Measuring Dead Space in the Manifold

In the FRC test, the Pressure and Volume changes caused by the animal's attempts to breathe help us derive the entire volume occluded from atmosphere. This volume is made up of the animal's lung volume at the time of the occlusion plus the dead space. So, if the animal is occluded at FRC, then after we determine the occluded volume, we only need to subtract out the dead space to compute FRC.

The following procedure describes how to measure the dead space. Keep in mind that you should only need to measure this if it is the first time you are using this apparatus or if you change the type of tracheal tube you are using (the dead space of your apparatus should be constant unless you change your apparatus). Once you have measured the dead space, this value must be entered into your hardware configuration in FinePointe Control Panel.

Boyle's Law Method

Before you perform the measurement, make sure the mouth pressure transducer is properly calibrated.

You will need:

- A tracheal catheter with the end sealed (airtight)
- A small syringe, perhaps 100µL or 50µL for a rat chamber. You should plan on injecting 20µL into the valve assembly.
- The current atmospheric pressure P_{atm}, in cm H_2O
- FinePointe open acquiring data so that you can view the mouth pressure signal

Procedure:

1. Attach your sealed tracheal catheter to the tracheal port, on the inside of the chamber. Make a guess at the dead space and set your syringe to roughly 0.02 or 0.03 times that amount. For example, if you guess that there is 0.85 mL of dead space, set the syringe for 20µL.
2. Attach the syringe onto the N_2 sample port.
3. Turn on the FRC valve (to close it).
4. Freeze the waveform in FinePointe Station and use on the cursor to read back the stable pressure. Check it is at zero. If not, make note of the current value. Return the trace to Live.
5. Inject the syringe. The Mouth pressure signal should rise, and if there are no leaks, it will remain steady at that level. If there is a leak, please use the suggestions in the chapter "Test for leaks in the valve assembly".
6. Freeze the waveform in FinePointe and use on the cursor to read back the stable pressure. Find the difference from that stable value to the pressure before you injected the syringe. Use this difference as ***ΔP*** in the following equation.

$$Deadspace = \frac{V_{injected} \times P_{atm}}{\Delta P}$$

   P_{atm} and ΔP must be in the same unit (cmH2O).

   The computed ***Dead space*** is in the same units that $\boldsymbol{V_{injected}}$ is specified.

Note the following to ensure a good measurement:

- Make sure the pressure signal did not get saturated. If it did, then you should repeat this process with a smaller volume. If you do, make sure your $\boldsymbol{V_{injected}}$ reflects what you injected.
- Your ***ΔP*** should be more than 10cm H_2O. If not, try increasing the volume you inject. If you do, make

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sure your $\boldsymbol{V_{injected}}$ reflects what you injected.

- Be sure to specify $\boldsymbol{P_{atm}}$ in cm H_2O units—it should be somewhere near 1030 cm H_2O

Alternate Boyle's Law Method Without P_{atm}

If $\boldsymbol{P_{atm}}$ is not known, you can use this method to measure the dead space. This method requires you to make two injections, and to find two $\boldsymbol{\Delta P}$ values. The two injections that you make must differ by a known volume (referred to here as $\boldsymbol{V_b}$).

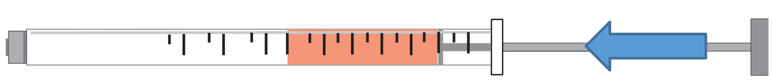
Ideally, you would want to make that known volume about ¼ the volume of the estimated dead space. And the volume you will inject ($\boldsymbol{V_{injected}}$) could be 10 times smaller. It is desirable to use the same syringe to both provide the $\boldsymbol{V_b}$ and to allow you to inject $\boldsymbol{V_{injected}}$.

So, if you estimate the dead space is 0.85ml, then you might want $\boldsymbol{V_b}$ to be around 200µL and you will want to inject about 20µL. This will require you to use a 250µL syringe, and so the accuracy of the 20µL injection is diminished from what it would be if you used a 50µL syringe. In short, you need to make a tradeoff between accuracy of the $\boldsymbol{V_{injected}}$ and the accuracy of $\boldsymbol{V_b}$.

To improve the situation, instead of injecting only 20µL, inject 50µL. This will improve your ability to have accurate values for both $\boldsymbol{V_{injected}}$ and $\boldsymbol{V_b}$. However, it requires that you recalibrate the mouth pressure to be less sensitive, to avoid saturation.

Procedure:

1. Calibrate Mouth Pressure so that you get an Effective Range of +/- 90 cmH2O. You will have to reduce the gain on the **M Pressure** preamplifier.
2. Attach your sealed tracheal catheter to the tracheal port, on the inside of the chamber.
3. Turn off the FRC valve (to open it).
4. Attach the syringe onto the N_2 sample port.
5. Pull the plunger of the syringe back 50µL (or $\boldsymbol{V_{injected}}$)
6. Turn on the FRC valve (to close it).
7. Freeze the waveform in FinePointe Station and use on the cursor to read back the stable pressure. Check it is at zero. If not, make note of the current value. Return the trace to Live.
8. Inject the syringe. The Mouth pressure signal should rise, and if there are no leaks, it will remain steady at that level. If there is a leak, please use the suggestions in the chapter "Test for leaks in the valve assembly".
9. Freeze the waveform in FinePointe Station and use on the cursor to read back the stable pressure. Find the difference from that stable value to the pressure before you injected the syringe. Use this difference as $\boldsymbol{\Delta P_0}$ in the equation below.
10. Turn off the FRC valve (to open it).
11. Pull the plunger of the syringe back 250µL (or $\boldsymbol{V_{injected}}$ + $\boldsymbol{V_b}$)
12. Turn on the FRC valve (to close it).
13. Freeze the waveform in FinePointe Station and use on the cursor to read back the stable pressure. Check it is at zero. If not, make note of the current value. Return the trace to Live.
14. Inject the syringe 50µL ($\boldsymbol{V_{injected}}$) to 200µL ($\boldsymbol{V_b}$) plunger position. The Mouth pressure signal should rise, and if there are no leaks, it will remain steady at that level.



15. Freeze the waveform in FinePointe Station and use on the cursor to read back the stable pressure.

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Find the difference from that stable value to the pressure before you injected the syringe. Use this difference as ***ΔP₁*** in the equation below.

$$Deadspace = \frac{\Delta P_1 \times V_b}{\Delta P_0 - \Delta P_1}$$

Pressure Conversions and Useful Information

Conversion	Multiply by this value
Inches Mercury to cm H2O	34.53
Torr to cm H2O	1.3595
mm of Mercury to cm H2O	1.3595
Pascal to cm H2O	0.0102
Kilopascal to cm H2O	10.197
mb to cm H2O	1.0197

Sizes	
ID of 14 Gauge Sheath (Rat)	1.524 mm
ID of PE260 Tubing (Rat)	1.778 mm
ID of 19 Gauge Stainless Tubing	0.889 mm
ID of 18 Gauge Stainless Tubing	1.0668 mm
ID of 17 Gauge Stainless Tubing	1.27 mm

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