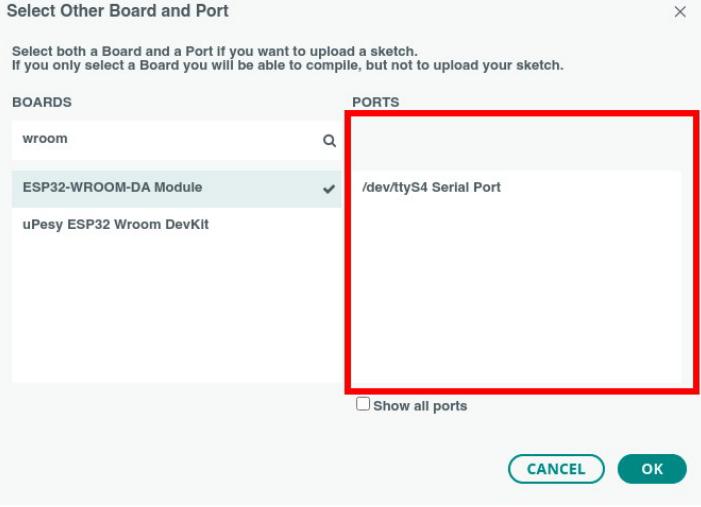
2	<p>To add a library to the Arduino IDE, Search and install the following libraries</p> <ul style="list-style-type: none"> <li>- <b>ArduinoJson</b> by Benoit Blanchon</li> <li>- <b>RTClib</b> by Adafruit <ul style="list-style-type: none"> <li>- May require additional downloads, complete these</li> </ul> </li> <li>- <b>LiquidCrystal I2C</b> by Frank de Brabander</li> <li>- <b>ESP32Encoder</b> by Kevin Harrington</li> <li>- <b>Adafruit_BusIO</b> by Adafruit</li> </ul>	<p><b>LIBRARY MANAGER</b></p> <p><b>ArduinoJSON</b></p> <p>Type: All</p> <p>Topic: All</p> <p>ArduinoJson by Benoit Blanchon &lt;blog.benoitblanchon.fr&gt; Version 6.21.0 <b>INSTALLED</b> ArduinoJson supports ✓ serialization, ✓ deserialization, ✓ MessagePack, ✓ fixed allocation, ✓ zero-copy, ✓ streams, ✓ filtering, and more. It is the most popular Arduino library on GitHub ❤️❤️❤️. Check out <a href="#">arduinojson.org</a> for a comprehensive documentation. A simple and efficient JSON library for embedded C++. <a href="#">More Info</a></p> <p>6.21.1 <a href="#">INSTALL</a></p> <p>CTBot by Stefano Ledda &lt;shurillu@tiscaliinet.it&gt; A simple, easy to use and straightforward Arduino library for using Telegram bots on ESP8266/ESP32 chips. In order to use this library you need the ArduinoJson library (release 5.13.5 or greater) installed. Inline and Reply keyboard supported. Localization messages supported. Fingerprint authentication and QRCODE decoding libraries.</p>
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## Downloading the Firmware

1	<p>Go to  <a href="https://github.com/Rosbash-Lab-FlyBox/FlyBox-firmware">https://github.com/Rosbash-Lab-FlyBox/FlyBox-firmware</a></p>	<p>ctallum/FlyBox-firmware Public</p> <p>Code Issues Pull requests Actions Projects Wiki Security Insights Settings</p> <p>main · 1 branch · 0 tags Go to file Add file &lt;&gt; Code About</p> <p>Firmware repo for Olin College-Brandeis Rosbash Lab FlyBox scope team</p> <p>ctallum transfer files from main Flybox Repo SodStee 13 minutes ago 2 commits</p> <p>src transfer files from main Flybox Repo 13 minutes ago</p> <p>.gitignore transfer files from main Flybox Repo 13 minutes ago</p> <p>README.md transfer files from main Flybox Repo 13 minutes ago</p> <p>firmware.h transfer files from main Flybox Repo 13 minutes ago</p> <p>firmware.ino transfer files from main Flybox Repo 13 minutes ago</p>
2	<p>Click on the green button labeled “Code” then choose “Download ZIP”</p>	<p>Go to file Add file &lt;&gt; Code Local Codespaces New</p> <p>Clone HTTPS SSH GitHub CLI</p> <p>git@github.com:ctallum/FlyBox-firmware.git</p> <p>Use a password-protected SSH key.</p> <p>Download ZIP</p>

3	Unzip the downloaded folder. It should be titled “FlyBox-firmware-main”	
4	<p>Within the “FlyBox-firmware-main” folder, open the “FlyBox-firmware-main.ino” file in the Arduino IDE</p> <p>It will ask if you want to create a new folder, click yes.</p> <p>Make sure the FlyBox-firmware-main folder has the following files inside:</p> <ul style="list-style-type: none"> <li>- firmware.h</li> <li>- FlyBox-firmware-main.ino</li> <li>- src folder (which has “screens” and “utils” folders inside)</li> </ul>	
5	Connect the ESP32 to your computer using a micro USB to USB A cable	

6	Open up the board selection menu									
7	Click "Select Other Board and Port"									
8	Search and select "ESP32-WROOM-DA Module"	<p>Select Other Board and Port <span style="float: right;">X</span></p> <p>Select both a Board and a Port if you want to upload a sketch. If you only select a Board you will be able to compile, but not to upload your sketch.</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left; padding: 5px;">BOARDS</th> <th style="text-align: left; padding: 5px;">PORTS</th> </tr> </thead> <tbody> <tr> <td style="padding: 5px;">wroom</td> <td style="padding: 5px;">Q</td> </tr> <tr> <td style="padding: 5px;"><b>ESP32-WROOM-DA Module</b></td> <td style="padding: 5px;">✓ /dev/ttyS4 Serial Port</td> </tr> <tr> <td style="padding: 5px;">uPesy ESP32 Wroom DevKit</td> <td style="padding: 5px;"></td> </tr> </tbody> </table> <p style="text-align: right; margin-top: -10px;"><input type="checkbox"/> Show all ports</p> <p style="text-align: right; margin-top: -10px; font-size: small;"><span style="border: 1px solid #ccc; border-radius: 15px; padding: 2px 10px; margin-right: 10px;">CANCEL</span> <span style="border: 1px solid #0072BC; border-radius: 15px; background-color: #0072BC; color: white; padding: 2px 10px;">OK</span></p>	BOARDS	PORTS	wroom	Q	<b>ESP32-WROOM-DA Module</b>	✓ /dev/ttyS4 Serial Port	uPesy ESP32 Wroom DevKit	
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9	On the right half of the menu, select the port your ESP32 is connected to, and click “OK” to close the menu	
10	Click to upload the file	
<b>Common Errors</b> <p>If you see a “library error,” search /users/“your name”/documents/Arudino/Libraries and delete any library you are not using or delete everything and start over</p> <p>If you see a “src/utils/lights.h” not found, you have to move the src folder into the FlyBox-firmware-main folder</p> <p>If you ever see a “Adafruit_I2CDevice.h” not found, it’s because this library is no longer used. The new library used is now “Adafruit_BusIO”</p> <p>Selecting the Board is relatively simple. However, 2 ports may appear after connecting the ESP32. Select the port that contains “USB to UART serial port” instead of a port that may have 4 digit numbers, such as “0001”</p>		

## Running a Test

To run a test, first design a test using <https://rosbash-lab-flybox.github.io/FlyBox/>. You can then download the test and upload it to the box. On the box, you can then select and run the file.

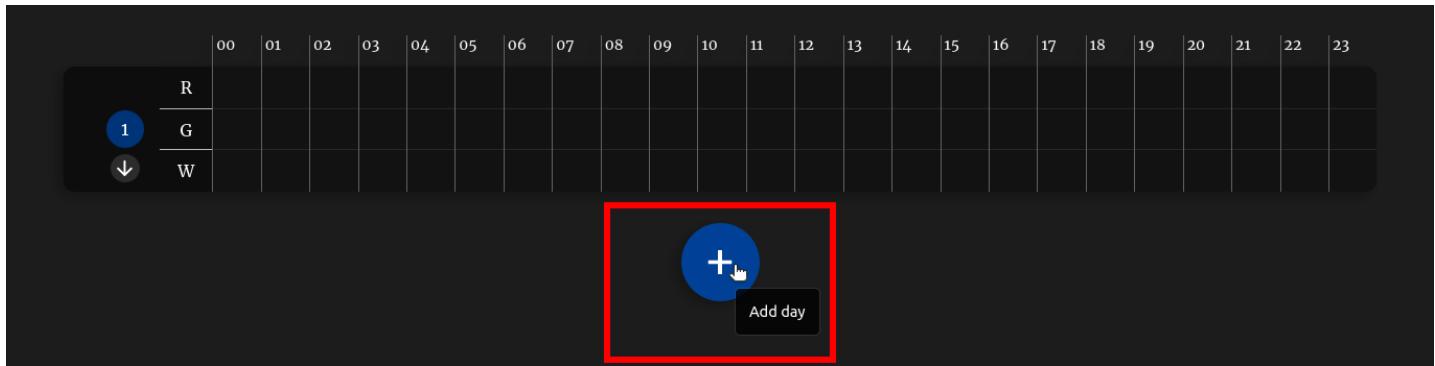
### Using the Rosbash Lab FlyBox Test Creator

The screenshot shows the Rosbash Lab FlyBox Test Creator interface. At the top left is the Brandeis University logo. To its right is the title "Rosbash Lab FlyBox Test Creator". Above the main content area are four buttons: a question mark icon, a circular arrow icon, "Upload test ↑", and "Download test ↓". Below these is a status message "1 Days, 0 Events". The main area features a 24-hour grid from 00 to 23. A vertical column on the left lists activities: "R" at the top, followed by "G" and "W" with arrows indicating they can be moved up and down. A blue "+" button is located in the center of the grid. The entire grid is labeled "1" in the top-left corner of its first row.

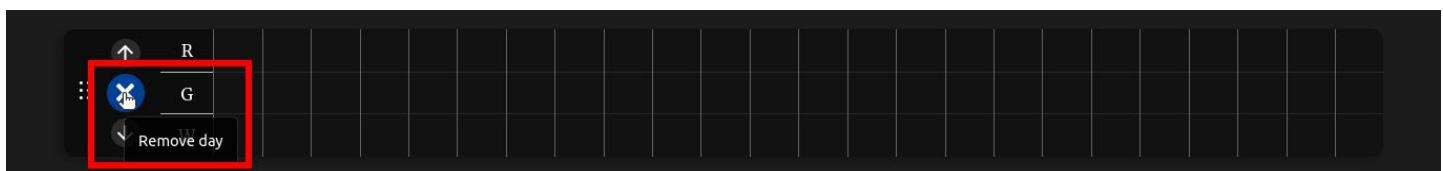
Above is an image of the test creator website. You can use this website to generate and modify test files. To create a test file, you can click and drag to create an event.

This screenshot shows the same interface but with two days of data. The top section "1 Days, 0 Events" has changed to "2 Days, 0 Events". The first day's grid is highlighted with a red box. It contains the same activity list (R, G, W) and a blue "+" button. The second day's grid is partially visible below it, also containing the same activity list and a blue "+" button. The days are numbered "1" and "2" in the top-left corner of their respective grids.

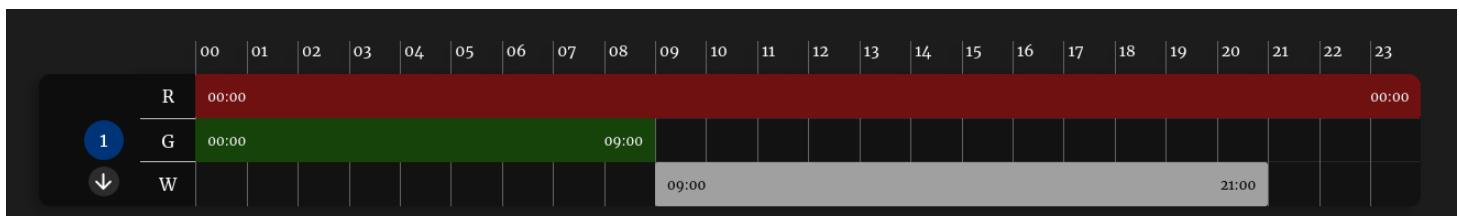
Each individual day of the test is grouped together as seen in the image above. The 24 hours of the day are displayed at the top of the day.



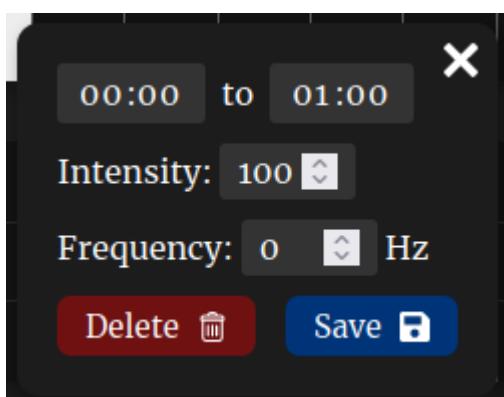
You can add more days to the test by clicking the large circular plus button at the bottom of the screen.



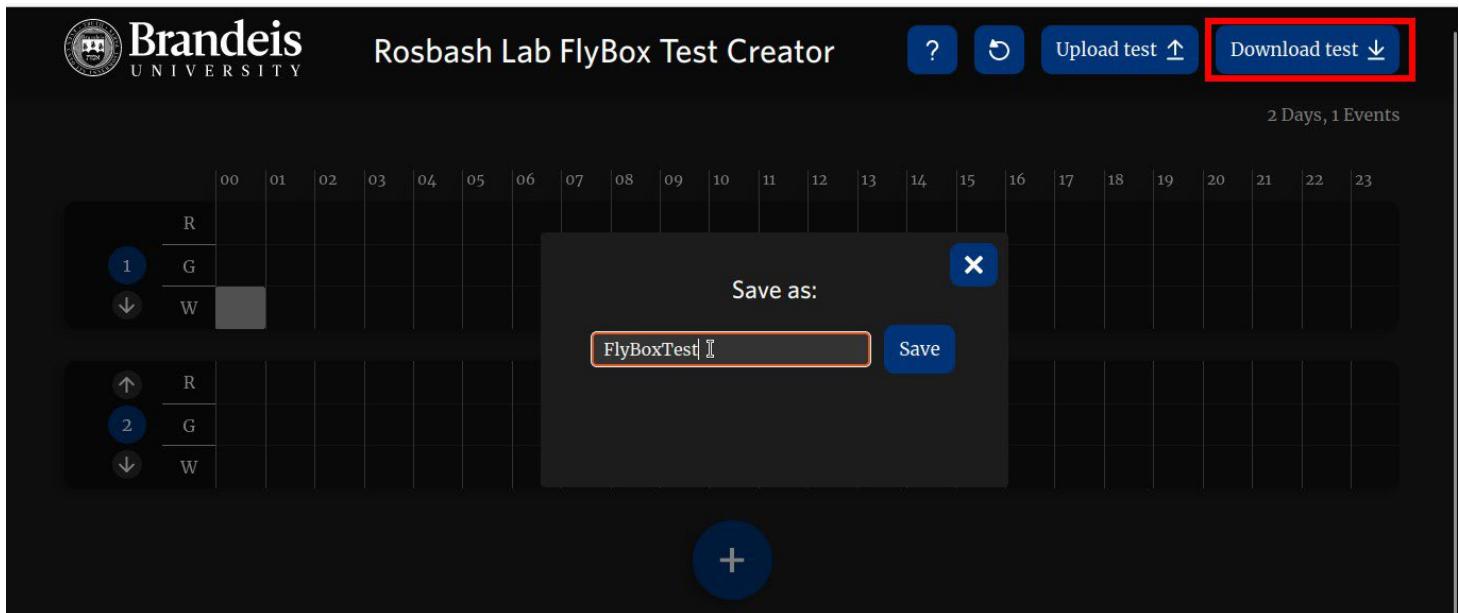
You can delete a day by hovering over the day indicator on the left.



Horizontally, across each day, you can add discrete lighting events. As shown in the image above, the top row is red light events, the middle row is green light events, and bottom is white light events.



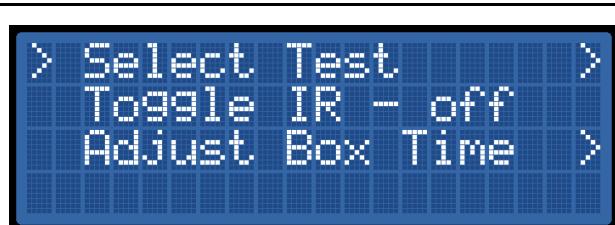
If you click on any event, there is an info panel that appears. In this modal, you can modify the start and stop time, the intensity of the light (from 0-100) and change the frequency of the lights (0 Hz is solid).



Once you are satisfied with your test, you can download the test file locally to your computer using the “Download test” button.

### Running a test file on the FlyBox

After downloading your test file from the FlyBox test creator website, load the file onto an SD card, and insert into the SD card reader slot on the right side of the FlyBox. The FlyBox camera can be connected to your computer using a USB B to USB A cable from the USB output on the back of the FlyBox.

1	<p>This is the intro screen for the box. Click the knob to enter the main menu screen.</p>	
2	<p>This is the main menu screen. You have three options here: select a test file, toggle the IR on and off, and change the time on the box.</p> <p>You can scroll between the options using the rotary knob.</p> <p>If you click <b>Select Test</b>, you will be brought to a screen where you can select your test file. You can scroll between the files using the rotary knob.</p> <p>If you click <b>Toggle IR</b>, you can turn the IR light on and off. The main menu screen will display the current status of the IR light.</p>	 

	<p>If you click <b>Adjust Box Time</b>, you will be brought to a screen where you can adjust the hour and minute on the box. You can change the hour and minute by clicking the hour and minute button on the main electronics panel. Clicking each button will increment the hour/minute by one minute.</p>	 
3	As you run a file, it has a live display indicating percent through the test, which lights are on/off, and the current time	
4	When the test is over, you have the option to return to the main menu by pressing the knob.	

# Appendix

## Calibrating IR

- The toggle IR option in the main menu will turn on the IR lights. While connected to the camera, you can twist the potentiometer (shown on the right) on the Main Electronics Board to decrease or increase the brightness.



## Camera focus

We have noticed issues with the camera sometimes not being able to focus on the tray of flies. This results in a pulling sort of effect as the camera repeatedly shifts focal length, trying to find an object to focus on. When we ran into this issue, we found that the solution was to simply replace the camera with a different one. This resolved the issue.

If switching to a different camera does not work, another solution is to lock the focus of the camera. To do this, you can use the Logitech Capture software ([download here](#)). Using the Logitech Capture software, it is possible to turn off auto-focus and have a fixed focal length. We found some success with this software in it being able to fix the pulsing focus issue.

Another solution is to shift the fly tray up by a small amount. Sometimes, just moving the tray up by a slight amount allows the camera to focus.

## Buying the PCBs from PCBway

<https://www.pcbway.com/>

Each of the manufactured boards should be available through PCBway's "Share and Sell Program." The links for each board are as follows:

- FlyBox Main Board:  
[https://www.pcbway.com/project/shareproject/W547848AS1D13\\_MainBoardGerber3\\_5fe985dc.html](https://www.pcbway.com/project/shareproject/W547848AS1D13_MainBoardGerber3_5fe985dc.html)
- FlyBox IR Board:  
[https://www.pcbway.com/project/shareproject/FlyBox\\_IR\\_Board\\_50aa739c.html](https://www.pcbway.com/project/shareproject/FlyBox_IR_Board_50aa739c.html)
- FlyBox RGW Board:  
[https://www.pcbway.com/project/shareproject/FlyBox\\_RGW\\_Board\\_fc0900b7.html](https://www.pcbway.com/project/shareproject/FlyBox_RGW_Board_fc0900b7.html)

Be sure to order it as PCB+Assembly if you are looking for the PCBs to arrive ready to use (around 4-5 weeks) or just PCB if you have the ability to solder yourself.

## General troubleshooting

If the box isn't turning on correctly, most likely something is wired incorrectly. Common issues are incorrectly wiring the display or the SD card reader. Check that these components have wires connected firmly and in the right place

## **Strange clock readings**

- It is possible that the coin battery in the RTC component has died. If so, replace battery and update the time using the build in time modification screen

- If the RTC is broken, replace component

### **Strange display artifacts**

- If the display is broken, there are a few symptoms. It might not turn on, it may stream random characters, or it may show incorrect text.

### Cleaning

- The FlyBox can be dusted with a microfiber cloth or canned air
- Use water and mild soap (such as dish soap) to clean the FlyBox. Be sure not to get electronic components wet.
- Don't use alcohol to clean the box, it can degrade the acrylic panels.
- Don't use abrasive cleaners, they will scratch the surface of the FlyBox

### FlyBox on a Budget

To lower the cost of the FlyBox, a couple changes can be made:

#### **PCBs**

If you have the ability to solder, you can order unassembled PCBs from PCBway and assemble them yourself. The components of the PCBs are listed in the PCB BOM on PCBway and will need to be ordered separately; they are not included in the FlyBox BOM.

#### **Fans**

The suggested Noctua fans are expensive. Cheaper brands of fans will have two wires, red and black. Put the black wire in the ground position, and the red wire in the power position on the IR/Fans board.

#### **MDF**

Instead of black opaque acrylic, medium density fiberboard can be used for the wall panels (cutsheets 1-6)

### Kitting

There are a number of small pieces of hardware (nuts, bolts, standoffs, etc.) that are easy to miscount or confuse for one another. This may not be an issue in the Rosbash lab. However, if kits are someday made to be sent to another lab or organization, a large bag of unsorted hardware won't be ideal. Instead, we recommend kitting the hardware, which means putting the proper quantity of each type of hardware into an individual bag. There are many companies that offer this service. Here are a few that we found:

- <https://www.associatedfastening.com/products-services/hardware-polybagging/>
- <https://www.mudgefasteners.com/custom-kitting-packaging>
- <https://www.autobag.com/markets-served/hardware-fasteners>
- <https://www.foremostfastener.com/kitting-custom-bagging-services.html>