

# Olfactometer

206A

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#### Aurora Scientific

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#### CAUTION

Maximum operating pressure is 30 psig (207 kPa). Do not exceed the maximum pressure ratings. Injury could result from use at higher pressures. Damage to the 206A may occur at pressures above the maximum rated pressure.

# **Revision History**

Date	Version Number	Document Changes
20-Feb-2017	1.0	Initial Draft
3-Mar-2017	1.1	Added Chapter 8 Appendix - Serial Communication Commands
5-Apr-2017	1.2	Added Chapter 9 Appendix – Connections
18-Aug-2017	1.3	Added Sequence Trigger Function – updated Chapters 4.7, 8.1.8-8.1.10 and 9

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## 1 Introduction

## 1.1 Scope and Purpose

The 206A Olfactometer is an instrument designed to deliver a series of odor concentrations in a precisely controlled fashion. Careful fluid design of the instrument ensures that odors released have a well-defined "square" profile with minimal delay between activation of the valve and the arrival of the odor at the subject location. Odor releases are repeatable and precise. The basic instrument provides 4 scent vials which can be increased in sets of 4 to provide 4, 8 or 12 vials.

## 1.2 Process Overview

The 206A Olfactometer is a system comprised of several parts.

- 1. Mass Flow Controllers to precisely control the flow through all parts of the system.
- 2. Vials for odorant storage that can be individually addressed to release a variety of different odors or odors of varying concentrations.
- 3. Valve manifolds that control which vial is being used for the odor release. Valve manifolds are available in 4 vial modules that can be easily added to a system to expand the total number of vials from 4 to 8 or a maximum of 12.
- 4. A Mixing valve to combine flows.
- 5. A Final valve to direct the flow to the experiment or to the exhaust.
- 6. Control circuitry for the above system.
- 7. Control software for Windows.

# 2 Quick Start Guide

This quick start guide will provide you with basic information that will allow you to get your new 206A Olfactometer up and running as quickly as possible.

## 2.1 Instrument Setup

The 206A Olfactometer is shipped in 1 or more boxes depending on how many extra vial modules you have purchased. Unpack all boxes included with the shipment and check the contents against the list provided below. If there are any issues with what you received, please <u>contact Aurora Scientific</u> as soon as possible.

ltem	Description
206A Mass Flow Controller (MFC) Subassembly	MFC Subassembly consisting of 3 mass flow controllers, main controller circuit board, inlet manifold and installed fittings.
206A Odor Vial Subassembly	Odor vial subassembly consisting of 2 inline manifold valves and the holders for 4 odor vials. Note: the 206A Odor subassembly is shipped already attached to the 206A Mass Flow Controller Subassembly.
206A Mixing Valve Subassembly	Mixing valve subassembly consisting of the mixing valve mounted on a plate with installed tube fittings. Note: the 206A Mixing Valve subassembly is shipped already attached to the 206A Odor Vial subassembly.
206A Final Valve Subassembly	Final valve subassembly consisting of the final valve with installed tube fittings. Note: the 206A Final Valve subassembly is shipped separate from the Mass Flow Controller, Odor vial and Mixing valve subassemblies.
206A Accessory Subassembly	Accessories for 206A including: Teflon hose 1/16" OD, Teflon hose 1/8" OD, spare inert fittings for 1/16" tube, spare inert fittings for 1/8" tube, 4 vials with septa, septa puncture tool, 4mm supply tube, 4mm one-touch tube fitting for supply tube, power supply, power cord, USB cable, software disk, extension cable for connecting the Final Valve to the 206A.

Table 1 Items included with a 206A Olfactometer

The following is a list of several items required in order to use the 206A Olfactometer. These items are not included with the 206A.

Required Items	Description
Clean/Dry Air Source	A source of clean and dry air is required by the 206A. This air can be supplied by various means including: high pressure gas bottle, lab air supply or a compressor. When using either lab air or a compressor you may also require an air dryer, air filter and a charcoal filter to remove odors from the air.
Air Dryer, Air Filter, Charcoal Filter	If the air source is lab air or a compressor then moisture, particles and odors may need to be removed from the air prior to entering the 206A. The Mass Flow Controllers must have clean, dry air or permanent damage could result. This extra equipment is not required if using a high pressure gas bottle as the air source.
Upstream Air Regulator	The Mass Flow Controllers, valves, tubing and fittings that are used in the 206A all have maximum pressure ratings. A regulator must be installed upstream of the 206A to control the inlet pressure. Inlet pressure must be in the range 5-30 psig (35-207 kPa). DO NOT EXCEED THE MAXIMUM PRESSURE RATING OF 30 psig (207 kPa).
Animal Nose Cone or Other Odor Delivery Fitting	The outlet of the Final valve should be piped to an animal nose cone or other fitting designed to deliver the odor to the subject. The Final Valve uses ¼-28, 1/8" inert fittings on the ports. In most cases the nose cone should include a method of terminating a 1/8" OD (outside diameter) Teflon tube.
Mount for Final Valve	The Final valve includes tapped holes on the bottom of the valve (4-40UNC). The Final Valve should be mounted to a plate or other fixation device to keep it in place near the animal.
Mount for 206A Olfactometer	The 206A Olfactometer includes rubber feet on the bottom of each subassembly. The Olfactometer can be placed on a flat, level surface or it can be hung off the back of the test chamber by use of the included hooks designed into each part. A horizontally mounted length of 20mm x 20mm aluminum extrusion is ideal for mounting the 206A using the built-in hooks.
Exhaust Vent	The 206A Olfactometer continuously releases odor through the Final valve Exhaust port while in use. To prevent a buildup of odor in the test facility the exhaust from the Final valve should be piped to an exhaust vent.

Table 2 Required items not included with a 206A Olfactometer



NOTE: Maximum operating pressure is 30 psig (207 kPa). Do not exceed the maximum pressure ratings. Injury could result from use at higher pressures. Damage to the 206A will occur at pressures above the maximum rated pressure.

#### 2.1.1 Installation of the 206A Olfactometer

- The 206A Olfactometer is shipped assembled and should either be placed on a flat, level surface on the attached rubber feet or mounted using the included hooks on a horizontally mounted length of 20mm x 20mm aluminum T-Slotted extrusion. (McMaster-Carr part number 5537T101).
- 2. Install the included software on a Windows PC.
  - a) Insert the software install CD into the PC which should launch the installer.
  - b) Follow the on-screen wizard to complete the installation.
  - c) Following successful completion of the installation you should see a new icon on the desktop that can be used to launch the software.
- 3. Once the software is installed then connect the USB port on the MFC Controller circuit board to a USB port on the PC.
- 4. Pipe the air supply to the 206A Olfactometer. (For the purposes of this quick start guide we will assume that you are using a high pressure air cylinder as your air source and therefore there is no requirement for drying, filtering or removing odors from the air source.)
  - a) Attach the included 1/8" NPT to 4mm one-touch adapter to the outlet of a two stage regulator. Use Teflon tape to seal the 1/8" NPT threads prior to screwing the fitting into the regulator.
  - b) Attach the regulator to the gas cylinder.
  - c) Push one end of the supplied 4mm blue air tubing into the one-touch fitting on the regulator and the other end into the inlet manifold fitting on the MFC subassembly. Note: depending on the distance between the gas cylinder and the 206A you could cut the 4mm tubing to an appropriate length.
  - d) Attach the Mixing Valve outlet to the Final Valve "Scent In" inlet using the supplied 1/8" Teflon tubing and the supplied ¼-28 inert fittings.
  - e) Attach the Exhaust bulkhead fitting (located on the Mixing valve plate) to the Final Valve "Air In" inlet by inserting one end of the 1/8" Teflon tubing into the one-touch fitting and using a supplied ¼-28x1/8" port fitting to attach the tubing to the Final Valve.
  - f) Attach the Final Valve electrical cable to the Final Valve circuit board either directly or by using the valve extension cable supplied with the accessories.
  - g) Attach the Final Valve "Animal" outlet (normally closed) to the experiment using 1/8" Teflon tubing and the appropriate inert fittings supplied.
  - h) Attach the Final Valve "Exhaust" outlet (normally open) to the exhaust using 1/8" Teflon tubing and the appropriate inert fittings supplied.
- 5. Plug the power supply into the 206A Mass Flow Controller (MFC) circuit board and, using the supplied power cord, plug the AC adapter into an appropriate AC outlet.
- 6. Open the valve on the air cylinder and set the pressure on the regulator to 30 psig (207 kPa).
- 7. Check the tubing for air leaks.

#### 2.1.2 Using the 206A Olfactometer

1. Place a small amount of the chosen liquid odorant(s), with or without a solvent, in each of the vials to be used. Ensure that only a small amount of odorant is placed in the vial.

# NOTE: THE ODORANT LIQUID LEVEL IN THE VIAL MUST BE LOWER THAN THE SHORT TUBE IN THE SEPTUM OF THE VIAL LID.

This is to ensure that only gaseous odors are injected into the olfactometer. If the short tube is in the liquid, then liquid will be run through the olfactometer. This level of contamination may not be able to be cleaned out of the olfactometer and may necessitate replacing the valve manifold and associated tubing. Screw the vial onto the lid and place the vial in the circular cutout.

- 2. Launch the 206A Control Software by clicking on the launch icon located on the desktop.
- 3. Configure the vials.
  - a) Click on **Config>Configure System** to open the System Configuration window. Note: if Odorants are already configured and you want to replace them then first click on the Clear Vials button at the bottom left of the Vial table data entry box.
  - b) Click on line 1 and then click on the **Add New Odorant** button and type in the name of the odorant.
  - c) Click on the Add New Solvent button and type in the name of the solvent.
  - d) Enter the **Odorant Volume Concentration in Liquid** in the box shown. Note: this is the volumetric concentration of the odorant in the solvent. If you are not using a solvent then you should set the **Odorant Volume Concentration in Liquid** to 100%.
  - e) Click on the **Set Selected Vial** button and you should see the odorant, solvent and concentration appear in the Vial table above.
  - f) Follow the same procedure to enter all odorants required by the study. Note: the 206A will auto detect the number of vials connected to the olfactometer and then grey out all vial numbers not present.
  - g) Next enter the Odor Flow Stabilization Delay in seconds (this is the time that it takes an odor to flow from a vial to the Final Valve). This time is normally in the 5-60 second range depending on the system flowrate specified and the length of tubing between the Mixing Valve and the Final Valve. If you find that during the first few releases the concentration is changing then you probably have the Odor Flow Stabilization Delay set too short. If this is the case increase this time.
  - h) Set the **Total System Flow Rate** in sccm (standard cubic centimeters per minute). This is the sum of the odor flow plus the dilution flow. Note: the maximum flow is limited to 950 sccm.
  - i) Click on the **Save Config** button to save the current configuration to a file.
  - j) Click on the **OK** button to set the vials and return the user to the main screen.
- 4. There are two operating modes for the software, option 1 is a manual control mode and option 2 is a sequence control mode:
  - Option 1 Manual Mode
  - a) On the Home window click on the drop down arrow next to the **Select Odor** box.

- b) Click on the odor of interest.
- c) Set the Total Flow % through the Vial as a percentage of the Total System Flowrate. This is the main method of changing the concentration of the odorant delivered to the experiment. Note: the odor mass flow controller is limited to a maximum flowrate of 100 sccm and, for best accuracy, a minimum flowrate of 5 sccm. Therefore, the range of possible vial flowrates is dependent on the Total System Flowrate. For example, assuming a Total System Flowrate of 950 sccm the maximum vial flowrate percentage would be 10.5% (100sccm/950sccm = 10.5%). The minimum vial flowrate percentage would be 0.53% (5sccm/950sccm = 0.53%). [Note: lower minimum vial flowrate percentages are possible as the mass flow controller manufacturer specifies a minimum flowrate of 1 sccm. However, for best accuracy and repeatability we suggest you limit flowrate to a minimum of 5 sccm.]
- d) Set the **Odor Delivery Duration** in milliseconds. This is the time that the odor will be delivered to the animal. The minimum duration time should be greater than 20 milliseconds for best results.
- e) Click on the button labelled Flow Odor to Exhaust to start the flow of the selected vial odor at the selected concentration and flowrate. The Deliver Odor to Animal button will remain greyed out until the Odor Flow Stabilization Delay time has completed.
- f) Click on the **Deliver Odor to Animal** button to deliver the selected odor to the animal.
- g) If at any time you want to see a diagram of which valves are active, then click on Config>Advanced Status and Control. The System Status window will open which will graphically show the flow path and which valves are on or off. The flowrates reported from the three mass flow controllers are also shown in this window.
- h) Click on the **Purge** button to purge the tubing of odor when the experiment is complete.
- i) To shut off the flow completely click on **Stop Flow** button.
- Option 2 Sequence Mode
- a) On the Home window click on the **Config** menu item and then click on **Sequencer**. Sequences are a method of automating an experiment.
- b) A sequence can be entered manually or the software can create a random sequence.
- c) Use the controls to enter a sequence.
- d) Click the **Run** button to execute the sequence.
- e) Click on the **Purge** button to purge the tubing of odor when the experiment is complete.
- f) To shut off the flow completely click on the **Stop Flow** button.

#### 2.1.3 Cleaning and Shutdown

- 1. Click on the **Purge** button to run clean air through all of the tubing. We suggest a minimum of 20 minutes of purge time to ensure odors are cleaned out of the olfactometer.
- 2. To perform a more complete cleaning do the following:
  - a) Place the "Animal" outlet and "Exhaust" outlet of the Final Valve in a beaker for collection of liquid.

- b) Remove all odor vials and replace them with vials filled with a cleaning solvent for the odors being used. In this case make sure the liquid level reaches to the top of the vial.
- c) Run the control software and select the vial that you want to clean. Once the valve opens liquid solvent will be run through the valves and the tubing. Allow the solvent to run until the solvent liquid level falls below the level of the outlet tube (short tube) in the vial.
- d) Shut off the flow (Stop Flow).
- e) Remove the vial and replace it with a clean one.
- f) Use the software to select the vial of interest again and then run clean air through the system for 30 minutes or until the valves and tubing are dry and all traces of odor are gone.
- 3. To shut down the 206A do the following:
  - a) Click on **Stop Flow**.
  - b) Turn off the air source, close the valve on the high pressure air cylinder.
  - c) Shut off the software by clicking on the **X** to close it or click on **File>Exit**.
  - d) Power off the 206A by unplugging the DC power jack or by unplugging the AC adapter.

# 3 General Operating Procedure

## 3.1 Theory of Operation

The 206A Olfactometer consists of four main sections: mass flow controllers complete with control electronics, odor vials with associated control valves and control circuits, Mixing valve, and Final valve. Additional odor vial sections, each consisting of four vials and valves, can be added to the base 206A to increase the total vial count from 4 to 8 to 12.



#### Figure 1 206A Piping Schematic

The Olfactometer generates precisely timed odor pulses by continuously flowing the odor flow to the Final valve where it is diverted to the exhaust until the odor challenge is required. Having the odor flowing continuously to the Final valve minimizes the time between actuation of the Final valve and deliver of the odor to the animal. In order to keep this time as short as possible the researcher should locate the Final valve in the animal chamber as close to the animal as is practical. We suggest the Final valve be located within about 10 cm of the animal. The design uses a second flow of fresh air that is also piped to the Final valve. This fresh air flow normally exits to the animal and when the Final valve is actuated the fresh air flow is sent to the exhaust. In order to ensure minimum delay time between valve actuation and arrival of the odor at the animal it is important that the odor flow rate and the fresh air

flow rate are equal. Equal flow rates also help ensure that the concentration profile delivered to the animal will be as "square" as possible by minimizes pressure and flow differences between the two flows. The fresh air flow is controlled by a third mass flow controller (MFC3) which is set to have a flow equal to the sum of the dilution flow plus the odor flow (MFC3 = MFC1 + MFC2).

Odor concentration is determined by two parameters: (1) the concentration of the odor placed in the vial and, (2) the relative flow rate of the odor and dilution gases. The odor concentration in the vial is controlled by diluting the odor solute with an appropriate solvent which lowers the head space concentration in the vial (concentration of gas above the liquid in the vial). The head space concentration in a particular vial is set by the researcher and this concentration can be further reduced by the relative flow rates of the dilution and odor flows as is set by the 206A control program. The odor concentration released to the animal can be calculated as follows.

$$C_{odor} = \frac{Q_{odor\ flow}}{Q_{Total\ flow}} \ x \ C_{HS}$$

Where  $C_{odor}$  is the concentration of the odor released to the animal,

 $Q_{odor flow}$  is the odor flow rate through the selected vial (MFC2 flow rate),

 $Q_{Total flow}$  is the total flow rate through the olfactometer (MFC1 flow rate + MFC 2 flow rate), and

 $C_{HS}$  is the concentration in the head space of the vial.

The range of  $C_{odor}$  is determined by both the odor flow rate and the total flow rate. Since the maximum odor flow rate is limited to 100 sccm (due to the mass flow controller chosen) and typical total flow rates are between 200 and 950 sccm the typical maximum odor concentration released to the animal is between 50% (100/200) and 10.5% (100/950) of the head space concentration. The minimum odor concentration released to the animal will be determined by the minimum flow rate of the odor mass flow controller which we recommend as 5 sccm (note: the mass flow controller manufacturer specifies flow rates as low as 1 sccm from the odor MFC but we don't recommend running the odor MFC this low). Therefore the minimum concentrations will also be dependent on the total flow rate and will range from 2.5% (5/200) down to 0.53% (5/950) of the head space concentration. This calculation is simplified for the researcher in our control program as the researcher only needs to enter the desired concentration percentage and then the program automatically sets the odor flow rate required to achieve this desired concentration based on the current total flow rate.

The standard 206A model includes four odor vials. These vials can be configured with different odors, the same odor with different concentrations or with no odor. In operation the olfactometer works by opening only one set of vial valves at a time. This allows the odor air (from MFC2) to flow through the selected vial and pick up the odor present in the head space above the odor liquid. This odor is then mixed with the dilution flow coming from MFC1 to create the total flow and this is delivered to the Final valve. When the animal is not being challenged the total flow (which includes the odor) is directed to the Final valve's Exhaust port. When the animal is challenged the Final valve switches the total flow to the Animal port of the Final valve and thus to the animal. At the same time fresh air (from MFC3) with a flow rate equal to the total flow (MFC1 + MFC2) is directed to the Final valve and exits from either the Animal port or the

Exhaust port depending on the state of the Final valve. Thus the animal experiences the same flow at all times but the flow is either fresh air or air with odor in it.

The Mixing valve is only opened under two circumstances, when a challenge with no vial is selected and when the olfactometer is being purged. When a challenge with no vial is selected in the control program the Mixing valve opens so that the air from MFC2 can mix with the air for MFC1 to maintain the same total flow as when odor flow is being produced. When the olfactometer is being purged all of the vial inlet and outlet valves close and the Mixing valve opens. This allows fresh air to pass through both sides of the olfactometer to clean any odor residue out of the tubing.

## 3.2 Odor Pulse Characteristics

A miniPID fast response photo-ionization sensor was used to measure the output odor pulse shape and timing produced by the 206A olfactometer. Figure 2 shows a typical trace, in this figure the green line shows the odor concentration and the red line shows when the Final valve was turned on and the off. The Final valve was actuated at the 50 ms mark of the test and was closed at the 250 ms mark for a total valve on time of 200 ms. As can be seen the odor pulse concentration starts to rise at about the 69 ms mark (19 ms after Final valve opening) and has reached steady state concentration by about the 89 ms mark (20 ms rise time). The closing delay, the time between turning off the Final valve and the odor pulse starting to fall, is longer (about 50 ms) than the opening delay. Both the opening and closing delays are functions of the valve actuation time, the internal flow design of the Final valve, and the volume of tubing downstream of the Final valve. For this test the tube between the Final valve and the miniPID inlet was 1/8" OD Teflon tubing with a length of about 8 cm. The fall time for the odor pulse can be seen to be about 50-60 ms. It is normal to see longer fall times for an odor pulse than rise times because extra time is required to purge the downstream tubing of odor molecules. Repeated testing of the 206A with a miniPID sensor revealed that the odor pulse characteristics are reasonably constant with a variation observed of 1-2 ms on the actuation time, rise time and fall time.



Figure 2 Odor Pulse and Final Valve Opening vs Time

## 3.3 Instrument Setup

#### 3.3.1 Unpacking the Instrument

The 206A Olfactometer is shipped in 1 or more boxes depending on how many extra Odor Vial Subassemblies you have purchased. Unpack all boxes included with the shipment and check the contents against the list provided below. If there are any issues with what you received, please <u>contact Aurora</u> <u>Scientific</u> as soon as possible.

Item	Description
206A Mass Flow Controller (MFC) Subassembly	MFC Subassembly consisting of 3 mass flow controllers, main controller circuit board, inlet manifold and installed fittings.
206A Odor Vial Subassembly	Odor vial subassembly consisting of 2 inline manifold valves and the holders for 4 odor vials. Note: the 206A Odor subassembly is shipped already attached to the 206A Mass Flow Controller Subassembly.
206A Mixing Valve Subassembly	Mixing valve subassembly consisting of the mixing valve mounted on a plate with installed tube fittings. Note: the 206A Mixing Valve subassembly is shipped already attached to the 206A Odor Vial subassembly.
206A Final Valve Subassembly	Final valve subassembly consisting of the Final valve with installed tube fittings. Note: the 206A Final valve subassembly is shipped separate from the Mass Flow Controller, Odor vial and Mixing valve subassemblies.
206A Accessory Subassembly	Accessories for 206A including: Teflon tubing 1/16" OD, Teflon tubing 1/8" OD, spare inert fittings for 1/16" tube, spare inert fittings for 1/8" tube, 4 vials with septa, septa puncture tool, 4mm supply tube, 4mm one-touch tube fitting for supply tube, power supply, power cord, USB cable, software disk, extension cable for connecting the Final valve to the 206A.
206B Odor Vial Subassembly (optional – purchased separately)	Odor vial subassembly consisting of 2 inline manifold valves and the holders for 4 odor vials. Note: the 206B Odor Vial Subassembly is shipped separate from the 206A Olfactometer.
206B Accessory Subassembly (optional – only present with 206B)	Accessories for 206B including: 4 vials with septa.

Table 3 Items included with a 206A Olfactometer

The following is a list of several items required in order to use the 206A Olfactometer. These items are not included with the 206A.

Required Items	Description	
Clean/Dry Air Source	A source of clean and dry air is required by the 206A. This air can be supplied by various means including: high pressure gas bottle, lab air supply or a compressor. When using either lab air or a compressor you may also require an air dryer, air filter and a charcoal filter to remove odors from the air.	
Air Dryer, Air Filter, Charcoal Filter	If the air source is lab air or a compressor then moisture, particles and odors may need to be removed from the air prior to entering the 206A. The Mass Flow Controllers must have clean, dry air or permanent damage could result. This extra	

	equipment is not required if using a high pressure gas bottle as the air source.
Upstream Air Regulator	The Mass Flow Controllers, valves, tubing and fittings that are used in the 206A all have maximum pressure ratings. A regulator must be installed upstream of the 206A to control the inlet pressure. Inlet pressure must be in the range 5-30 psig (35-207 kPa). DO NOT EXCEED THE MAXIMUM PRESSURE RATING OF 30 psig (207 kPa).
Animal Nose Cone or Other Odor Delivery Fitting	The outlet of the Final valve should be piped to an animal nose cone or other fitting designed to deliver the odor to the subject. The Final Valve uses ¼-28, 1/8" inert fittings on the ports. In most cases the nose cone should include a method of terminating a 1/8" OD (outside diameter) Teflon tube.
Mount for Final Valve	The Final valve includes tapped holes on the bottom of the valve (4-40UNC). The Final Valve should be mounted to a plate or other fixation device to keep it in place near the animal.
Mount for 206A Olfactometer	The 206A Olfactometer includes rubber feet on the bottom of each subassembly. The Olfactometer can be placed on a flat, level surface or it can be hung off the back of the test chamber by use of the included hooks designed into each part. A horizontally mounted length of 20mm x 20mm aluminum extrusion is ideal for mounting the 206A using the built-in hooks.
Exhaust Vent	The 206A Olfactometer continuously releases odor through the Final valve Exhaust port while in use. To prevent a buildup of odor in the test facility the exhaust from the Final valve should be piped to an exhaust vent.

Table 4 Required items not included with a 206A Olfactometer

NOTE: Maximum operating pressure is 30 psig (207 kPa). Do not exceed the maximum pressure ratings. Injury could result from use at higher pressures. Damage to the 206A will occur at pressures above the maximum rated pressure.

- 3.3.2 Initial Equipment Setup
  - 1. The 206A Olfactometer is shipped assembled and should either be placed on a flat, level surface on the attached rubber feet or mounted using the included hooks on a horizontally mounted length of 20mm x 20mm aluminum T-Slotted extrusion. (McMaster-Carr part number 5537T101).
  - 2. Install the included software on a Windows PC.
  - a) Insert the software install CD into the PC which should launch the installer.
  - b) Follow the on-screen wizard to complete the installation.
  - c) Following successful completion of the installation you should see a new icon on the desktop that can be used to launch the software.
  - 3. Once the software is installed then connect the USB port on the MFC Controller circuit board to a USB port on the PC.

- 4. Pipe the air supply to the 206A Olfactometer. (For the purposes of this document we will assume that you are using a high pressure air cylinder as the air source and therefore there is no requirement for drying, filtering or removing odors from the air source.)
- a) Attach the included 1/8" NPT to 4mm one-touch adapter to the outlet of a two stage regulator. Use Teflon tape to seal the 1/8" NPT threads prior to screwing the fitting into the regulator.
- b) Attach the regulator to the gas cylinder.
- c) Push one end of the supplied 4mm blue air tubing into the one-touch fitting on the regulator and the other end into the inlet manifold fitting on the MFC subassembly. Note: depending on the distance between the gas cylinder and the 206A you could cut the 4mm tubing to an appropriate length.
- d) Attach the Mixing valve outlet to the Final valve "Scent" inlet using the supplied 1/8" Teflon tubing and the supplied ¼-28 inert fittings.
- e) Attach the Exhaust bulkhead fitting (located on the Mixing valve plate) to the Final Valve "Air" inlet by inserting one end of the 1/8" Teflon tubing into the one-touch fitting and using a supplied ¼-28x1/8" port fitting to attach the tubing to the Final valve.
- f) Attach the Final valve electrical cable to the Final valve circuit board either directly or by using the valve extension cable supplied with the accessories.
- g) Attach the Final valve "Animal" outlet (normally closed) to the experiment using 1/8" Teflon tubing and the appropriate inert fittings supplied.
- h) Attach the Final valve "Exhaust" outlet (normally open) to the exhaust using 1/8" Teflon tubing and the appropriate inert fittings supplied.
- 5. Plug the power supply into the 206A Mass Flow Controller (MFC) circuit board and, using the supplied power cord, plug the AC adapter into an appropriate AC outlet.
- 6. Open the valve on the air cylinder and set the pressure on the regulator to 30 psig (207 kPa) maximum.
- 7. Check the tubing for air leaks.

At this point the olfactometer is installed and ready to use. Familiarize yourself with the software (chapter 4) and start using the olfactometer.

## 3.4 Loading a Vial

The 206A Olfactometer uses standard, unprocessed, 40 ml amber vials with a bonded septum (available from Aurora Scientific Inc., part # 206-VIAL or from Fisher Scientific, part # 14-965-103). Since these vials are unprocessed they are economically packaged in a case quantity of 72 vials in a shrink-wrapped tray that are ready for your in-house cleaning and certification process.

#### 3.4.1 Insertion of Teflon hoses through Vial Septum

An accessory kit is supplied with the 206A which includes a 16GA x 1.5'' hypodermic needle that can be used to assist in inserting the 1/16'' OD Teflon flow entry and exit tubes through the septum of the vial. Use the following procedure:

- 1) Remove the old vial and cap from the desired vial location in the olfactometer. To do this unscrew the cap, discard the vial and then hold the two 1/16" Teflon tubes on the outside of the cap as you pull the cap off of the tubes. Discard the cap.
- 2) Remove the cap from a fresh vial.
- 3) Push the 16GA needle, supplied in the accessory kit, through the septum from inside the cap.
- 4) Thread one of the two Teflon tubes through the 16GA needle from outside the cap to inside the cap. *Note: take care not to stab your finger with the 16GA needle tip.*

- 5) Remove the 16 GA needle from the septum. This should leave the Teflon tube passing through the septum from the outside to the inside of the cap.
- 6) Repeat steps (2) through (5) for the second 1/16" Teflon tube.
- 7) Screw the vial back onto the cap and place the vial in the holder of the olfactometer.

#### 3.4.2 Filling and Installing a Vial

You will note that the inlet tube into a vial is much longer than the outlet tube from a vial. The purpose of this is to ensure good mixing of the clean air entering the vial with the odor present in the head space of the vial. The incoming air is introduced low down in the vial (either just above the liquid/air interface or even below the liquid level). The outgoing air is collected near the top of the vial (near the cap). Because of this arrangement it is important that the liquid level in the vial is never high enough to immerse the outlet tube in the liquid. If the outlet tube is in the liquid then, when the vial valves open, liquid will be injected into the olfactometer. This will lead to significant contamination of the valves and all downstream tubing which may require replacement of all of the valves and all of the tubing.

Note: take care to ensure that the liquid level in the vial is ALWAYS below the level of the shorter Teflon tube present in the vial.

#### 3.4.2.1 Method 1 – Filling and Installing a Vial

- 1) Whenever an odor vial is changed we recommend purging the olfactometer to remove trace amounts of the original odor that may have absorbed into the downstream tubing/valves.
- 2) If you are changing an odor you MUST use a clean vial and vial cap/septum. DO NOT REUSE a vial or cap with a different odor.
- 3) If you are simply adding more of the same liquid then there is no need to change the vial/cap.
- 4) We recommend only changing vials when the olfactometer is not in use. Press the **Stop Flow** button on the main window to stop all flow through the olfactometer before changing vials.
- 5) Relieve pressure in the old vial by clicking on **Config>Advanced Status and Control** and then clicking on the vial you want to change. Ensure the vial valves are green (Open) which will open the vial to atmosphere and relieve any pressure present in the old vial. *Note: If necessary, vials can be replaced while the olfactometer is in use but you should never replace the vial currently in use.*

*Caution: If you haven't stopped the olfactometer flow or relieved the pressure in the old vial then take care when unscrewing the cap from the old vial to slowly relieve the pressure.* 

- 6) Remove the old vial and cap.
- 7) Close the vial valves by pressing the **Stop Flow** button or by clicking on the vial on the System Status window which you open with **Config>Advanced Status and Control**.
- 8) Replace the cap with the new one using the procedure in section 3.4.1.
- 9) Place a small amount of odor solute in the bottom of a clean, dry vial. In most cases you will be using something less than 10 ml of solute.
- 10) If required, dilute the solute with an appropriate solvent at the ratio required by your experimental procedure. We recommend limiting the solute plus solvent volume to less than 20 ml.
- 11) Screw the vial onto the cap and place the vial in the holder in the Olfactometer.
- 12) Check that the flow exit tube (the shorter of the two tubes in the vial) is well above the liquid/air interface. If not then remove the vial, and reduce the amount of liquid in the vial, and reinstall the vial on the cap before placing it back in the holder.
- 13) You are now ready to use the new vial.

Note: if changing from a strong odor to a less strong odor you may need to replace the two 1/16" diameter Teflon tubes that are in the end cap. These tubes can absorb strong odors into the plastic and outgas the strong odor after the vial has been replaced. You may also need to purge the unit when changing odors to remove any residue of the strong odor present in the downstream tubing/valves.

Alternative methods of filling a vial:

For very strong odors it may be advisable to fill the vial in a remote location away from the experiment so as to not introduce odor into the experimental area. To add a strong odor to a vial without contaminating the experimental air we suggest using one of the following methods.

#### 3.4.2.2 Method 2 – Fill Vial in a Fume Hood or in a Remote Location

- 1) Perform steps 1 through 7 of procedure 3.4.2.1.
- 2) Remove the 1/16" diameter Teflon tubes from the inlet and outlet manifold valves that lead to the vial you want to change. To do this simply unscrew the fittings from the valves.
- 3) Take the old vial complete with the inlet and outlet tubes to a fume hood or remote location.
- 4) If you have been using a strong odor then it is recommend that you replace the inlet and outlet tubes with new tubing. If you intend to reuse the tubes then remove them from the old vial's lid and insert the tubes in the septum of a new vial cap (see section 3.4.1).
- 5) Add the odor to a new vial and screw the cap onto the vial.
- 6) Take the new vial complete with 1/16" tubes back to the olfactometer and screw the fittings back into the manifold valves. ENSURE YOU ATTACH THE LONGER TUBE TO THE INLET VALVE AND THE SHORTER TUBE TO THE OUTLET VALVE.
- 7) Using this method will minimize any odor being released in the experimental area.

#### 3.4.2.3 Method 3 – Fill Vial with a Syringe

- 8) Perform steps 1 through 8 of procedure 3.4.2.1.
- 1) Screw a new clean, dry vial onto the cap and place it in the olfactometer.
- 2) The simplest method of filling a vial without releasing the odor in the experimental area is to use a syringe.
- 3) Prepare the odor liquid plus solvent in a remote location or in a fume hood. Fill a new disposable syringe with a sufficient amount of liquid for the experiment. To do this simply place the needle into the odor liquid and pull back on the plunger to fill the syringe. We suggest using a needle size between 20GA and 28GA.
- 4) Remove the needle from the liquid and pull some air into the hypodermic syringe thus emptying the needle. Wipe the exterior of the needle to remove any odor residue.
- 5) Take the syringe to the olfactometer and add the odor to a new vial which is already attached to the olfactometer by pushing the hypodermic needle through the vial's septum and then injecting a sufficient amount into the vial.
- 6) Remove the syringe from the septum and dispose of it in a safe manner.
- 7) Using this method will minimize any odor being released in the experimental area.

#### 3.4.2.4 Method 4 – Modify each Vial to Include a Filling Port

- 1) Insert a third 1/16" Teflon tube through the septum of each vial.
- 2) Attach a 1/6" tube to Luer adapter fitting to the end of the tubing.
- 3) Attach a shutoff valve with built-in Luer fittings to the Luer/tube adapter.

- 4) Attach a disposable syringe (without a hypodermic needle) filled with odorant to the shutoff valve.
- 5) Perform steps 1 through 8 of procedure 3.4.2.1.
- 6) Screw a new clean, dry vial onto the cap and place it in the olfactometer.
- 7) Open the shutoff valve.
- 8) Inject odorant into the vial using the syringe.
- 9) Close the shutoff valve. Note: leave the syringe attached to the valve to prevent a release of odor into the experimental area.
- 10) Using this method will minimize any odor being released in the experimental area.

## 4 Control Software

## 4.1 Software Setup

The 206A Olfactometer is supplied with a Software Install CD for Windows. Inserting this CD into a PC should launch the installer. Follow the on-screen wizard to complete installation of the control software. Following successful completion of the install wizard, the user should find an icon on the desktop to launch the control software.

## 4.2 Getting Started

Once the software is launched for the first time, the user must first configure the vials. See message in **Select Odor** text box shown in Figure 3.

Olfactometer Control	×
File Config Help	
SCIENTIFIC Performance. Precision. Progress.	
Select Odor Flow 0 sccm Temp -17.2 C	Flow On
You must Configure your vials.	Odor
Total Flow % C Odor Delivery 1000	Purge
through Vial 5 🐨 % Duration 1000 🐨 ms	Animal
Flow Odor To Exhaust Purge To Animal	Stop Flow
Flow Stopped	

Figure 3: Initial Startup

Selecting the **Config>Configure System** menu option will bring the user to the System Configuration screen, see Figure 4. From this screen, the user can select the odorants in their vials, set the Odor Flow Stabilization Delay, and set the Total System Flow Rate.

#### 4.2.1 Configure the Vials

- a) If odorants are already configured and you want to replace them click on the Clear Vials button at the bottom left of the Vial table data entry box.
- b) Click on line 1 and then click on the **Add New Odorant** button and type in the name of the odorant.
- c) Click on the Add New Solvent button and type in the name of the solvent.
- d) Enter the **Odorant Volume Concentration in Liquid** in the box shown. Note: this is the volumetric concentration of the odorant in the solvent. If you are not using a solvent then you should set the **Odorant Volume Concentration in Liquid** to 100%.
- e) Click on the **Set Selected Vial** button and you should see the odorant, solvent and concentration appear in the Vial table above.

- f) Follow the same procedure to enter all odorants required by the study. Note: the 206A will auto detect the number of vials connected to the olfactometer and grey out all vial numbers not present.
- g) Next enter the Odor Flow Stabilization Delay in seconds (this is the time that it takes an odor to flow from a vial to the Final Valve). This time is normally in the 5-60 second range depending on the system flowrate specified and the length of tubing between the Mixing Valve and the Final Valve. If you find that during the first few releases the concentration delivered to the animal is increasing then you probably have the Odor Flow Stabilization Delay set too short. (The scent is diluted in the tubing because you haven't displaced the full volume of the downstream tubing before releasing the first challenge to the animal). If this is the case increase the time.
- h) Set the **Total System Flow Rate** in sccm (standard cubic centimeters per minute). This is the sum of the odor flow plus the dilution flow. Note: the maximum flow is limited to 950 sccm.
- i) Click on the Save Config button to save the current configuration to a file.
- j) Click on the **OK** button to set the vials and return the user to the main screen.

	Odorant	Solvent	Concentratio
1	1-propanethiol	Water	0.00%
2	2-pentanol	Water	4.00%
3	1-decanol	Water	4.00%
4	Limonene	Water	25.00%
5	N/A	N/A	N/A
6	N/A	N/A	N/A
7	N/A	N/A	N/A
8	N/A	N/A	N/A
9	N/A	N/A	N/A
10	N/A	N/A	N/A
11	N/A	N/A	N/A
12	N/A	N/A	N/A
CI	ear Vials		Odorant Vol Concentratio
)dorar	nt Add New Odorant	Solvent Add New Sol	vent in Liquid
	ene 💌	Water	💌 25 🌩 🤊
imon			
Limon			Set Selected Via
Limon		Odor Flow Stabilization D	Set Selected Via

Figure 4: System Configuration Window

#### 4.2.2 Deliver an Odor

- a) On the Home window (Figure 3) click on the drop down arrow next to the **Select Odor** box.
- b) Choose the odor you want to release.
- c) Set the Total Flow % through Vial as a percentage of the Total System Flowrate. This is the main method of changing the concentration of the odorant delivered to the experiment. Note: the odor mass flow controller is limited to a maximum flowrate of 100 sccm and, for best accuracy, a minimum flowrate of 5 sccm. Therefore, the range of possible vial flowrates is dependent on the Total System Flowrate. For example, assuming a Total System Flowrate

of 950 sccm the maximum vial flowrate percentage would be 10.5% (100sccm/950sccm = 10.5%). The minimum vial flowrate percentage would be 0.53% (5sccm/950sccm = 0.53%). [Note: lower minimum vial flowrate percentages are possible as the mass flow controller manufacturer specifies a minimum flowrate of 1 sccm. However, for best accuracy and repeatability we suggest you limit flowrate to a minimum of 5 sccm.]

- d) Set the **Odor Delivery Duration** in milliseconds. This is the time that the odor will be delivered to the animal. The minimum duration time should be greater than 20 milliseconds for best results.
- e) Click on the button labelled Flow Odor to Exhaust to start the flow of the selected vial odor at the selected concentration and flowrate. The Deliver Odor to Animal button will remain greyed out until the Odor Flow Stabilization Delay time has completed.
- f) Click on the **Deliver Odor to Animal** button to deliver the selected odor to the animal.
- g) If at any time you want to see a diagram of which valves are active, then click on Config>Advanced Status and Control. The System Status window will open which will graphically show the flow path and which valves are on or off. The flowrates reported from the three mass flow controllers are also shown in this window.
- h) When the experiment is complete click on the **Purge** button to purge the tubing of odor.
- i) To shut off the flow completely click on **Stop Flow** button.

## 4.3 Menus

#### 4.3.1 File Menu

The file menu contains only one option, Exit.

#### 4.3.1.1 Exit Menu Item

Selecting this menu option will **Exit** the program. Upon exiting the program, the mass flow controllers are all set to zero flow and all valves are de-energized. This also happens when the program is closed via the "X" in the top right of the window.

#### 4.3.2 Config Menu

The Config menu contains 5 options. Configure System, Advanced Status and Control, Valve Check, Sequencer, and Stop Flow.

#### 4.3.2.1 Configure System Menu Item

Selecting this menu option will open the **Configure System** window. See section 4.5 for more details on the operation of this window.

#### 4.3.2.2 Advanced Status and Control Menu Item

Selecting this menu option will open the **Advanced Status and Control Window**. See section 4.6 for more details on the operation of this window.

#### 4.3.2.3 Valve Check Menu Item

Selecting this menu option will sequentially open and then close all vial valves attached on the 206A. This menu is only used to quickly check correct operation of the olfactometer.

#### 4.3.2.4 Sequencer Menu Item

Selecting this menu option will open the **Sequencer** window. The Sequencer window allows the user to create test sequences which consist of a vial to be used, the delay time before the challenge occurs, and the odor delivery duration. Over 100 steps can be included in a sequence which can then be edited, randomized and saved for later recall. Pressing the **Run** button on the Sequencer window will start the sequence running. See sections 4.7 and 4.8 for more details on the operation of this window.

#### 4.3.2.5 Stop Flow Menu Item

Selecting this menu option will set the mass flow controllers to zero flow and close all valves except the mixing valve. This will stop all flow through the system.

#### 4.3.3 Help Menu

The help menu contains only one option, About.

#### 4.3.3.1 About Menu Item

Selecting this menu option will open the **About** window and provide the user with the program version number and contact information for Aurora Scientific.

#### 4.4 Main Control Window

The main control window provides simple control of an olfaction experiment. See Figure 5 and the descriptions below for an explanation of each part of the window.

Olfactometer Control	×
File Config Help	
SCIENTIFIC Proformance. SCIENTIFIC Progress.	
Flow 805 sccm Board 23.9 C Sensor 0 C Select Odor	Flow On
Vial 1: 1-propanethiol (0.00%)	Odor
Total Flow % [ Odor Delivery 2000	Purge
through Vial 5 🐨 % Duration 2000 🐨 ms	Animal
Flow Odor To Exhaust Purge Deliver Odor To Animal	Stop Flow
Flowing Vial 1 To Animal	

Figure 5: Main Control Window of Configured System

#### 4.4.1 Status Indicators

Along the top, bottom, and right edges of the window are several status indicators which give status information about the current state of the system.

#### 4.4.1.1 Numeric Flow Indicator

At the top left of the window is the numeric **Flow** indicator. It shows current total flow rate through the system in real-time. In the example in Figure 5, it is easy to see that the current flow is 805 sccm.

#### 4.4.1.2 Board Temp and Sensor Temp Indicators

To the right of the numeric Flow indicator, also along the top edge of the window are two temperature indicators, the first, **Board Temp**, shows the current temperature in degrees C from an on-board temperature sensor in real-time, the second, **Sensor Temp**, shows the current temperature in degrees C from an optional thermistor that can be located in any desired location where the temperature should be monitored (e.g., vial liquid temperature).

#### 4.4.1.3 **Status** LEDs

Down the right hand edge of the window are several **Status** LEDs. These indicate the current system state, and are controlled by sensor feedback.

#### Flow On LED

The Flow On LED illuminates any time the flow is non-zero.

#### Odor LED

The **Odor** LED illuminates any time an odor vial is connected to the flow.

#### Purge LED

The **Purge** LED illuminates any time the odor flow is connected to the downstream tubing via the mixing valve and not through any vial.

#### Animal LED

The **Animal** LED illuminates any time the Final valve is energized and the odor stream is going to the animal. This LED is off if the odor stream is flowing to the exhaust.

#### 4.4.1.4 **Status** Bar

The **Status** bar is located along the bottom edge of the window, and provides a simple sentence describing the state of the flow, what odor it contains, and whether it is flowing to the exhaust or to the animal.

#### 4.4.2 Controls

The center portion of the window is occupied by a series of controls used to change the system output.

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#### 4.4.2.1 Select Odor Drop-down Menu

The **Select Odor** drop-down menu allows the user to select the desired vial to flow odor from. Each menu item is labelled with vial number, odorant name, and odorant concentration in solvent. See Figure 6.

Olfactometer Control	×
File Config Help	
SCIENTIFIC Performance. Precision. Progress.	
Select Odor Flow 0 sccm Temp -13.9 C	Flow On
Vial 1: 1-propanethiol (0.00%)	Odor
✓ Vial 1: 1-propanethiol (0.00%)	Ourge
Vial 2: 2-pentanol (4.00%)	Animal
Vial 3: 1-decanol (4.00%)	
Vial 4: Daves Odor (4.00%)	Stop Flow
Flow Stopped	

Figure 6: Vial Selection Drop-down Menu

#### 4.4.2.2 Total Flow % through Vial Input Box

This control allows the user to select the percentage of the total flow which goes through the vial. The total flow will consist of the flow through the vial plus the direct flow through the olfactometer. This is the main method of changing the concentration of the odorant delivered to the experiment.

Note: the odor mass flow controller is limited to a maximum flowrate of 100 sccm and, for best accuracy, a minimum flowrate of 5 sccm. Therefore, the range of possible vial flowrates is dependent on the Total System Flowrate. For example, assuming a Total System Flowrate of 950 sccm the maximum vial flowrate percentage would be 10.5% (100sccm/950sccm = 10.5%). The minimum vial flowrate percentage would be 0.53% (5sccm/950sccm = 0.53%).

Caution: lower minimum vial flowrate percentages are possible as the mass flow controller manufacturer specifies a minimum flowrate of 1 sccm. However, for best accuracy and repeatability we suggest you limit flowrate to a minimum of 5 sccm.

#### 4.4.2.3 Odor Delivery Duration Input Box

This control allows the user to select how long the Final valve will remain open when the user clicks the **Deliver Odor to Animal** button.

#### 4.4.2.4 Flow Odor to Exhaust Button

This button sets the values to flow the selected vial at the selected percentage to the exhaust. If the user has selected values which would exceed the control limits of the mass flow controllers, an error message is produced.

#### 4.4.2.5 Purge Button

This button sets the values to flow clean air only to the exhaust via the Mixing value and closes all vials from the flow.

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#### 4.4.2.6 Deliver Odor to Animal Button

This button opens the Final valve for the specified duration, thereby delivering the odor to the animal. This button will be disabled for the **Stabilization Delay Time** after clicking either the **Flow Odor to Exhaust** or **Purge** buttons.

## 4.5 System Configuration Window

The **System Configuration** window is used to set the contents of the vials, the system's total flow and the stabilization delay. See Figure 7 and the descriptions below for more details.

	Odorant	Solvent	Concentration
1	1-propanethiol	Water	0.00%
2	2-pentanol	Water	4.00%
3	1-decanol	Water	4.00%
4	Limonene	Water	25.00%
5	N/A	N/A	N/A
6	N/A	N/A	N/A
7	N/A	N/A	N/A
8	N/A	N/A	N/A
9	N/A	N/A	N/A
10	N/A	N/A	N/A
11	N/A	N/A	N/A
12	N/A	N/A	N/A
CI Odorai	ear Vials nt Add New Odorant S	olvent Add New So	Odorant Volu Concentratic Ivent in Liquid
Limon	ene 💌	Water	25 🚔 %
			Set Selected Vial
	Oc	lor Flow Stabilization [	Delay 60 🚔

Figure 7: System Configuration Window

#### 4.5.1 Controls

The System Configuration window has several controls to configure the vials and set the stabilization delay and system flow rate.

#### 4.5.1.1 Vials List Box

At the top of the window is a list box which describes the current setup of each vial.

Note: the 206A will auto detect the number of vials connected to the olfactometer and grey out all vial numbers not present.

#### 4.5.1.2 Clear Vials Button

Below the Vials list box is the **Clear Vials** button. This button will set all vials to have no odorant or solvent.

#### 4.5.1.3 Odorant Drop-down Menu

The **Odorant** drop-down menu provides the user a list of available odorants to choose from. The user can add odorants to this list via the **Add New Odorant** button. See Section 4.5.1.4 for more details.

#### 4.5.1.4 Add New Odorant Button

The **Add New Odorant** button allows the user to add odorants to the drop down list. Clicking the Add New Odorant button will open a dialog box for the user to add the desired odorant.

#### 4.5.1.5 Solvent Drop-Down Menu

The **Solvent** drop-down menu provides the user a list of available solvents to choose from. The user can add solvents to this list via the **Add New Solvent** button. See Section 4.5.1.6 for more details.

#### 4.5.1.6 Add New Solvent Button

The **Add New Solvent** button allows the user to add solvents to the drop down list. Clicking the Add New Solvent button will open a dialog box for the user to add the desired solvent.

#### 4.5.1.7 Odorant Volume Concentration in Liquid Data Entry Box

This control allows the user to specify the odorant concentration (by volume) that is in the vial.

#### 4.5.1.8 Set Selected Vial Button

The **Set Selected Vial** button sets the values of the **Odorant** drop down menu, the **Solvent** drop down menu, and the **Odorant Volume Concentration in Liquid** controls into the **Vials** list box on the selected row, and then advances the selected row to the next row.

#### 4.5.1.9 Odor Flow Stabilization Delay Data Entry Box

The **Odor Flow Stabilization Delay** data entry box allows the user to specify how long the flow takes to flow from the vial manifolds to the Final valve. This delay disables the **Deliver Odor to Animal** button for the delay time each time the odor is changed, to ensure that the tubing between the vials and the Final valve is fully evacuated of the previous odor.

#### 4.5.1.10 Total System Flow Rate Data Entry Box

The **Total System Flow Rate** control allows the user to set the desired total flow rate. This flow rate is the sum of the dilution mass flow controller and the odor mass flow controller. The total flow is limited to 950 sccm due to the limits of the mass flow controllers.

#### 4.5.1.11 MFC3 Compensation Factor Data Entry Box

The **MFC3 Compensation Factor** control allows the user to fine tune the fresh air flow rate to ensure that it is exactly equal to the odor flow rate. The olfactometer includes three mass flow controllers (MFC) that control the vial air flow rate, the dilution air flow rate and the fresh air flow rate. In operation the software sums the vial flow rate with the dilution flow rate and then sets the fresh air flow rate equal to this total flow rate. Due to small discrepancies in the MFCs it is possible that the fresh air flow will not be

equal to the total flow. In this case the **MFC3 Compensation Factor** allows the user to increase or decrease the fresh air flow rate until it is equal to the total flowrate. If the fresh air flow rate is too low, then enter a value greater than 1.0, if it is too high then enter a value less than 1.0. In most cases the compensation factor will be between 0.980 and 1.020. The MFC3 Compensation Factor is stored in the Config file.

#### 4.5.1.12 Load Config Button

The Load Config button allows the user to load a previously saved configuration.

#### 4.5.1.13 Save Config Button

The **Save Config** button allows the user to save the current configuration to a file for later recall using the **Load Config** button.

#### 4.5.1.14 Cancel Button

Closes the window without making any changes.

#### 4.5.1.15 **OK** Button

Closes the window and updates the main window with the current system configuration settings.

## 4.6 Advanced Status and Control Window

Open the Advanced Status and Control window by clicking on **Config>Advanced Status and Control**. When the Advanced Status and Control window opens it will display a graphical representation of the system status of the olfactometer in real-time. Clicking on the various valves and vials on the diagram will actuate them manually. Clicking on a mass flow controller will allow the user to set the flow rate manually. The software auto detects the number of vials connected to it and then updates the diagram to show all vials in the system. Figure 8 shows the System Status of a 206A with 4 vials while Figure 9 shows the same window with a 206A and a 206B installed for a total of 8 vials.

In both figures MFC1 is the dilution flow, MFC2 is the odor flow and MFC3 (the bottom one in the diagram) is the fresh air flow. The outlet of MFC3 is connected to the Clean Air In line on the Final valve. As vial valves are turned on and off the diagram of the valve changes from an X (valve off) to a flow through line (valve on) and the background colour of the valve changes from white (valve off) to green (valve on). Likewise the Final valve symbol changes to show the actual flow pattern through the valve and the background colour changes to green to show the valve is energized and odor flow is being sent to the Animal port.



Figure 8: Advanced Status and Manual Control Window with 206A – 4 Vials



Figure 9: Advanced Status and Manual Control Window with 206A and 206B – 8 Vials

## 4.7 Sequencer Window

The Sequencer Window allows the user to create a sequence of challenges to be executed by the olfactometer with pre-defined timing or based on the Sequence Trigger signal. The user can manually create a sequence to their specifications or they can have the software generate a random sequence using their chosen parameters, see Figure 10.

2	Sequencer			x
	Challenge Seq	uence		
	Vial	Delay To Challenge (s)	Odor Delivery Duration (ms)	^
	1	50	150	
	2	Trig	150	
	No Vial	50	150	
	3	Trig	Edge	
				-
	Total	100 s	450 ms	*
	Elapsed			
		[		-
	Move Up	Move Dowr	Remove	<u> </u>
	Shuffle	Load File	Save File	e
	Ge	enerate Rand	om	
			Odor	
		Delay to	Delivery	
	Vial	Challenge (s	) Duration	(ms)
	No Vial 💌	50 🌲	150	×
		Ext Trig	Falling	Edge
			Add	
		Show Du	ration Detail	s
		Cancel	Run	

Figure 10: Sequencer Window

#### 4.7.1 Sequencer Controls

#### 4.7.1.1 Challenge Sequence List Box

The **Challenge Sequence** list box shows the sequence as it will run. Rows can be added or deleted from the list using the other controls on the window.

#### 4.7.1.2 Total/Elapsed boxes

At the bottom of the Challenge Sequence list box are a four boxes which describe the **Total** and **Elapsed** time of the sequence. The Total time is generated as the challenge sequence is created. The Elapsed time is shown while the sequence is running.

#### 4.7.1.3 Move Up Button

The **Move Up** button moves the selected row up by one row in the sequence.

#### 4.7.1.4 Move Down Button

The **Move Down** button moves the selected row down by one row in the sequence.

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#### 4.7.1.5 *Remove* Button

The **Remove** button removes the selected row from the sequence.

#### 4.7.1.6 Shuffle Button

The Shuffle button keeps all of the rows in the sequence the same, but randomizes their ordering.

#### 4.7.1.7 Load File Button

The Load File button is used to load in a previously stored sequence from a file.

#### 4.7.1.8 Save File Button

The **Save File** button is used to save a sequence to a file for later use.

#### 4.7.1.9 Generate Random Button

The Generate Random button is used to open the Random Sequence Generator. See Section 4.8 for more details.

#### 4.7.1.10 Vial Drop Down

The Vial drop down is used to select which vial you want to add to the sequence with the Add button.

#### 4.7.1.11 Delay to Challenge (s) Data Entry Box

The **Delay to Challenge (s)** data entry box is used to enter the delay in seconds before the challenge is released. Note: this delay can never be less than the **Odor Flow Stabilization Delay** time that is set on the Config menu.

#### 4.7.1.12 Ext Trig Check Box

The **Ext Trig** check box is used to allow the sequence to be triggered by an external trigger signal. The trigger voltage must be a minimum of 1 volt to trigger and a maximum of 10 volts. Note: we are using an A/D input to monitor the trigger line and for this reason the maximum voltage is set by the A/D maximum which is 10 volts. Please note that the Final Valve will be triggered by the rising edge of the trigger pulse. Therefore, if the sequence includes a vial change, then you must include the Odor Flow Stabilization Delay time before the end of the previous step in the sequence before you issue the trigger. Failure to include this delay will result in unknown concentrations being released to the animal. This delay provides the time necessary for the new odor to travel the length of the tubing between the vials and the Final Valve. Step two in the sequence shown in Figure 10 shows a **Trig** in the Delay to Challenge (s) column which means that upon completion of the previous step of the sequence the Final Valve will open when the rising edge of the trigger signal is received by the olfactometer. In step two the Odor Delivery Duration (ms) is set to 150 which means that after the trigger opens the Final Valve it will stay open for 150ms and then close. Note: the sequencer proceeds to the next line of the sequence after the longer of either the **Odor Delivery Duration** or the trigger level falling back to ground (falling edge of trigger signal). For the case where you use a pulse trigger (a short duration on and then off of the trigger voltage) the next sequence step will start as soon as the Final Valve closes. For the case of a level trigger (the trigger goes high and stays high for the duration of the Final Valve opening) the next sequence step will start as soon as the olfactometer sees the falling edge of the trigger signal (i.e., when the trigger goes low).

#### 4.7.1.13 Odor Delivery Duration (ms) Data Entry Box

The **Odor Delivery Duration (ms)** data entry box is used to enter the duration of the challenge in milliseconds.

#### 4.7.1.14 Falling Edge Check Box

The **Falling Edge** check box is used to allow the trigger signal to control closing of the Final Valve. Step 4 in the sequence shown in Figure 10 shows the Final Valve will open on the rising edge of the trigger (**Trig**) in the **Delay To Challenge (s)** column and will close on the falling edge of the trigger (Edge) in **Odor Delivery Duration (ms)** column. In order to use the Falling Edge trigger the user must us a level trigger (the trigger goes high and stays high for the duration of the Final Valve opening and then goes low).

#### 4.7.1.15 Add Button

The **Add** button is used to add a new row to the sequence based on the current values of the Vial, Delay to Challenge, and Odor Delivery Duration controls.

#### 4.7.1.16 Show Duration Details Button

The **Show Duration Details** button is used to open a window with detailed information about odor delivery per vial as described by the sequence in the Challenge Sequence list box, see Figure 11. This table provides a simple method of seeing the total **Delay to Challenge** and total **Odor Delivery Duration** times per vial as well as the grand totals for the entire sequence. Note: if your sequence uses an external trigger to control the release then the times associated with the triggered releases will not be known by the program and will therefore be set to zero in the Duration Details.

Note: the Duration Details window must be closed before continuing editing on the Sequencer window.

#### 4.7.1.17 Cancel Button

The **Cancel** button is used to exit the Sequencer window.

#### 4.7.1.18 **Run** Button

The **Run** button is used to run the sequence described in the Challenge Sequence list box. While a sequence is running, the Challenge Sequence list box will highlight the current step in the sequence, and the olfactometer Main window will be disabled so the user cannot accidentally interfere with the sequenced operation. Also, while a sequence is running, the Run button changes into an Abort button, which stops the running sequence immediately. At the conclusion of a running sequence, the main window is re-enabled and the Abort button changes back to Run.

Duration Details					
		Delay To Challenge (s)	Odor Delivery Duration (ms)		
	No Vial	40	400	l	
	Vial 1	20	200		
	Vial 2	20	200		
	Vial 3	20	200		
	Vial 4	20	200		
	Vial 5	0	0		
	Vial 6	0	0		
	Vial 7	20	200		
	Vial 8	0	0		
	Vial 9	0	0		
	Vial 10	0	0		
	Vial 11	0	0		
	Vial 12	0	0		
	Total	140 s	1400 ms		
Grand Total Experiment Duration 00:02:21.400					

Figure 11: Sequence Duration Details Window

#### 4.8 Random Sequence Generator

The Random Sequence Generator will generate a random challenge sequence according to a set of rules the user defines. The user sets the controls to their desired settings and clicks Generate, and this populates the sequencer with a randomly generated sequence that matches the settings chosen. To open the Random Sequence Generator window click on the **Generate Random** button on the Sequencer window, see Figure 12. Note: the Random Sequence Generator does not include external triggers in the sequence.

#### 4.8.1 Generate Random Sequence Controls

#### 4.8.1.1 Number of Challenges Data Entry Box

The **Number of Challenges** data entry box allows the user to select the total number of challenges that will be included in the generated sequence.

#### 4.8.1.2 Included Vials Checkboxes

The **Included Vials** checkboxes allow the user to select which of the attached vials should be included in the randomized challenge. The user can select all vials, some vials, or no vial as a valid option. Vials not present due to the hardware setup are shown greyed out. In this example 8 vials are present in the hardware setup so Vials 9-12 are greyed out.

2	Generate Random Sequence
	Number of Challenges 10 🚔
	Included Vials
	Vial 1 Vial 5 Vial 9 Vial 2 Vial 6 Vial 10
	Vial 3 Vial 7 Vial 11 Vial 4 Vial 8 Vial 12
	Allow No Vial as a valid Challenge
	Minimum Odor Delivery Duration 50 🚽 ms
	Maximum Odor Delivery Duration 200 🚔 ms
	Odor Delivery Duration Step Size 50 🚔 ms
1	Minimum Delay To Next Challenge 🛛 🚖 s
Ν	Aaximum Delay To Next Challenge 👩 🚔 s
	Delay To Next Challenge Step Size 5 🚔 s
	Cancel Generate

Figure 12: Random Sequence Generator

#### 4.8.1.3 *Minimum Odor Delivery Duration (ms)* Data Entry Box

The **Minimum Odor Delivery Duration (ms)** data entry box allows the user to select the shortest duration the randomizer will deliver odor for in milliseconds.

#### 4.8.1.4 Maximum Odor Delivery Duration (ms) Data Entry Box

The **Maximum Odor Delivery Duration (ms)** data entry box allows the user to select the longest duration the randomizer will deliver odor for in milliseconds. Note: the minimum and maximum times can be set the same if all odor delivery durations need to be the same.

#### 4.8.1.5 Odor Delivery Duration Step Size (ms) Data Entry Box

The **Odor Delivery Duration Step Size (ms)** data entry box allows the user to set the step size the randomizer will use when selecting durations within the range described by the minimum and maximum controls in milliseconds.

#### 4.8.1.6 Minimum Delay to Next Challenge (s) Data Entry Box

The **Minimum Delay to Next Challenge (s)** data entry box allows the user to select the shortest delay the randomizer will wait between challenges in seconds.

#### 4.8.1.7 Maximum Delay to Next Challenge (s) Data Entry Box

The **Maximum Delay to Next Challenge (s)** data entry box allows the user to select the longest delay the randomizer will wait between challenges in seconds.

#### 4.8.1.8 **Delay to Next Challenge Step Size (s)** Data Entry Box

The **Delay to Next Challenge Step Size (s)** data entry box allows the user to set the step size the randomizer will use when selecting delay times within the range described by the minimum and maximum controls in seconds.

#### 4.8.1.9 Cancel Button

The **Cancel** button closes the Random Sequence Generator without generating a random sequence.

#### 4.8.1.10 Generate Button

The **Generate** button populates the Sequencer window with a randomly generated sequence that meets the parameters specified by the user and then closes the Random Sequence Generator window, see Figure 13.

2	Sequencer			×
	Challenge Seq	uence		
	Vial	Delay To Challenge (s)	Odor Delivery Duration (ms)	*
	4	60	200	
	8	25	200	
	1	60	150	
	1	45	100	
	4	30	150	
	4	60	50	
	8	50	150	
	4	55	200	
	3	60	50	
	7	35	150	
				-
	Total	480 s	1400 ms	<u> </u>
	Elapsed			-
	Move Up	Move Dowr	Remove	2
	Shuffle	Load File	Save Fil	e
	Ge	nerate Rand	om	
			Odor	
		Delay to	Delivery	
	Vial	Challenge (s	) Duration	(ms)
	No Vial 💌	20 🚖	20	-
			Add	
		Show Du	iration Detail	s
		Cancel	Run	

Figure 13: Example Challenge Sequence Generated by Random Sequence Generator

## 5 Calibration and Maintenance

## 5.1 Calibration

The flow rate of the three mass flow controllers should be checked periodically. This can be done by connecting either an electronic flow meter or simple rotameter to the Exhaust outlet of the Final valve. Open the **Config>Advanced Status and Control** window to provide manual control of the mass flow controllers and all valves.

## 5.1.1 Calibration of Mass Flow Controllers

Before starting ensure all mass flow controllers are set to zero flow and all valves are closed. Ensure an adequate air supply is available and is connected to the olfactometer.

#### 5.1.1.1 Mass Flow Controller 1 – Dilution Air

Click on MFC1 on the diagram and enter the flow rate you wish to calibrate the mass flow controller at (maximum flow rate is 1000 sccm). Next ensure all valves are turned off, simply click on a valve to toggle its state from ON to OFF. At this point all air passing through MFC1 should be exiting from the Exhaust port of the Final valve. Measure the flow rate at the exhaust exit using an electronic flow meter or a rotameter.

#### 5.1.1.2 Mass Flow Controller 2 – Odor Air

Click on MFC2 on the diagram and enter the flow rate you wish to calibrate the mass flow controller at (maximum flow rate is 100 sccm). Next ensure all valves are turned off, simply click on a valve to toggle its state from ON to OFF. Now turn on the Mixing valve by clicking on the diagram. At this point all air passing through MFC2 should be exiting from the Exhaust port of the Final valve. Measure the flow rate at the exhaust exit using an electronic flow meter or a rotameter.

#### 5.1.1.3 Mass Flow Controller 3 – Fresh Air

Click on MFC3 on the diagram and enter the flow rate you wish to calibrate the mass flow controller at (maximum flow rate is 1000 sccm). Next ensure all valves are turned off, simply click on a valve to toggle its state from ON to OFF. At this point all air passing through MFC3 should be exiting from the Animal port of the Final valve. Measure the flow rate at the Animal port using an electronic flow meter or a rotameter.

#### 5.1.1.4 Balancing Mass Flow Controller 3 and Total Flow

Connect an electronic flow meter or a rotameter to the Animal port of the Final valve. Use the System Configuration window (Config>System Configuration) to set the Total System Flow Rate to the desired total flow. We suggest you set the total flow to a value close to what you normally use in your experiments. Ensure that the MFC3 Compensation Factor is set to 1.000. Place a clean vial in the vial location 1 and select that vial on the main window from the Select Odor drop down menu. Set the Total Flow % through Vial on the main window to a typical value that you use during your experiments (alternatively run this procedure several times at a range of Total Flow % through Vial values). Set the Odor Delivery Duration on the main window to a long enough value that you can read the flow rate when the odor is switched, we suggest using 2000 – 5000 ms. Click on the Flow Odor to Exhaust button to start

the flow. Wait until the **Deliver Odor to Animal** button is active (not greyed out). Record the flow rate shown on the rotameter. Click the **Deliver Odor to Animal** button and record the flow rate. When you click on the **Deliver Odor to Animal** button the Final valve switches the Animal port (where you have your rotameter attached) from the Fresh air line from MFC3 to the total flow line from MFC1 + MFC2. These two flow rates should be equal. If you observe that the Fresh air flow is less than the total flow then increase the **MFC3 Compensation Factor** on the **System Configuration** window to be greater than 1.000 (we suggest you change the compensation factor in 0.01 steps. Press the **OK** button and then the **Flow Odor to Exhaust** button again. Once again compare the flow rate at the Animal port of the Final valve before and after you press the **Deliver Odor to Animal** button. If the flow rates are still not the same repeat the process of adjusting the **MFC3 Compensation Factor**. Once you determine the correct compensation factor press the **Save Config** button to save this configuration.

#### 5.2 Maintenance

#### 5.2.1 Recommended Daily Maintenance

Daily maintenance is important to ensure minimal cross contamination of odors. The frequency of this maintenance may need to be more or less often depending on the number of experiments being run and the number of different odors being used. For greater numbers of experiments and greater numbers of different odors the frequency and duration of this maintenance may need to be increased.

The following procedure should be completed at the end of each day.

1) Purge the system for a minimum of 30 minutes. Click on the **Purge** button to start the purge process. All vial valves will be closed and the Mixing valve will be opened. Air will then be run through all of the olfactometer tubing to remove any odor residue present.

#### 5.2.2 Recommended Monthly Maintenance

Monthly maintenance is important to ensure continued reliable operation of the olfactometer. The frequency of this maintenance may need to be more or less often depending on the number of experiments being run and the number of different odors being used. For greater numbers of experiments and greater numbers of different odors the frequency of this maintenance may need to be increased.

The following procedure should be completed at least once per month.

- Replace all vials with new, empty ones and purge through each vial separately for 30 minutes. From the main window select the first vial from the Select Odor drop down list, change the Total Flow % through Vial to 10%, click on Config>Configure System and set the Total System Flow Rate to 950 sccm. Click on Flow Odor to Exhaust and let run for 30 minutes.
- 2) Repeat step (1) for each vial in the system. This should remove odor residue from the vial valves and vial tubing.

- 3) Purge the system for a minimum of 30 minutes. Click on the **Purge** button to start the purge process. All vial valves will be closed and the Mixing valve will be opened. Air will then be run through all of the olfactometer tubing to remove any odor remaining odor residue.
- 4) In many cases severe contamination is best handled by replacing all of the olfactometer tubing and fittings downstream of the mass flow controller subassembly. The tubing used is 1/16" OD and 1/8" OD Teflon tube. The fittings are: ¼-28 inert fittings for 1/16" and 1/8" tubing. All of these parts can be purchased from <u>Aurora Scientific</u>.
- 5) If odor contamination is still present after steps (1) through (4) then we suggest flushing the system with liquid solvent.
  - a. Replace each vial with a fresh vial containing the solvent for the odor that was present in that vial location making sure to fill all solvent vials to the top of the vial (40ml liquid). By filling the vial you will cause liquid to be driven from the vial into the downstream valves and tubing. This liquid solvent should decontaminate these downstream parts. Under no circumstances should you ever run liquid odor solute through the valves and tubing as this will damage the system and require replacement of all valves and tubing.
  - b. Unscrew the ¼-28 inert fitting from the scent port on the Final valve. Place the tube into a beaker. Ensure that the tube will not come out of the beaker when the flow starts and cover the beaker to protect from splashing of the solvent as it exits the tube.
  - From the main window select the first vial from the Select Odor drop down list, change the Total Flow % through Vial to 10%, click on Config>Configure System and set the Total System Flow Rate to 950 sccm. Click on Flow Odor to Exhaust and let run for 30 minutes.
  - d. Repeat for each vial in the system. This should remove odor residue from the vial valves and vial tubing.
  - e. Replace all vials with clean ones and then repeat Steps (1) through (3) above to dry out the valves and tubes.
  - f. Note: you can create a cleaning sequence using the Sequencer and then **Run** this sequence overnight to clean and purge the olfactometer.
- 6) In extreme contamination cases you may need to replace the vial manifold valves, Mixing valve and possibly the Final valve as well. If you do replace these components then you must also replace all tubing and fittings after the mass flow controller subassembly. <u>Contact Aurora</u> <u>Scientific</u> to purchase replacement parts.

# 6 Performance Guarantee, Technical Support, Warranty and Repair Information

Aurora Scientific is dedicated to providing you with products that allow you to meet your research goals. For this reason we offer a performance guarantee, technical support and a new product warranty. Our performance guarantee ensures you purchase the correct instrument for your research. Technical assistance is always free and will be available for the life of your product. If you do have a problem with a product then please know that all Aurora Scientific products are covered by a three-year warranty covering both parts and labour. If you need to return a product to us for repair then consult the final section of this chapter for returns information.

## 6.1 Performance Guarantee

Our performance guarantee states: if for any reason a new product does not meet your research needs then you can return it to Aurora Scientific for exchange or a full refund. The performance guarantee only applies to new products and must be exercised within 60 days of receipt of the instrument.

## 6.2 Technical Support

Technical assistance is always free and will be available for the life of your product. Please don't hesitate to contact us if you have any technical support issues. Contact us by telephone, email, fax, or regular mail.

# 6.3 Technical Support Contact Information and Return Shipping Addresses

#### Canada, USA, South America, Middle East, Africa

Aurora Scientific 25 Industry St. Aurora, Ontario, CANADA L4G 1X6 Attn: RMA Returns Tel: +1-905-727-5161 Fax: +1-905-713-6882 Email (all Aurora Scientific Offices): <u>techsupport@aurorascientific.com</u> Web Site: <u>www.AuroraScientific.com</u>

#### Europe

Aurora Scientific Europe Hilton House 3 Ardee Road Rathmines, Dublin 6, Ireland Attn: RMA Returns Tel: +353-1-525-3300 Fax: +353-1-443-0784

#### Asia, Australia, New Zealand

Aurora Scientific Asia Unit B, 10/F Charmhill Centre 50 Hillwood Road Tsimshatsui, Kowloon, Hong Kong Attn: RMA Returns Tel: +852-3188-9946 Fax: +852-2724-2633

## Distributors

Japan Kantoh Electronics Co., Ltd. 1-25-14, Nakacho, Meguro-ku Tokyo, 153-0065, Japan Tel: +81-03-5773-5028 Fax: +81-03-5773-5029 Email: <u>info@Kantoh-elec.co.jp</u> Web site: <u>http://www.kantoh-elec.co.jp</u>

## 6.4 Warranty

Products manufactured by Aurora Scientific Inc. are guaranteed to the original purchaser for a period of three (3) years. Under this warranty, the liability of Aurora Scientific is limited to servicing, adjusting and replacing any defective parts that are of Aurora Scientific manufacture. Aurora Scientific is not liable to the customer for consequential or other damages, labour losses or expenses in connection with or by reason of the use or inability to use the products manufactured by Aurora Scientific.

Guarantee of parts and components not manufactured by Aurora Scientific shall be the same as the guarantee extended by the manufacturer of such components or parts. Where possible such parts returned to Aurora Scientific will be sent to the manufacturer for credit or replacement. Ultimate disposition of these items will depend upon the manufacturer's decision.

All shortages must be reported within ten (10) days after receipt of shipment.

Except where deviations are specified in literature describing particular products, the limited warranty above is applicable to all Aurora Scientific products, provided the products are returned to Aurora Scientific and are demonstrated to the satisfaction of Aurora Scientific to be defective.

Transportation costs of all products returned to Aurora Scientific must be borne by the customer and products must be returned to Aurora Scientific within three years after delivery to the original purchaser. Aurora Scientific cannot assume responsibility for repairs or changes not authorized by Aurora Scientific or damage resulting from abnormal or misuse or lack of proper maintenance.

Repair or service work not covered under the limited warranty will be billed at current service rates.

NO EXPRESS WARRANTIES AND NO IMPLIED WARRANTIES WHETHER FOR MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR USE, OR OTHERWISE OTHER THAN THOSE EXPRESSLY SET FORTH ABOVE WHICH ARE MADE EXPRESSLY IN LIEU OF ALL OTHER WARRANTIES, SHALL APPLY TO PRODUCTS SOLD BY AURORA SCIENTIFIC INC, AND NO WAIVER, ALTERATION OR MODIFICATION OF THE FOREGOING CONDITIONS SHALL BE VALID UNLESS MADE IN WRITING AND SIGNED BY AN EXECUTIVE OFFICER OF AURORA SCIENTIFIC INC.

## 6.5 Returning Products to Aurora Scientific for Repair

There are a few simple steps that must be completed before returning your product to Aurora Scientific.

1. Obtain a Return Material Authorization number (RMA#).

<u>Go to our website</u> or contact our technical support department to obtain a RMA #. We require the serial number of the product along with your contact information, i.e., your name, institution, phone number and email address.

2. Package your instrument.

Use the original packaging materials if available. If you do not have original packaging, then ensure that the product is wrapped in bubble pack and placed in a sturdy corrugated cardboard box.

3. Prepare Customs documents.

Canadian Clients: no customs documents are required, skip to step 4.

European Clients: no customs documents are required, skip to step 4 and ship to Aurora Scientific Europe.

Asia, Australia and New Zealand Clients: no customs documents are required, skip to step 4 and ship to Aurora Scientific Asia.

**USA and Rest of the World Clients**: We will supply you with a Commercial Invoice (CI) that must be included with the shipment. Please look for the CI in our return email message. You must sign and date the commercial invoice.

Place three (3) copies of the CI in an envelope and mark the outside CUSTOMS PAPERS ENCLOSED. Attach the envelope to the outside of the box.

4. Choose a shipper and prepare the waybill.

European Clients: ship your instrument to Aurora Scientific Europe in Dublin, Ireland.

Asia, Australia and New Zealand Clients: ship your instrument to Aurora Scientific Asia in Hong Kong.

Canadian, USA and all other Clients: ship your instrument to Aurora Scientific in Ontario, Canada.

You may ship your instrument back to us via the courier of your choice or via parcel post. If possible we prefer that you ship via FedEx. You are responsible for both the shipping and brokerage charges so please mark the waybill accordingly. Please don't ship freight collect. Shipments sent freight collect will be received but you will be invoiced for the shipping charges when your instrument is returned.

5. Prepare and send a purchase order.

After we receive the instrument we will evaluate it and contact you with the estimated repair cost. We require a purchase order before we can repair and return your instrument. Please fax or email us the purchase order at your earliest convenience.

# 7 Specifications

#### 206A Olfactometer

Modules Included: Mass Flow Controllers [SCCM]:	Mass Flow Controller, 4 CH Odor, Mixing valve, Final valve Odor: 0-100 Dilution Air: 0-1000 Exhaust Air: 0-1000
Total Flowrate Range [SCCM]:	100-950
Odor Vials:	4
Odor Dilution Range [%]:	0.1 – 10
Odor Pulse Duration [ms]:	20 – continuous
Odor Pulse Delay [ms <sup>+</sup> ]:	40
Communication Port:	USB (mini-B)
Final Valve Sync Output:	Hi = 4V - Final Valve flows to Exhaust
	Lo = 0V - Final Valve flows to Animal
Sequence Trigger Input:	TTL signal triggers opening of Final Valve and indexing of sequence
	Hi > 1V (maximum 10V) - Final Valve opens, flows to Animal Lo = 0V - Final Valve closed, flows to Exhaust
General Specifications	
Operating Temperature [°C]:	0 - 40
Power Required:	In: 100 - 240 VAC, 50/60 Hz. @ 1.8A
Power Consumption [w]:	15
Dimensions [cm]:	5.2 Mass Flow Control Modulo: 17L x 16W/x 21H
Dimensions [cm]:	A CH Oder Medule (2000): 17L x 16W X 21H
	Complete 206A: 38L x 16W x 21H
206A Software	
Recommended PC Specifications	
Processor:	Quad-core Intel or AMD
RAM:	4 GB
Hard Disk:	500+ GB
Operating System:	Windows 7/8/10
Coffeena Functiona	

Software Functions

Main Window:

Indicators: Flow, Board Temp, Sensor Temp, Status line, LEDs: Flow On, Odor, Purge, Animal

<sup>&</sup>lt;sup>1</sup> Final Valve switching to 99% of desired odor concentration

		Parame Deliver Animal	eter Entry: Select Odor, Total Flow % through Vial, Odor y Duration, Flow Odor to Exhaust, Purge, Delivery Odor To , Stop Flow
Menu l File:	tems:		
	Exit:		Exits program
Config:			
	Configure System:		Vial Definition List, editing controls, Odor Flow Stabilization Delay, Total System Flow Rate, MFC3 Compensation Factor, Load Config, Save Config
	Advanced Status and Manual Co	ntrol:	Real-time display of flow rate settings and valve state, click on valves to turn on and off, click on mass flow controllers to set flow rate
	Valve Check:		Cycles through all vial valves first turning them on and then turning them off
	Sequencer:		Challenge Sequence list, Total and Elapsed Time indicators, editing controls, Generate Random Sequence, Vial selection, Delay to Challenge time, Odor Delivery Duration, Show Duration Details, Run. Timed or triggered operation of Final Valve and sequence indexing
	Generate Random Sequ	ence:	Number of Challenges, Vial selection, Minimum Odor Delivery Duration, Maximum Odor Delivery Duration, Odor Delivery Duration Step Size, Minimum Delay to Next Challenge, Maximum Delay to Next Challenge, Delay to Next Challenge Step Size, Generate
	Stop Flow:		Stops flow
Help:			
	About:		version number, contact information

## 8 Appendix – Serial Communication Commands

To communicate with a 206A, you must connect it to your PC via a USB port. The 206A will connect to the PC and appear as a serial COM port. The PC operating system will normally assign the connection to either COM3 or COM4. User software should be designed to search all COM ports until it finds one which responds to 206A style commands and then use this port for communication.

## 8.1 Commands

The following is a list of the valid commands, and examples of their usage along with the replies returned from the 206A. In all cases the instrument acknowledges a command as soon as it is received by either a successful reply or by an unsuccessful reply.

The I2C address of a given 206A is set by the I2C Address switches on the mass flow controller (MFC) circuit board. All 206A olfactometers are shipped with the I2C address set to 1.

## 8.1.1 Valve

The valve command is used to set the state of any of the olfactometer's valves. The valves are numbered from 1 to 32 as follows:

Valve Number	Location/Function
1 (J18)	MFC circuit board/cycles with Final valve
	Can be used to monitor Final valve actuation – contact Aurora Scientific Inc. for wiring information.
2-6 (J19-J23)	MFC circuit board/Extra valve connectors
7 (J1)	Final/Mixing valve
8 (J2)	Final/Final valve
9-16 (J4-J11)	Vial Module 1/Vials 1 – 4
17-24 (J4-J11)	Vial Module 2/Vials 5 – 8
25-32 (J4-J11)	Vial Module 3/Vials 9 – 12

Table 5 Valve Number and Location

Command: valve <I2C ADDR> <VALVE ID> {on | off}

e.g.: valve 1 9 on

Description: Turn on valve 9 located on Vial module 1 (this is the vial 1 inlet valve).

## 8.1.2 Vial

The vial command is used to set the state of a pair of valves on the 206A connected to each vial. The vials are numbered as follows:

Vial Number	Location/Function
1–3	MFC board
4	MFC/Final valve-Mixing valve
	Do not use the Vial command to control the Final valve or Mixing valve
5-8	Vial Module 1/Vials 1 – 4
9–12	Vial Module 2/Vials 5 – 8
13–16	Vial Module 3/Vials 9 – 12

Table 6 Vial Number and Location

Command	vial <i2c addr=""> <vial id=""> {on off}</vial></i2c>
e.g.	vial 1 5 on
Description:	Turn on the inlet and outlet valves to vial 1 on the first vial module.

## 8.1.3 MFC

The MFC command is used to set the set point of the Mass Flow Controllers (MFCs) on the 206A. The set point is expressed as a number between 0 and 1, and is scaled to the flow rate of the MFC. The MFC's are numbered as follows:

MFC 1 = Hi flow (1000 SCCM) for Odor Dilution Air

MFC 2 = Lo flow (100 SCCM) for Odor Flow

MFC 3 = Hi flow (1000 SCCM) for Clean Air to Exhaust

Command MFC <I2C ADDR> <MFC ID> <SETPOINT>

e.g. MFC 1 1 0.4

Description: Set MFC 1 to a flow rate of 400 SCCM.

## 8.1.4 findModules

The findModules command is used to determine how many vial modules are connected to the 206A.

CommandfindModules <I2C ADDR>e.g.findModules 1Description:Inquire number of modules attached to olfactometer at address 1.Successful Reply:2

## 8.1.5 Identify

This command is used to identify the 206A.

Command	identify
e.g.	identify
Description:	Inquire as to identity of device attached to COM port.
Successful Reply	y: Aurora Scientific 206A

## 8.1.6 Final

This command is used to open the Final valve for a hardware timed duration in milliseconds.

e.g. final 1 400

Description: Open final valve for 400 milliseconds then close it. Note: this command also energizes the valve 1 connector that is on the MFC circuit board. This connector can be used to monitor the timing of the Final valve.

CAUTION: all valves are powered by 24VDC. Direct connection of a valve connector to a digital I/O line on an A/D card could result in damage to the I/O port. Special connection wiring is required. Contact Aurora Scientific Inc. for wiring information if you want to use the valve 1 connector to monitor the Final valve timing.

## 8.1.7 Temp

This command is used to query the Temperature in degrees C reported by the onboard or external thermistors. The temperature sensor ID's are as follows:

1 = Onboard Thermistor

2 = External Thermistor

Command	temp <i2c addr=""> <sensor id=""></sensor></i2c>
e.g.	temp 1 1
Description:	Inquire temperature of onboard thermistor
Successful Reply	26.43

## 8.1.8 resetTrig

The resetTrig command is used to reset the trigger counter to zero.

Command	resetTrig <i2c addr=""></i2c>
e.g.	resetTrig 1
Description:	Reset trigger count.

## 8.1.9 checkTrig

The checkTrig command is used to check the current value of the trigger counter. The trigger counter increments on the falling edge of each trigger pulse input.

Command	checkTrig <i2c addr=""></i2c>
e.g.	checkTrig 1
Description:	Check the trigger count.
Successful Repl	y: 4

## 8.1.10 setTrig

The setTrig command is used to set the duration in ms of the final valve opening on the rising edge of trigger input. Setting a duration of zero ms will cause the final valve to remain open until the falling edge of the trigger input.

Command	setTrig <i2c addr=""> <duration></duration></i2c>
e.g.	setTrig 1 100
Description:	Set trigger duration to 100ms.

# 9 Appendix – Connections

Connections to the olfactometer and between the various sections of the olfactometer are presented in this Appendix.

Connector	Label	Pins	Description
J4		1-6	Programming Header
J5		1, 2	Thermistor
JG	Analog	1, 7	Control/Readout for MFC3 – Exhaust Air
J7	Digital	7, 8	Pin 7 = TTL sync with Final Valve Open, Pin 8 = GND
J8	Analog In	1, 2	Sequence Trigger In, BNC inner = Trigger, BNC outer = GND
J9	+5V/GND	1, 2	5VDC Output Power
J10		1-10	Ribbon Connector to Vial Module
J11	24V/GND	1,2	Power for MFC3 – Exhaust Air
J12	24V/GND	1,2	24VDC Output Power
J13, J14	I2C	All	I2C Communications Ports
J15	MFC1	All	MFC1 – Dilution Air
J16	MFC2	All	MFC2 – Odor Air
J17	LAST	1, 2	Jumper to set MFC PCB as last Vial board – do not install
J18		1, 2	Extra Final Valve Connection
J19-J23		1, 2	Valve Connections
J24	24V	1, 2	24VDC Input Power

Table 7 provides a list of connections to the Mass Flow Controller circuit board.

Table 7 MFC Board Connections

Photo 1 shows the Mass Flow Controller (MFC) circuit board with important connections labelled.



Photo 1 Mass Flow Controller Circuit Board

Table 8 provides a list of connections to the Valve Driver circuit board.

Connector	Label	Pins	Description
J1	LAST	1, 2	Jumper to set last Vial board – install on last vial module only
J2		1-10	Ribbon Connector to from MFC board or upstream Vial Module
J3		1-10	Ribbon Connector to next Vial Module or Final Board
J4-J11		1, 2	Valve Connections

Table 8 Valve Driver Board Connections

Photo 2 shows the Valve Driver circuit board with important connections labelled.



Photo 2 Valve Driver Circuit Board

Table 9 provides a list of connections to the Valve Driver circuit board.

Connector	Label	Pins	Description
J1	MIXING	1, 2	Mixing Valve Connector
J2	FINAL	1, 2	Final Valve Connector

Table 9 Final Valve Board Connections

Photo 3 shows the Final Valve circuit board with important connections labelled.



Photo 3 Final Valve Circuit Board