Regenerating a glovebox

→ Open the N₂ tank in the closet and check the gas level to make sure it has ≥1000 psi remaining.
   If it has <1000 psi, change the tank prior to starting the regeneration cycle.
   *****The amount of N₂ is critical to ensure that there will be enough to finish the entire regeneration cycle. H₂ is highly explosive at high concentrations... you MUST have enough N₂ to maintain a 95%:5% mixture of N₂:H₂!*****

→ For the triple box, make sure that the hose from the valve is attached for the regeneration (check fermentor mixer)

→ If the box needs to be thawed, turn off the refrigeration
   Functions → next → Box cooling (Module 1 and 2) (should turn from green to red)
   “The triple box compressors are all controlled by “Box cooling ”; the double box compressors are controlled separately so be sure to turn off both Box cooling module 1 and module 2.

→ Turn off the circulation and turn on the regeneration
   Functions → Circulation (should turn from green to red) → regeneration (green to red)

Conversely,

Confirm that you want to regenerate the glovebox by touching “yes”.

→ Return to the closet and open the valve to allow the gas to flow into the lab. Open the valve in the lab to the box. The valve in the lab typically does not need to be fully opened.

→ Gas should start flowing and you might hear a howling noise. This is normal.

→ You need to confirm that the flow is appropriate for the regeneration.
   → The N₂ should be on already.
   → Check that the flow is appropriate. Find the flow meter on the glovebox. The ball should be steadily floating between 15-20. If it is not, adjust the flow by closing/opening the valves in the labs that lead to the gloveboxes. Once this is done, return to the cylinder closet.

→ Turn on the H₂ tank. The person before you should have checked to make sure there is enough for a regen. It only takes approximately 100 psi H₂.

→ Now comes the fun part! You need to make sure the N₂:H₂ mixture is approximately 95%:5%.
Look at the two balls on the “high” side of the flow meter. This corresponds to the N₂. Make a note of where the glass (black) and stainless steel are on the flow meter. On the charts on the wall to the right, find the numbers that correspond to the levels of the two balls on the “scale reading” column.

- Note the “flow rate” numbers corresponding to the “scale reading” numbers.
- Average the numbers that correspond to the flow of the N₂.
- Calculate 5% of this number (multiply by 0.05). Remember this number.

Example:

<table>
<thead>
<tr>
<th>Scale reading</th>
<th>Flow rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>&quot;High&quot; N₂ reading (glass): 65</td>
<td>9.1038</td>
</tr>
<tr>
<td>&quot;High&quot; N₂ reading (st. stl): 35</td>
<td>8.1410</td>
</tr>
</tbody>
</table>

Average flow rate is 8.62
5% of this is: 0.431 ← This is the flow setting you will want for H₂.

Look on the charts corresponding to H₂ levels. Find the number you just calculated on the “flow rate” side of the H₂ charts. These correspond to the “scale reading” for the glass and stainless steel balls for the H₂. The “scale reading” numbers should correspond to the flow of the H₂ on the “low” side of the flow meter.

Example:

<table>
<thead>
<tr>
<th>Scale reading</th>
<th>Flow rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>&quot;Low&quot; H₂ reading (glass): 98</td>
<td>422 (this number is 1000x the calc #)</td>
</tr>
<tr>
<td>&quot;Low&quot; H₂ reading (st. stl): 52</td>
<td>0.425</td>
</tr>
</tbody>
</table>

You would then adjust the H₂ flow by the regulator on the H₂ tank or flow meter so the balls are reading at approximately the levels above.

- If the H₂ balls are not moving when you adjust the flow:
  - turn down the flow on the N₂ line
  - make sure the flow meter valve to the “low” side is open
  - increase the pressure of the H₂ line

Once the flow has been adjusted and the H₂ is as close to 5% of N₂ as possible, return to the lab. Check the flow on the flow meter on the glovebox is still between 15-20.

On the glovebox panel, click that the flow has been confirmed and the regeneration should begin.

Regenerations take approximately 16-17 hours. The O₂ in the box will increase. The regeneration is finished when the “Regeneration” tab goes from green to red and the “Circulation” button becomes red again.

If the O₂ levels are high (~<50 ppm), purging the box might be useful (circulation should still be off)

- Functions → Next → Quick purge
  - Purging the box pulls gas directly from the cylinders into the box and will go through a tank pretty quickly. Keep an eye on your N₂ (or Argon) levels.
  - Once the O₂ levels are around 30-40ppm, turn off the purge.

Turn on the circulation by touching “circulation” (should go from red to green). The O₂ levels should drop quickly to <1ppm.

If you turned off the refrigeration, turn it back on by touching the “box cooling (module 1 and 2) buttons

- Be sure to close the valve in the lab, the valve in the closet and shut off both tanks. If a tank is too low for an additional regeneration, replace it. If H₂ is low, please be sure to tell the tank ordering person as we do not keep extras of these tanks.

Note the H₂O pulled off during the regeneration and empty the flask in the sink. Note that you completed a regeneration in the glovebox log (near the computer for the glovebox AKTA).