PHYSIOTEL DIGITAL L11R RESPIRATION CHANNEL CALIBRATION INSTRUCTIONS USING HANS RUDOLPH PNEUMOTACH AMPLIFIER CONNECTED TO ABDC AMPLIFIER

Calibration of the PhysioTel Digital L11R versus a Pneumotach requires acquisition of both respiratory signals in a synchronized manner. It also requires calibration of the pneumotach signal versus a reference. This technical note will document the steps required for acquiring signals simultaneously and provide a process for calibrating the pneumotach and the implant.

BACKGROUND

The respiratory impedance channel of the PhyioTel Digital L11R is intended to be calibrated after the animal has fully recovered from surgical implantation of the device. When not calibrated, the raw impedance signal from the L11R implant can be used to calculate respiration rate and look at relative respiratory volume changes from baseline. If more precise data is desired to measure and calculate respiratory volume and volume changes, the raw impedance signal must be calibrated to a source of known flow rate and volume.

EQUIPMENT NEEDED

Configuration of the PhysioTel Digital L11R implant requires a desktop computer with Ponemah 5.32 or greater installed. This technical note does not cover details of PhysioTel Digital hardware and implant configuration. For additional information on this please see the Ponemah manual or PTD configuration page on the DSI Support Center. The following additional equipment is needed to perform device calibration.

DSI P/N	Description
600-2029-001 or 600-2030-001	Respiratory Mask (Dog/Pig) or Respiratory Helmet (NHP) (with spare neck seals as p/n 600-2031-001)
600-130x-00x	Pneumotach of proper flow range for your species (See Flow Range Determination)
276-0944-001	Hans Rudolf Pneumotach Amplifier
PNM-P3P-7002XS or PNM-P3P-7002	ACQ7700-USB (2 Slot) Acquisition Interface Kit or ACQ7700-USB (6 Slot) Acquisition Interface Kit
13-7715-04	ABDC 4 amplifier for ACQ7700
271-0401-001*	Quad04 Sync Counter board for ACQ 7700 Interface - *Spare PCI slot is needed in PC
274-0030-002	Sync Cable for synchronizing ACQ 7700 Interface with Sync board
600-1260-001	Mass Flowmeter for calibration verification
601-2200-001 or 601-2200-002	DSI Bias flow 200V (Optional – not needed if another constant flow source is available) or DSI Bias flow 100V (Optional – not needed if another constant flow source is available)

Flow Range Determination

Proper pneumotach selection depends upon what species and size of animal you are using and whether or not a heating unit is required. Please reference the below table.

Animal Size	Flow Range (L/min)	Pneumotach Model with heater	Pneumotach Model without heater
2-5 kg	0 – 35	600-1303-001	600-1303-002
5-20 kg	0 – 160	600-1305-001	600-1305-002

CALIBRATION CONSIDERATIONS

Calibrating the L11R is a 2-part process.

- Part 1: Calibrate Pneumotach to known flow source.
- Part 2: Calibrate L11R to Pneumotach

No need to have the animal nearby when calibrating the Pneumotach in 1st calibration step.
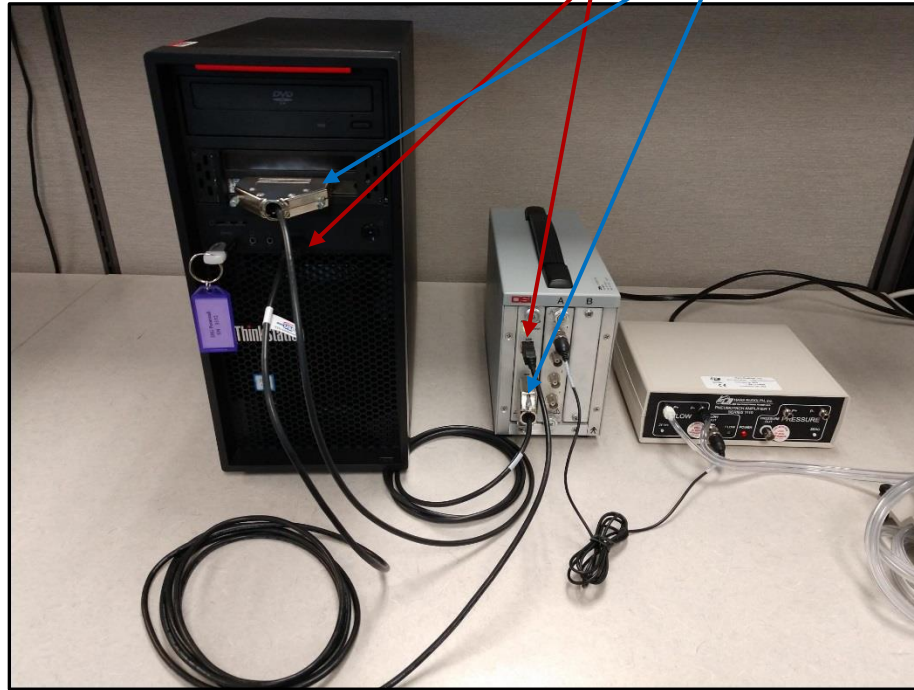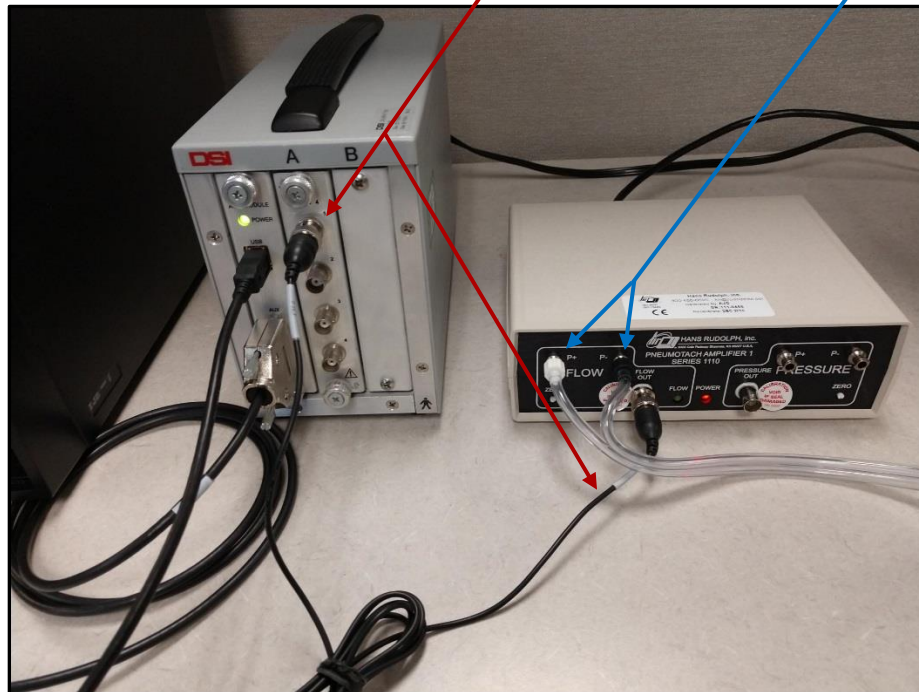
CONNECTING HARDWARE
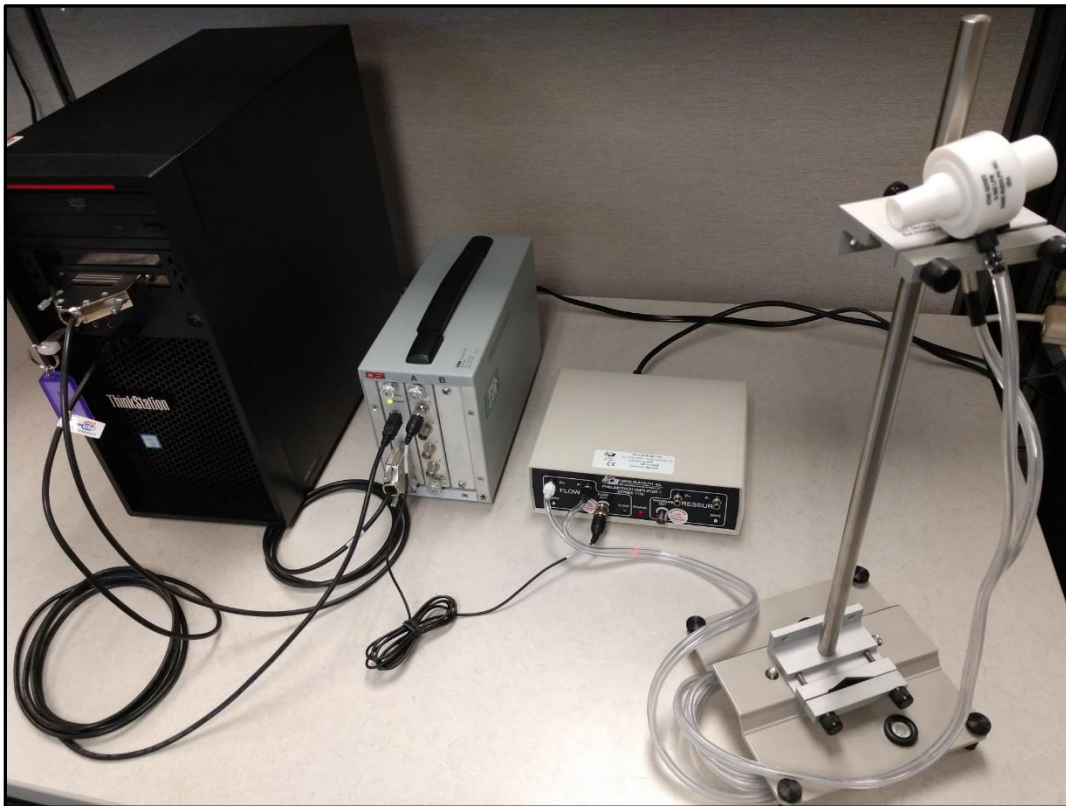
Connect all necessary equipment.

- Plug the ACQ 7700 and the Hans Rudolph Pneumotach Amplifier into a power source.

- Connect the Sync cable from the ACQ7700 to the Sync Card connector on the PC.
- Connect the ACQ 7700 USB cable to an open USB port on the PC.

- Connect the Pneumotach tubing to the Hans Rudolph Pneumotach Amplifier on the Flow side.
- Connect the analog Flow Out to the first available connection on the ACQ 7700 ABDC amp.


- Make sure all hardware is connected and powered on before launching Ponemah software.

PART 1: CALIBRATING THE PNEUMOTACH

In the first part of calibration, the pneumotach must be calibrated to a source of known, constant flow. This can be a DSI Bias Flow unit or a compressed air tank with a regulator that can be controlled.

A two-point calibration is performed consisting of a low and a high value. The typical low calibration reference is no (zero) flow. The typical high calibration reference is a constant positive flow from an air tank, or a constant negative flow (vacuum) from a bias flow supply. As with any calibration one must take care to assure that the calibration covers the expected range of use during application.

To verify high flow rate, connect the Mass Flowmeter to power and to the flow source.
The Flowmeter is directional: Ensure the flow is in the correct direction when connecting the Flowmeter.
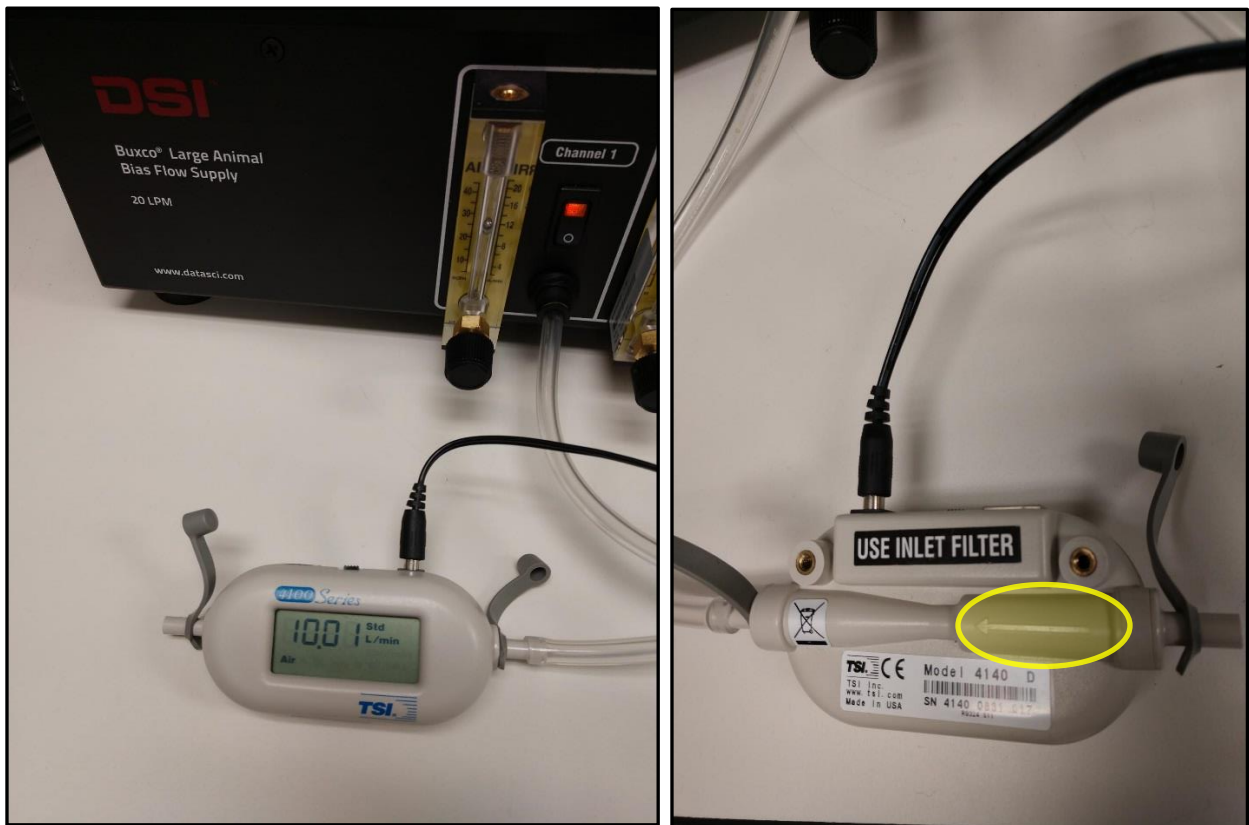
Open the Ponemah software and under the Options – Application Configuration menu make sure the acquisition interface is set to **ACQ7700 USB – PhysioTel Digital**.
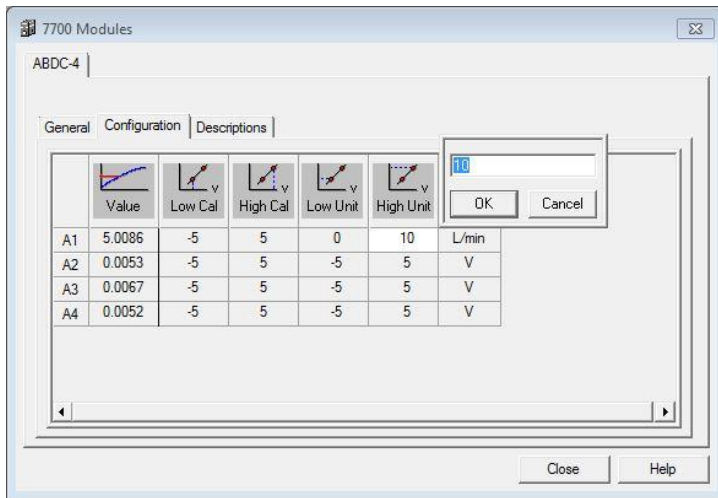
1. Go to the Hardware – 7700 Amplifier Setup menu.
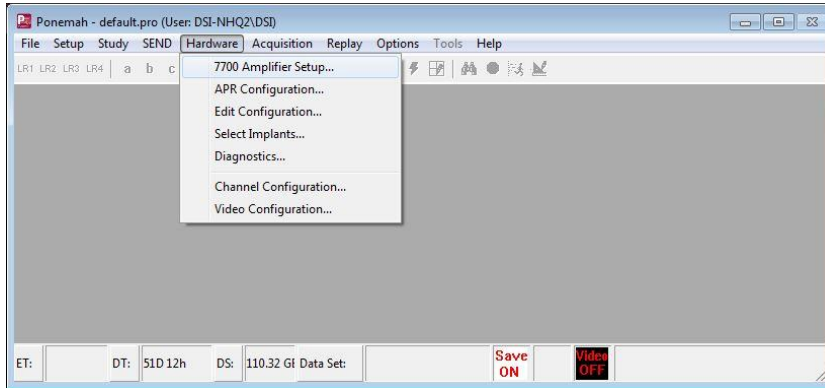
2. In the ABDC-4 Amplifier Tab, select the Configuration sub-tab.
   *Note: Double click in a box to modify a value.*
   a. In the example, a low flow of 0 L/min and a high flow of 10 L/min will be used.
   b. For channel A1, set the Low Unit to a value of 0 and the High Unit to a value of 10.

c. Set the Units to L/min. If flow is measured in mL/sec, set the Units to mL/sec.
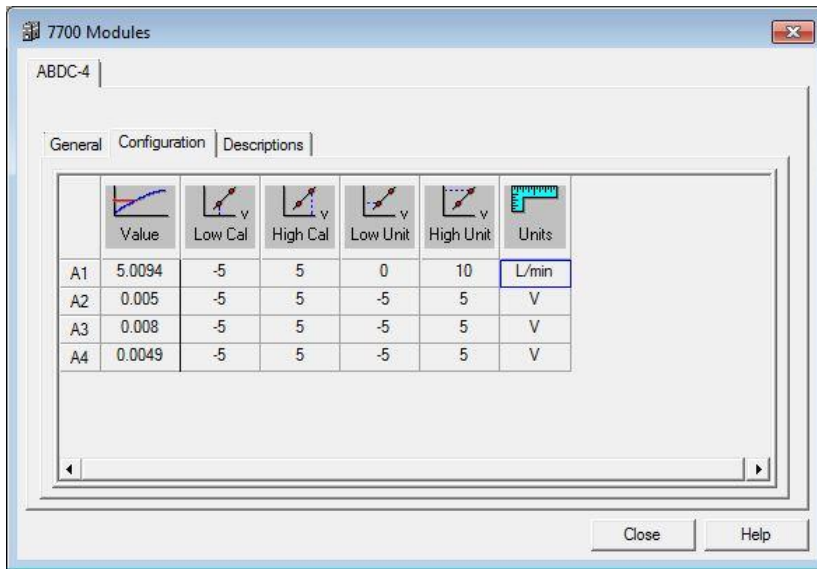
3. **Low Cal** - This will be the voltage that corresponds with the Low Unit. Have the pneumotach open to air (no flow). Double click the Low Cal box and make sure measurement is set to Average. The measurement time is recommended at 5 seconds. Select 'Measure' - **DO NOT select 'OK'**. Once the 5 second measurement completes the window will close and the average voltage will be entered automatically.
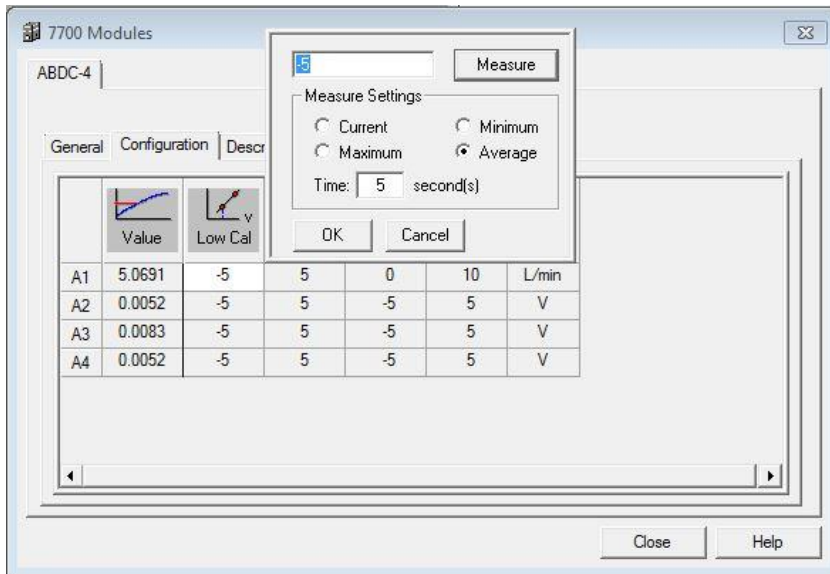
4. **High Cal** – This will be the voltage that corresponds with the High Unit. Connect the pneumotach to the flow source and make sure air is flowing at the known rate. Double click the High Cal box and make sure measurement is set to Average. The measurement time is recommended at 5 seconds. Select 'Measure' - **DO NOT select 'OK'**. Once the 5 second measurement completes the window will close and voltage will be entered automatically.
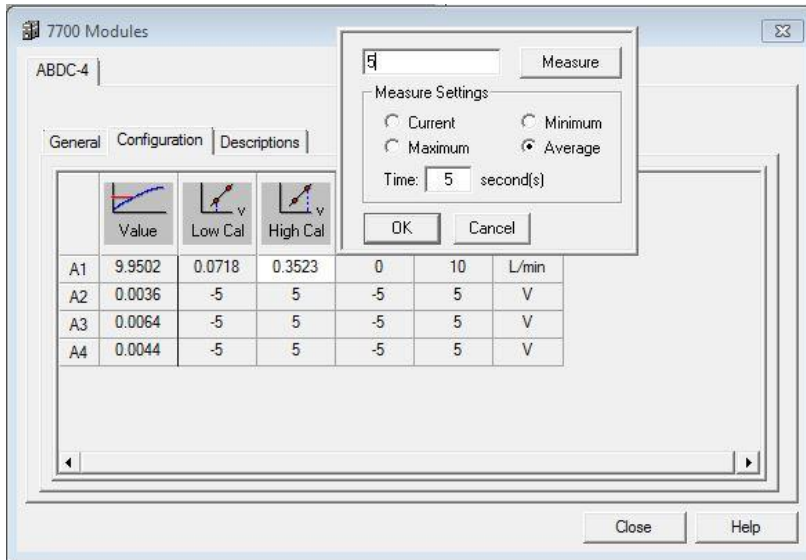
5. **Value** – verify calibration. Once calibration is completed it can be verified by turning the bias flow source on/off and viewing the calibrated signal value in the Value column. With bias flow off the value should be close to 0, and with the bias flow on the value should be at the high flow rate (in the example it is 10L/min). Calibration of the pneumotach is complete – select Close to close the window.
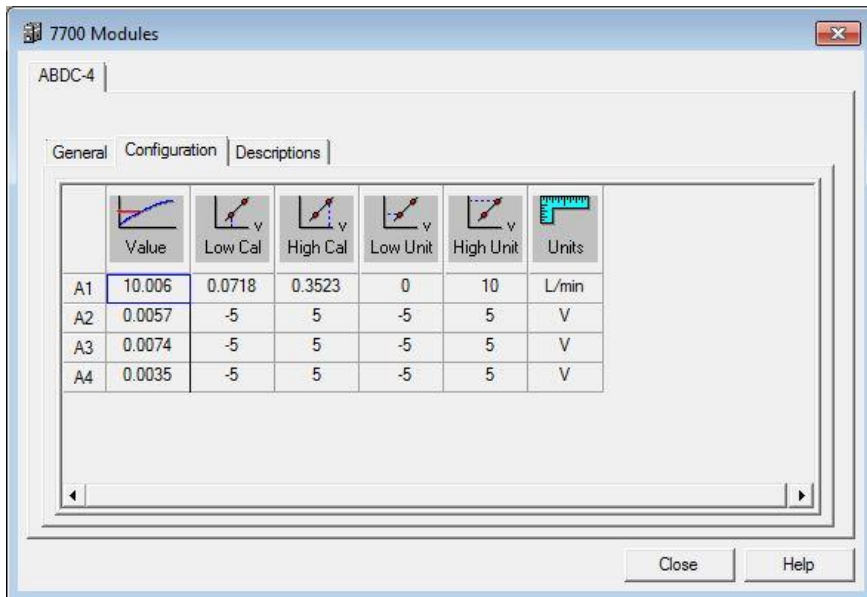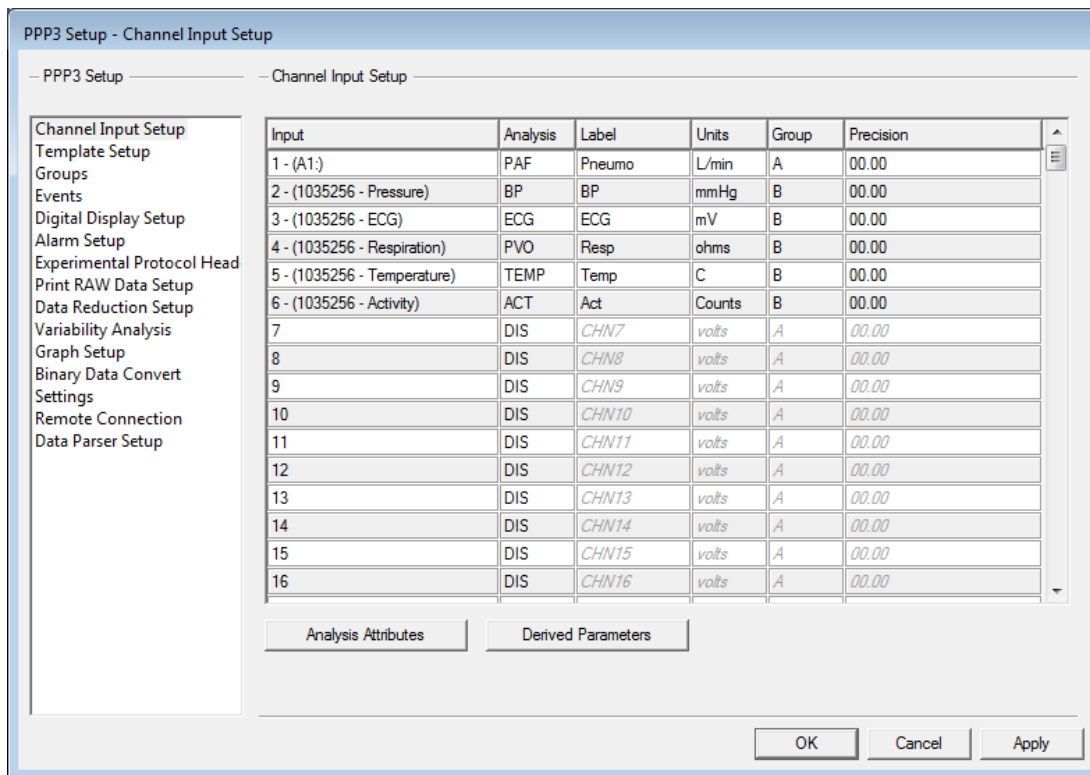

6. In the main Ponemah window select File – Save As to save the protocol. This will allow naming of the protocol so it can be easily identified later on if needed. It can be helpful to use the word 'calibration'.

PART 2: CALIBRATING THE L11R

In the second part of calibration the L11R implant is sampled simultaneously with the pneumotach attached to a mask (dog) or helmet (primate). The two signals are then compared in the Ponemah software to complete calibration. Calibrating the L11R impedance channel versus a pneumotach assumes the user has followed part 1 of the calibration technical note to set up and calibrate the pneumotach and configured the Ponemah software in such a way that it may be used in a synchronized manner with the L11R implant (using a pneumotach interface connected to ACQ7700USB, synchronized to the PhysioTel Digital CLC).

Ensure the desired L11R implant is turned on and configured. For additional information on PhysioTel Digital implant configuration please see the Ponemah manual or PTD configuration page on the DSI Support Center.
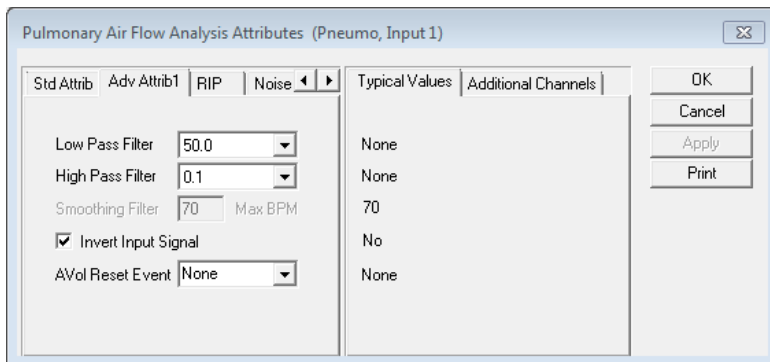
1. Configure the channels in Channel Input Setup.
   a. In Ponemah go to Setup – P3 Setup and select Channel Input Setup.
   b. Set the pneumotach channel to analysis type of PAF. In the example this is channel 1 with a label of A1.
   c. Set the L11R respiratory channel to analysis type of PVO. In the example this is channel 4.
   d. Label other channels as indicated and disable unused channels.
   e. Set the Pneumotach channel to a different group than the implant channels. In the example pneumotach is Group A while the implant is Group B.


PPP3 Setup - Channel Input Setup

PPP3 Setup

Channel Input Setup
Template Setup
Groups
Events
Digital Display Setup
Alarm Setup
Experimental Protocol Head
Print RAW Data Setup
Data Reduction Setup
Variability Analysis
Graph Setup
Binary Data Convert
Settings
Remote Connection
Data Parser Setup

Input	Analysis	Label	Units	Group	Precision
1 - (A1:)	PAF	Pneumo	L/min	A	00.00
2 - (1035256 - Pressure)	BP	BP	mmHg	B	00.00
3 - (1035256 - ECG)	ECG	ECG	mV	B	00.00
4 - (1035256 - Respiration)	PVO	Resp	ohms	B	00.00
5 - (1035256 - Temperature)	TEMP	Temp	C	B	00.00
6 - (1035256 - Activity)	ACT	Act	Counts	B	00.00
7	DIS	CHN7	volts	A	00.00
8	DIS	CHN8	volts	A	00.00
9	DIS	CHN9	volts	A	00.00
10	DIS	CHN10	volts	A	00.00
11	DIS	CHN11	volts	A	00.00
12	DIS	CHN12	volts	A	00.00
13	DIS	CHN13	volts	A	00.00
14	DIS	CHN14	volts	A	00.00
15	DIS	CHN15	volts	A	00.00
16	DIS	CHN16	volts	A	00.00

Analysis Attributes   Derived Parameters

OK   Cancel   Apply

2. Check PAF analysis settings.
    a. Highlight the pneumotach channel set as PAF and select 'Analysis Attributes' button on bottom of page.
    b. In the Std Attrib tab ensure that the Primary Signal is set to Flow and that the Input Flow Units are set to the unit that was used during the pneumotach calibration in part 1.
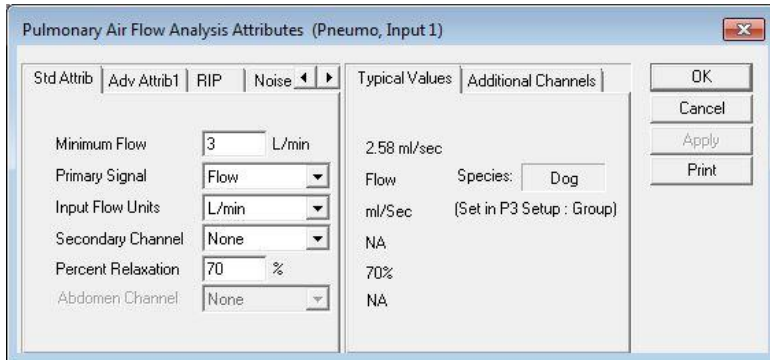

    c. In the Adv Attrib1 tab there are no filters applied by default. A low pass and high pass filter may be applied to help filter noise. See the Appendix for pneumotach filtering recommendations.
   d. Select OK to close.

3. Check PVO analysis settings.
    a. In the Channel Input Setup, highlight the L11R impedance channel set as PVO and select 'Analysis Attributes' button on bottom of page.
    b. In the Std Attrib tab ensure that the Calculated Flow Units is set to the unit that was used during the pneumotach calibration in part 1.
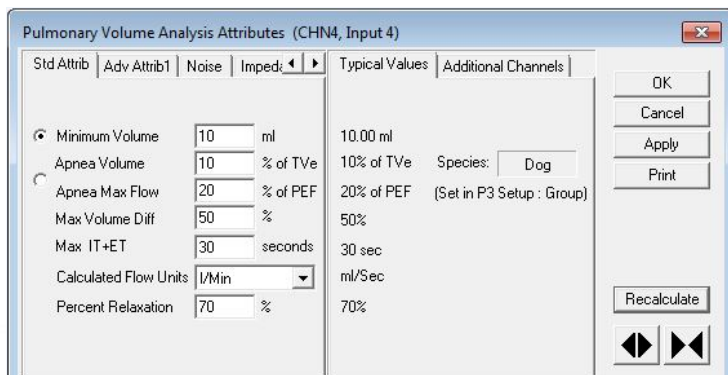
c. In the Adv Attrib1 tab filters may be applied to help filter noise. See the Appendix for L11R implant filtering recommendations.

d. Select OK to close.
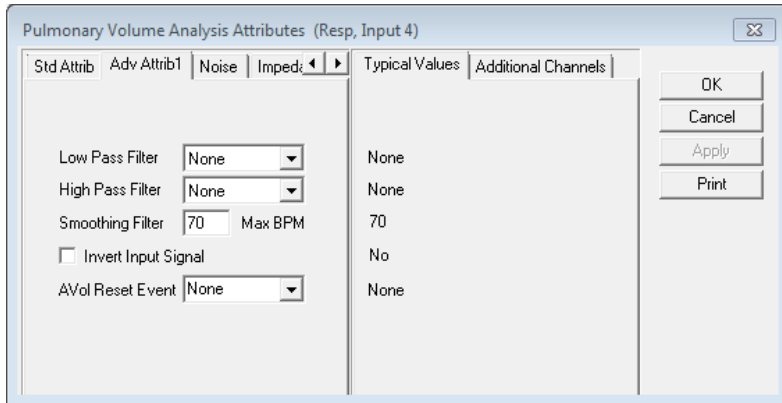
4. In the P3 Setup Menu select Graph Setup to configure graph pages.

a. Enable a graph page for the respiratory channels and set up with desired signal presentation. In the example both the pneumotach channel and impedance channel are shown with a flow and a volume presentation.

b. Once graph page is configured, select OK to close the P3 Setup.
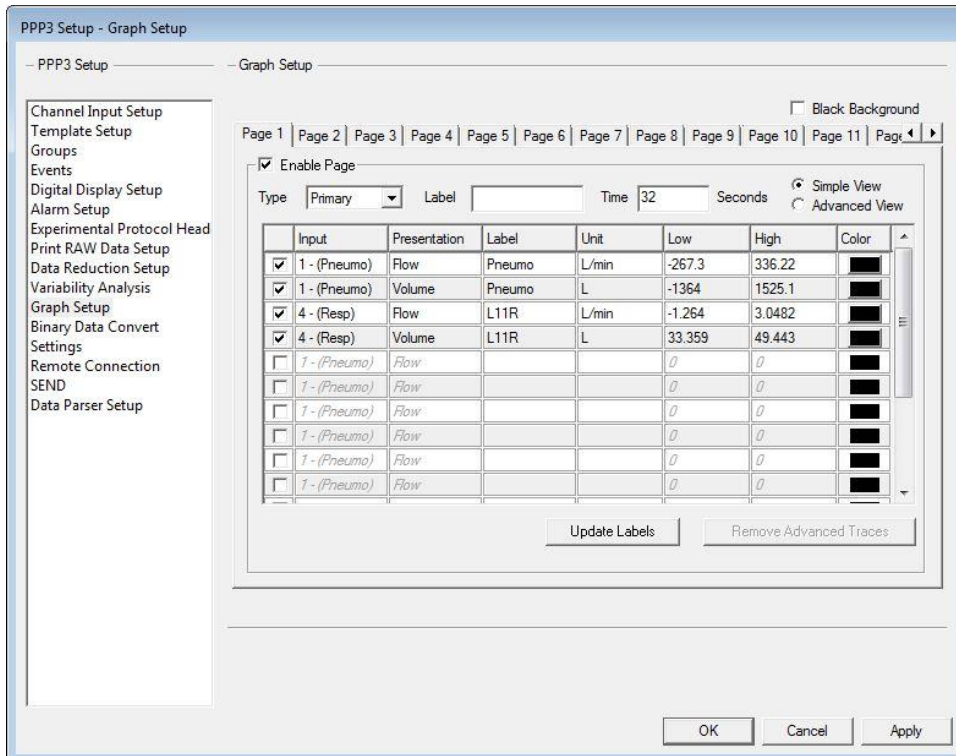
5. From the main Ponemah menu select File – Save to save recent changes to the Protocol.

6. Verify sample rates.
   a. From the main Ponemah window select Acquisition – A/D Sample Rate
   b. Set the Primary sample rate to either 500Hz or 1000Hz. Set the Secondary rate to 50Hz.
   c. Click in the boxes under Rate to set as Primary or Secondary for each channel as indicated.
   d. Click OK to close the window. Save the protocol changes using File – Save.

7. Begin acquisition.
   a. Select the Acquisition menu and choose Data Set Name. This will allow naming of the data file so it can be easily identified later on if needed. It can be helpful to use the subject name and the word 'calibration'.
   b. Select the Acquisition menu and choose Start Acquisition to begin the data collection.

8. Ponemah will begin data collection and the graph page will begin to display incoming data.
   a. The data collection for the calibration period may be collected for as long or short as desired; however, for calibration purposes a maximum data segment of 5 consecutive minutes may be used.
   b. It is strongly recommended to only calibrate during periods when the impedance signal baseline is consistent and bad data marks are not present.
   c. Prior to calibration it is recommended to acclimate the animals to the mask (dog) or helmet (primate) so that they are relaxed during the data collection. The data will preferably be with the animal in a posture representative of the posture which they will be in during a normal data collection.

9. Put on the mask (for dog) or helmet (for primate) and wait until the animal is relaxed and the data being collected is stable and relatively clean. Ideally, Ponemah will be identifying and triggering cycles for both the pneumotach channel and the impedance channel as in the example.
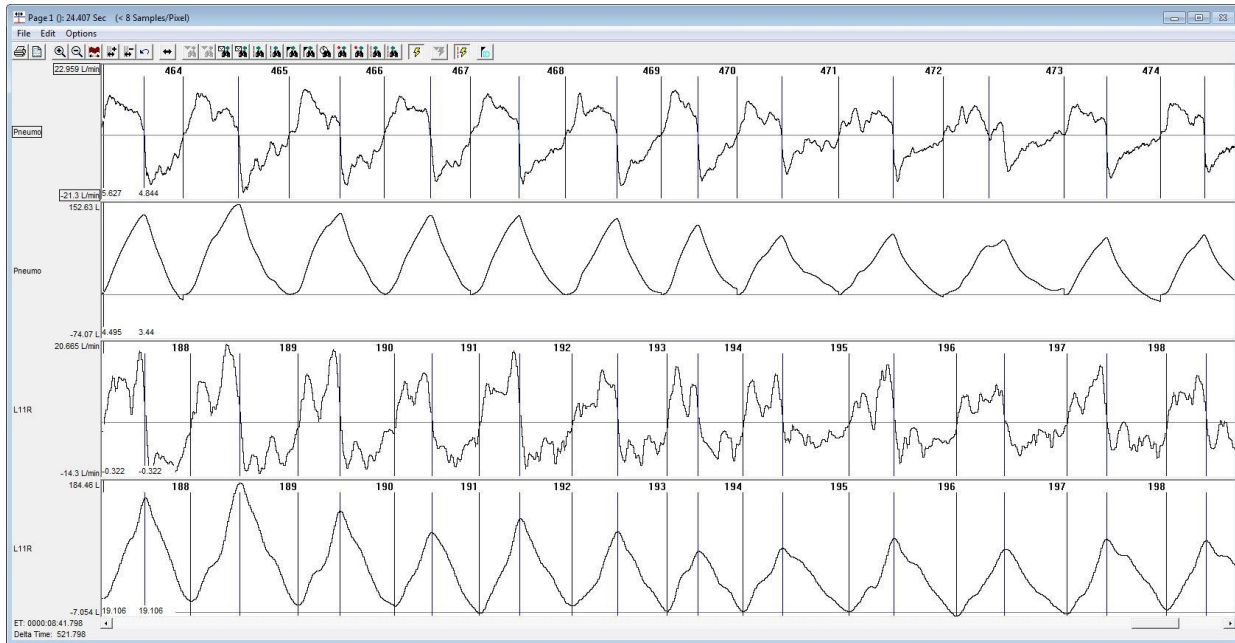

10. Data collection can be run as long as desired, but should have **AT LEAST 50** good breaths within a 5 minute segment. When enough data has been collected, select Functions – Stop Acquisition.
11. Calculate and apply calibrations in Review Mode.
    a. From main Ponemah window select File – Open Review File and select the calibration file. Load all channels.
    b. In the graph page right click on PAF channel (pneumotach) and select Analyze. Verify cycles are triggering. If not, adjust settings as needed in Std Attrib.
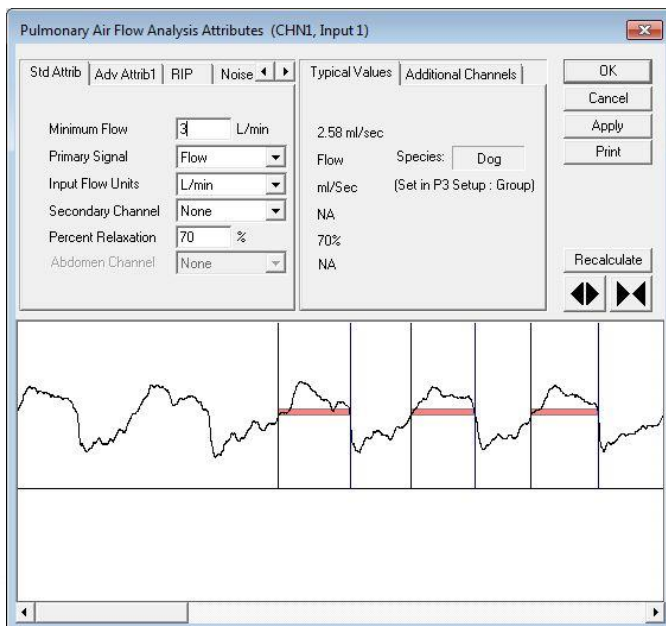
      c.   Observe the main graph page and verify that pneumotach and impedance signal are in sync and inspiration correlates with a positive shift in signal. If inspiration is not positive and the two signals are out of phase with each other, select the Adv Attrib1 tab and choose the Invert Input Signal box.

      d.   Select OK and reanalyze the entire channel.
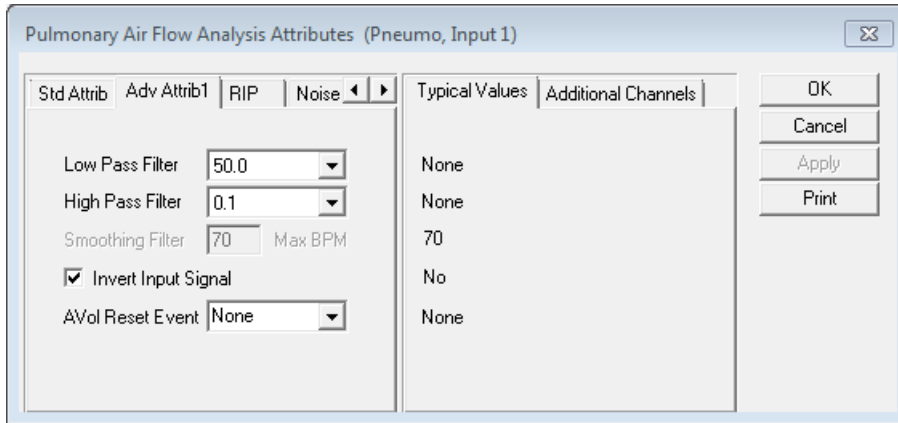
      e.   In the graph page right click on PVO channel (impedance) and select Analyze. Verify cycles are triggering. If not, adjust settings as needed in Std Attrib.

      f.   Select OK and reanalyze the entire channel.
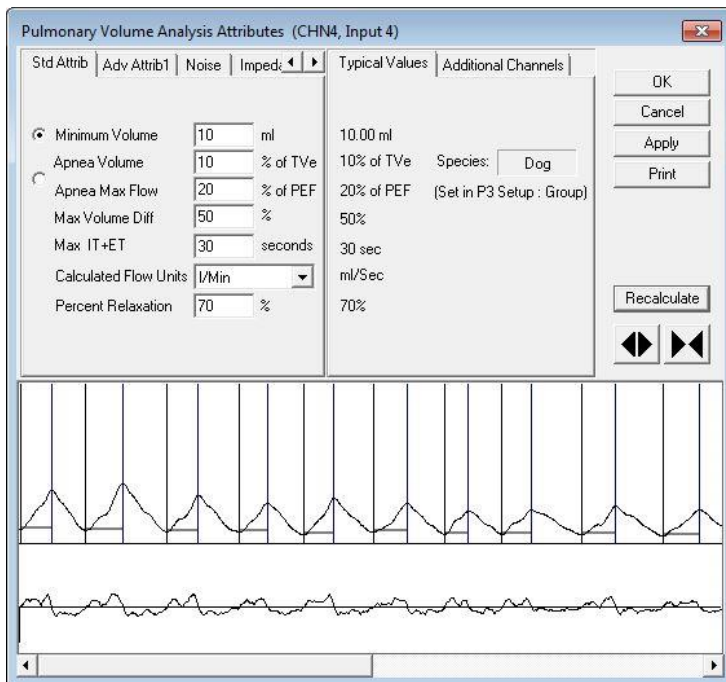
g. Go to Impedance tab in the PVO module and select the pneumotach channel for calibration in the drop down menu.

h. Select the Calibration button.
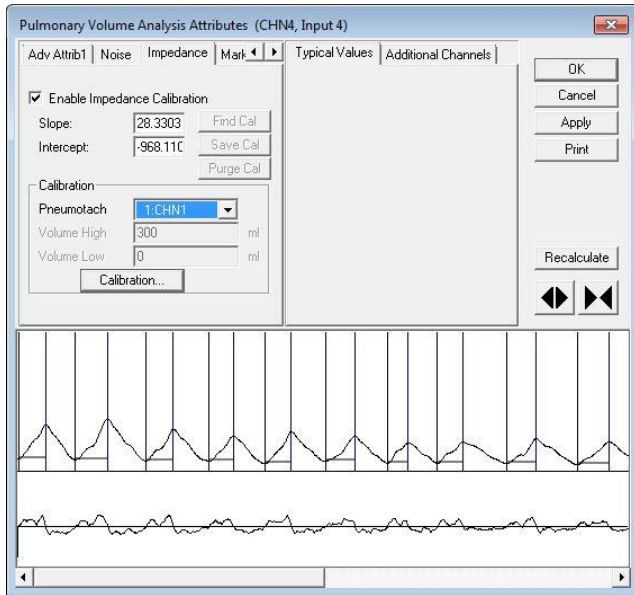
Note: The Fixed Volume entry is disabled when using the Pneumotach option.

i. The Impedance Calibration Dialog will display up to a five-minute segment of data from the user selected area of normal waveforms. The green bar located near the top of the dialog is the Calibration Segment Bar. The Calibration Segment Bar allows the user to select which breaths to calibrate versus the pneumotach signal. The bar can be shortened or elongated using the computer mouse. Multiple segments can be added to maximize the number of typical breaths used in the calibration. Additional segments are added by right clicking in the Calibration Segment Bar area.
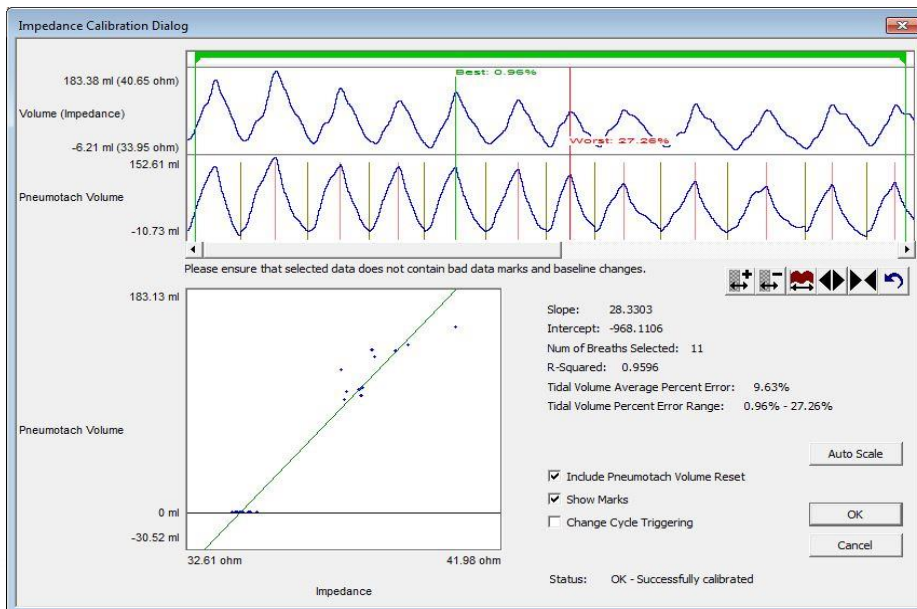
    j.   After placing the Calibration Segment Bar(s) the slope and intercept will automatically be calculated. Also displayed are the Number of Breaths Selected (determined by the placement of the Calibration Segment Bar(s)); Number of Breaths Used in the calibration; the Average Percent Error [(Vtotal - VPneumo)/VPneumo] of the breaths used in the calibration and the Percent Error Range.

    k.   It is best practice to find segments of data that are clean and that the worst % error is minimized.

    l.   Once the error is minimized and the calibrations are acceptable, select the OK button.

    m.   The calibration Slope and Intercept value from the Impedance Calibration dialog will be automatically entered.
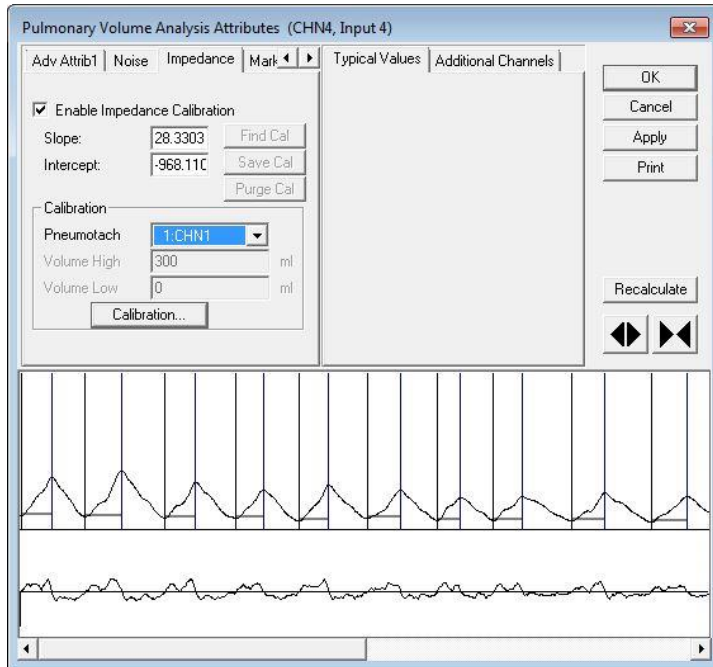
    n.   Select OK to reanalyze the entire dataset with the new calibration applied. Graph scaling and analysis settings will need to be updated after the calibration is applied.

    o.   The calibration will need to be completed for each L11R implant.

12. Apply the calculated calibration values to future recordings.

    a.   The calculated Slope and Intercept from the calibration session must be recorded along with any filtering applied during the calibration.

    b.   The recorded slope and intercept calibration values along with filtering options for each implant will need to be applied to the study protocol.

    c.   Calibration can be repeated as often as desired.

APPENDIX

Filtering Considerations

- Set the filtering by keeping in mind the respiration rate of the animal. Any filtering applied to the calibration sessions **MUST** also be consistently applied throughout the study. For example, if you use a 30 Hz low pass filter for the calibration session, a 30 Hz LPF must be applied on the study days the calibrations are used on.
- Apply either a low pass filter **OR** a smoothing filter, but not both.

Channel/Waveform	Minimum Sample rate to apply Low/High Pass filter	Low Pass Filter	High Pass Filter	Smoothing filters
Systemic Blood Pressure	≥100 Hz	≥30 Hz	None	N/A
Left Ventricular Pressure	≥500 Hz	None	None	N/A
ECG	≥500 Hz	None	< 0.5 Hz	N/A
Respiration - JET with PAF module	≥50 Hz	≥10 Hz	<1.5 Hz	≥20
Respiration – L11R with PVO	≥50 Hz	≥10 Hz	None	≥20
Respiration - Pneumotach	≥50 Hz	≥30 Hz	<1 Hz	N/A

Table 1: Recommended Sample Rates and Filters to Apply

DSI™