Measurement of O2 consumption for Drosophila

1. Turn on the oxygen monitor and then the computer and open the software OOIsensors.
2. To calibrate the oxygen sensor, log off and the log on the program. Create "new" and save in a designated folder. Turn on scan and pipette 25 mg/ml Sodium Diothionite into the chamber. Set cursor to the second peak around 600 nm wavelength. Calibrate oxygen/CO_2 using single temperature. Set concentration as 0% place cursor on intensity and select scan standard to obtain the reading. Remove solution and wash the chamber with distilled water many times. After washing, remove stir bar from chamber. Dry chamber. Set next concentration as 2000 (20% x 100), assuming ambient oxygen concentration is 20%. Select curve fit to obtain data curve and "update channel calibration".
3. Seal the ventilation hole with a piece of parafilm.
4. Put dry ice in an ice-bucket and place several layers of paper towel on top of the dry ice.
5. Anesthetize the drosophila by dumping some of them on top of the paper towels.
6. Quickly pick 4 flies of the same gender by using a soft brush and put them one by one into the oxygen monitor chamber.
7. Let the flies recover from the anesthesia. Make sure that none of them are injured. Replace the dead or injured with healthy ones.
8. After about 10 min, cover the chamber with a piece of parafilm and seal it by pushing the stopper. Record the oxygen consumption for exactly 5 min.
9. Open the chamber and blow fresh air into the chamber. Make sure the flies do not get away (the trick is not let them get to the top of the chamber. Once they get to the top, they fly away).
10. Once the oxygen level return to the ambient level, repeat step 8.
11. Repeat the measurement 3 more times.
12. After finishing the measurements, save the Timechart.
13. Open the Timechart by using slope calculation software. Measure each of the oxygen consumption slop and calculate the average slop for the 5 repeats.