DAILY PROTOCOL – NOVEMBER 2021

Initial Check

1. Move the PC cursor to activate the screen and check if any of the green lights from the flowers are on. Normally no green lights should be visible unless a nose-poke occurs. If you see a light that is on, check to see if the infrared sensor is blocked by nectar or other residue. Write this down in log if so.
2. Go to the computer and check to see how many visits the animals made. Check the tab "Analysis" to see which side the animals preferred, and then run the shiny app called shiny_gen_timing (on the Desktop) with the data from the previous night.
3. Check to see if any of the animals have drunk less than the recommended amount. Write this down in the log if so.
4. Make a note of the volume consumed by the bats as calculated by the Shiny app in the Log.
5. Remove the nectar reservoir from the Experiment Room. Measure how much nectar is left in the reservoir by pouring it in the measuring cylinder. Make a note of it.
6. If any bat has not drunk enough, place a cup of honey water in the cage during the cleaning procedure.
7. If there is already honey water in any of the cages remove them before starting the cleaning procedure.
8. Check the temperature and humidity in the experiment room on the PC in the preparation room. Write these down in the Log.
9. Check and see if there are big air bubbles in the system. Write this down if so.
10. Check to see if the small Teflon tubes are connected to the flower heads.
11. Remove the tubes from the flower heads and put them in the plastic cups to drain.
12. Quit the program.
13. Take a copy of the previous night's data on a USB, marked with the previous date in the folder called 'Configs'.

Cleaning and filling the system

1. Open the PhenoSoft Control program on the Desktop.
2. Click on the 'Open' tab at the top and open the program 'Flush_dynspeed'.
3. On non-alcohol cleaning days, when the program is open, click on the start button for the 'clear' procedure. Once the procedure is done, take the syringe from the pump. Wash the syringe thoroughly at the sink and wipe it dry.
4. On alcohol-cleaning days proceed as follows: place the nectar tube in de-calcified water and run the 'Rinse' procedure; when the Rinse procedure is finished, place the Nectar tube in 70% alcohol; run the 'Alcohol' procedure; when the Alcohol procedure is finished wash and rinse the syringe; replace the syringe in the pump and transfer the Nectar tube back into the de-calcified water; open the Nectar valve and fill the syringe – the liquid will be cloudy white; open the Waste valve, close the Nectar valve and empty the syringe; fill and empty the syringe this way until the water runs clear; replace the syringe in the pump and run the 'FinalFlush' procedure.
5. While the system is being cleaned, proceed with the steps in the soup kitchen.
6. Take the syringe back to the system. Place the Nectar tube in a reservoir of fresh nectar. Place the lid of the reservoir back on.
7. Click on the down arrow for the pump in the program so the pump moves without the syringe attached to it.
8. Click on the Nectar valve to open it. Fix the syringe to the tube and draw some nectar into the syringe. Push the plunger up and down and adjust the syringe so there's no air bubbles in the syringe and the nectar flows smoothly.

9. Stop the pump by clicking on the little x between the up and down arrows. Place the syringe in the pump, adjusting the plunger so it fits nicely.
10. Fix the screw so the syringe is held in place.
11. Close the Nectar valve.
12. Click on the start button on the 'fill' procedure.

Soup kitchen

1. After measuring how much nectar is in the reservoir, throw away the old nectar. Turn on the hot water, fill up the reservoir and let the water run a little so the reservoir is properly washed.
2. Turn off the hot water, rinse the reservoir with cold water and keep it to drain.
3. If there is no more fresh nectar in the fridge, prepare fresh nectar by mixing 86 grams of sugar with 400 mL of water from the sink. Measure the solution once the sugar has dissolved. The concentration should be 17% ± 0.2.
4. Adjust the water and sugar until the proper concentration is reached.
5. Pour approximately 250 mL of sugar solution into one of the clean reservoirs. Take it into the experiment room for the fill procedure.
6. Remove the Eppendorf tubes from the cages. Weigh them directly and write down the weights for each of the cages that had tubes in them.
7. Clean the Eppendorfs that had extra food in them using the wire and some wet tissue. Rinse them out.
8. Throw away the water from the Eppendorfs that had water and refill them with fresh water from the sink.
9. Mix 1.2 grams of Nektar-Plus and 1.8 grams of Milasan. You will find both of these in the fridge. Add 9 grams of water to it so that the whole thing weighs 12 grams.
10. Use the pipette to fill the Eppendorf tubes for all the cages.
11. Weigh the Eppendorfs and write down the weights to enter into the Log.
12. Take the tubes to the cages and fit them to the proper place with the zip-ties. Do not open them yet.

Testing the program

1. Once the pump has finished refilling the system you can make test visits.
2. Quit the program. Start PhenoSoft Control again and this time open the config with today's date.
3. Make the test visits.
4. Once the test visits are made, open the Eppendorfs with the food for the cages that have them.
5. Open the Nectar valve. Hit the down arrow on the pump until the pump is fully refilled.
6. Close the Nectar valve. Quit the program.
7. Now start PhenoSoft Control again with the config with today's date. Do not touch the flowers after this!

Final check

1. Is the log filled up fully?
2. Are the cage doors all properly closed?
3. Are all the tubes inside the flower heads?
4. Are the food tubes opened in the cages that have them?
5. Is the proper config opened in the program?
6. Has the log been filled?