

Using the LI-6800

Portable Photosynthesis System



Version 1.4

LI-COR[®]

Using the LI-6800 Portable Photosynthesis System

for Bluestem OS™ version 1.4

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The LI-6800 is protected by patents 8,610,072, 8,692,202, and 8,910,506. Additional patents pending in the U.S. and other countries.

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LI-6800 Portable Photosynthesis System						
Component Name	Hazardous Substances or Elements					
	Lead (Pb)	Mercury (Hg)	Cadmium (Cd)	Chromium VI Components (Cr ^{VI})	Polybrominated Biphenyls (PBB)	Polybrominated Diphenyl Ethers (PBDE)
Console	X	O	O	O	O	O
Sensor Head	X	O	O	O	O	O
Chambers and Light Sources	X	O	X	O	O	O
Power Supply	X	O	O	O	O	O
Carrying Case	O	O	O	O	O	O

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O: Indicates that said hazardous substance contained in all of the homogeneous materials for this part is below the limit requirement of GB/T 26572.

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LI-6800便携式光合作用系统						
部件名称	有害物质限制要求					
	铅(Pb)	汞(Hg)	镉(Cd)	六价铬(Cr ^{VI})	多溴联苯(PBB)	多溴二苯醚(PBDE)
主机	X	O	O	O	O	O
分析仪头部	X	O	O	O	O	O
叶室和光源	X	O	X	O	O	O
电源	X	O	O	O	O	O
便携箱	O	O	O	O	O	O





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	The product is marked with this symbol if a Chassis Ground connection is required.
	The product is marked with this symbol to indicate that a direct current (DC) power supply is required.
WARNING	Warnings must be followed carefully to avoid bodily injury.
CAUTION	Cautions must be observed to avoid damage to your equipment.
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Note	Notes contain important information and useful tips on the operation of your equipment.

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A bit of history

The LI-6800 is LI-COR's fourth generation photosynthesis system.

When designing the LI-6800, we endeavored to create a new type of photosynthesis system—one that can push our understanding of photosynthetic processes, but still be accessible to novices and be well-suited to those who are just learning the principles of photosynthesis. Therefore, we automated as many processes as possible and created a modern user interface that is easier to learn and navigate, but we left the platform open so new features can be added, both by us and by advanced users who find a need for programs that are not already built into the device.

In addition, we improved the performance of the gas analyzers and fluorometer, which will lead to noticeable improvements in data quality and may improve your confidence in the results that the instrument provides.

We thank you for your investment, and trust that this instrument will serve you well for years to come. We stand ready to support and help as you put it to work. Welcome to the LI-6800.

About this manual

These days, we hear that people expect instruments that are like smart phones—simple to use even by people who are completely unfamiliar with the underlying principles. In research and discovery, this is both good and bad. You may not want to learn a new programming language to advance in your field, but you may find it difficult to do something novel without a thorough understanding of what the instrument does. So, where to begin?

Fortunately, much of the instrument can be learned simply by using it, so don't be afraid to dig right in. This book is here to introduce the instrument, help you if you get stuck, and help you correct any issues that may arise.

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Section 1.

Introduction to the LI-6800

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Introduction to the LI-6800

The LI-6800 is designed to non-destructively probe processes involved in leaf-level photosynthesis. With the LI-6800, you can obtain real-time measurements on two distinct, but coupled, photosynthetic processes.

First, the instrument measures uptake of carbon dioxide (CO_2) and release of water vapor (H_2O) by the leaf with high precision infrared gas analyzers. Using a mass balance approach based on input and output of CO_2 and H_2O from the leaf cuvette, the LI-6800 computes net CO_2 assimilation (A) and transpiration (E). Additional measured parameters, including leaf temperature, allow the instrument to calculate other important physiological parameters, including stomatal conductance (g_{sw}) and intercellular CO_2 concentration (C_i).

Second, the LI-6800 quantifies fluorescence yield (Φ_F), or re-emission of photons by chlorophyll associated with photosystem II using a detector that is only sensitive to fluorescence generated by a low intensity modulating light source. Fluorescence yield provides information about the light reactions of photosynthesis, including the quantum yield of photosystem II (Φ_{PSII}), electron transport rate (ETR), and many other processes, such as non-photochemical quenching (NPQ). When equipped with a fluorometer, the LI-6800 simultaneously measures leaf-level gas exchange and chlorophyll fluorescence over the same leaf area.

Thus, the LI-6800 provides complete, real-time information about both the light reactions and the carbon reactions of photosynthesis. For a detailed description of the photosynthetic parameters and how they are measured, go to *Leaf-level photosynthesis and its measurement* on page C-1.

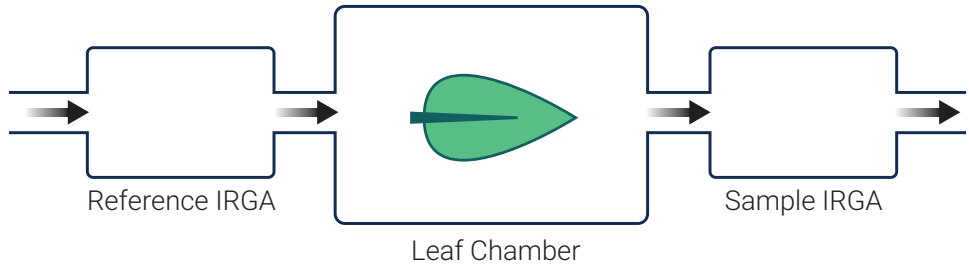
Suggested Instrument Reference: LI-COR, Inc. (2019) LI-6800 Portable Photosynthesis System, Software Version 1.4. LI-COR, Inc. Lincoln, NE.

Suggested Documentation Reference: LI-COR, Inc. (2019) Using the LI-6800 Portable Photosynthesis System; Instruction manual for software version 1.4. LI-COR, Inc. Lincoln, NE.

The gas exchange system

The LI-6800 is an open gas exchange system, which means that the measurements of photosynthesis and transpiration are based on the differences in CO_2 and H_2O

in an air stream that is entering and exiting the leaf cuvette (*Figure 1-1* below).



$$CO_2 \text{ Assimilation} = \frac{\text{Flow} \times (\text{Sample } CO_2 - \text{Reference } CO_2)}{\text{Leaf Area}}$$

$$H_2O \text{ Transpiration} = \frac{\text{Flow} \times (\text{Sample } H_2O - \text{Reference } H_2O)}{\text{Leaf Area}}$$

Figure 1-1. In an open system, photosynthesis and transpiration are computed from the differences in CO_2 and H_2O between air that is affected by the leaf (sample) and air that is not (reference).

The LI-6800 is different from traditional photosynthesis systems for many reasons, but three of the big differences follow: Firstly, the gas analyzers are in the head, right next to the leaf chamber. Secondly, the air stream, which is split so it can flow through both the reference and sample gas analyzers, splits in the sensor head rather than the console. This means that conditioned air doesn't flow through two different tubes until the head. And thirdly, the system supports relatively high flow rates so it can rapidly flush chamber air.

These features eliminate plumbing-related time delays and allow the instrument to respond quickly to leaf changes. For example, if stomata close, the control system immediately detects the drop in water vapor and can compensate. Similarly, a sudden change in light level will cause an immediate change in photosynthetic rate, which will be detected as a change in the CO_2 concentration. The speed of detection is not a function of the system's flow rate, as in traditional systems, since the sample infrared gas analyzers (IRGAs) are very close to the cuvette.

The LI-6800 console provides automated mechanisms for modifying the CO_2 and H_2O concentrations in incoming air (*Figure 1-2* on the next page). The instrument can scrub CO_2 and water vapor from the air stream, and add both to achieve nearly

any concentration desired, making it easy to maintain stable gas concentrations throughout measurements.

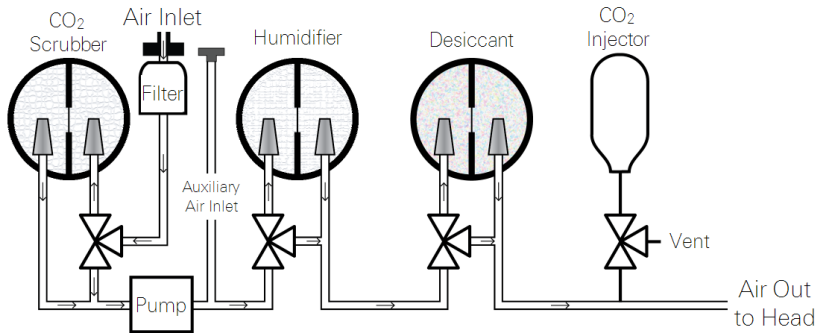


Figure 1-2. LI-6800 flow path through the console. In addition to the indicated components, there are pressure sensors, flow controllers, mixing volumes, proportioning valves, and vents.

The head has a valve system that partitions the flow between the reference and sample gas analyzers. This system of valves also vents chamber air when matching the gas analyzers. The valve mechanism also can restrict the flow to pressurize the chamber.

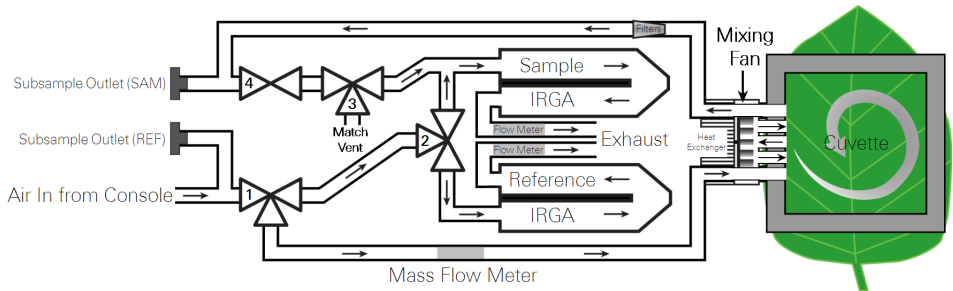
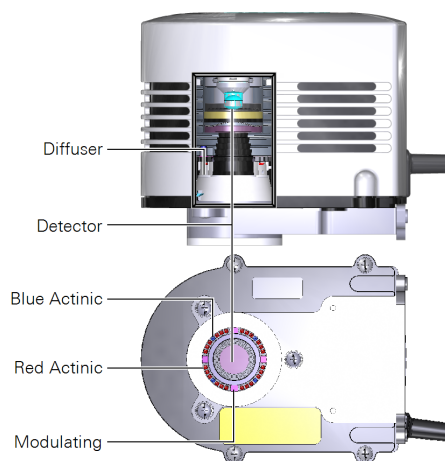


Figure 1-3. LI-6800 head flow control system. 1 indicates the flow partition valve, 2 and 3 are the match valves, and 4 is the chamber pressurization valve.

The fluorescence system

The Multiphase Flash™ Fluorometer is a light source and chamber that can be used for combined gas exchange and fluorescence measurements, or either one alone. The fluorometer uses various colored LEDs to emit light and a detector to measure fluorescence. For an introduction to the fluorometer, see *Multiphase Flash™ Fluorometer: Blinded by the light* on page 3-51.



LED	Center λ	Purpose
Blue Actinic	475 nm	Steady-state <ul style="list-style-type: none"> Maximum steady-state intensity is $1000 \mu\text{mol m}^{-2} \text{s}^{-1}$
Red Actinic	625 nm	Steady-state, saturating flash, and multiphase flash <ul style="list-style-type: none"> Maximum steady-state intensity is $2000 \mu\text{mol m}^{-2} \text{s}^{-1}$ Maximum saturating flash intensity is $16,000 \mu\text{mol m}^{-2} \text{s}^{-1}$
Red Modulating	625 nm	Modulated measuring <ul style="list-style-type: none"> Modulation is $1 \mu\text{s}$ wide pulses with programmable frequency from 1 Hz to 250 kHz Maximum steady-state peak intensity is $100 \mu\text{mol m}^{-2} \text{s}^{-1}$, steady-state average intensity depends on peak intensity setting and modulating frequency setting
Far-Red	735 nm	Steady-state and dark-pulse <ul style="list-style-type: none"> Maximum steady-state intensity is $20 \mu\text{mol m}^{-2} \text{s}^{-1}$

What's what?

If you have just taken delivery of the LI-6800 check the packing list and verify that you have received everything that you ordered. The following pages present the components available for the LI-6800.

Console

Part Number
LI-6860

The console provides the operating system, interface, air conditioning, and data logging. The right side of the console has the power button, head and fluorometer cable connectors, air supply to head connector, and the power cable connector. A 25-pin D connector provides customizable digital and analog inputs and outputs (see *Configuring auxiliary channels* on page 4-46). The accessory connector is to control a second light source.

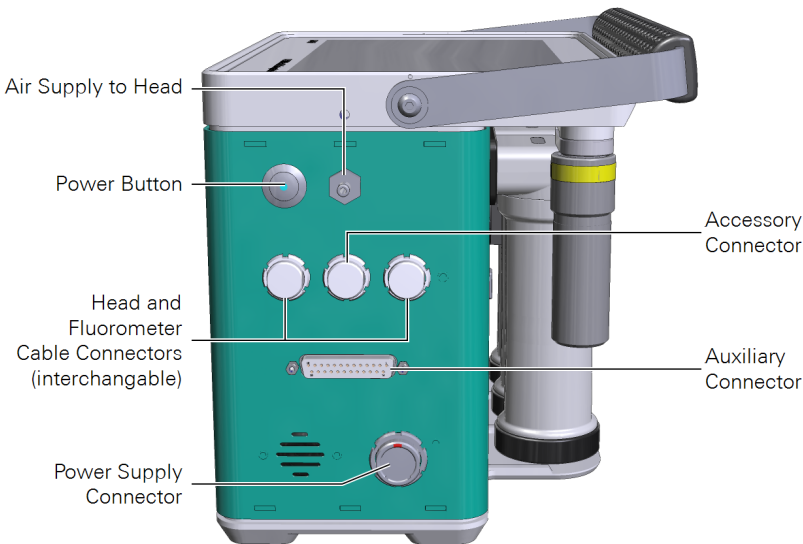


Figure 1-4. Console cable connections and air outlet.

The back (Figure 1-5 on the facing page) of the console has an Ethernet port (LAN) to connect to a network with the Ethernet cable. It has two USB Type A host ports and one USB Type B slave port. You can insert a USB storage device into the host port and transfer files to and from the LI-6800. The USB Type B slave port is not supported currently, but stay tuned for updates.

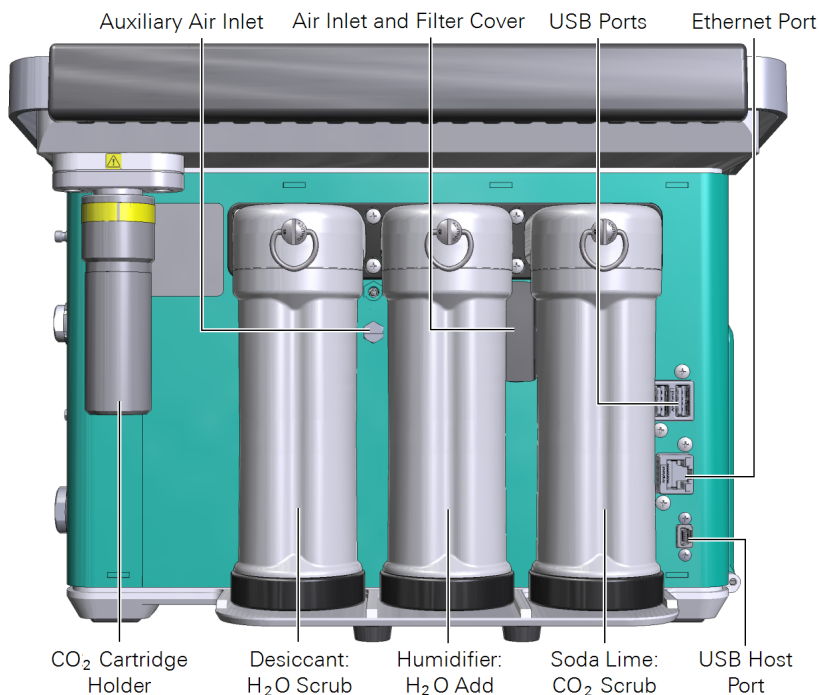


Figure 1-5. Air intakes, conditioning components and Ethernet connection.

The CO₂ scrub column, humidifier column, desiccant column, and CO₂ cartridge holder are on the back of the console. The columns hold materials that condition the air stream—to establish the CO₂ and water vapor concentrations that you specify. The air inlet cover provides access to the intake air filter. The Auxiliary Air Inlet is to inject a custom gas mix, if you want to start with a gas other than ambient air, such as a tank of low O₂ for measurements under non-photorespiratory conditions.

Sensor head

Part Number
LI-6850

The sensor head includes the sample and reference gas analyzers, flow meters, a spring-loaded latching mechanism, a Peltier thermoelectric cooler, and a system of valves. A leaf chamber and optional light source connect to the head. The chambers feature independently-articulated hinges to help form a good seal around unevenly-shaped leaves. Chambers also have three positions:

- **Open:** The lower chamber is all the way open, ready to accept a leaf.
- **Parked:** The first stop on the way to closed. Squeeze the handle until it clicks, then release. This is to adjust the position of a leaf before clamping down on it. Store and ship the head with the chamber in this position.
- **Closed:** This is the position for making measurements. Squeeze the handle firmly and then release it. The chamber will close, compressing the gaskets.

Two stabilizing posts (part number 9868-518) are included with some chambers. The posts should be used for laboratory measurements and removed for field measurements to protect them from damage.

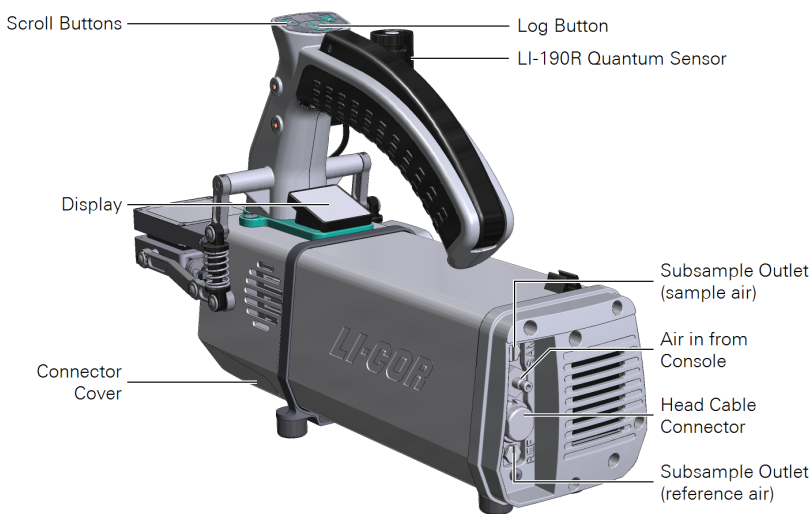


Figure 1-6. The head features the handle and latch mechanism, interchangeable leaf chambers, a display, LI-190R quantum sensor, the log button, and scroll buttons.

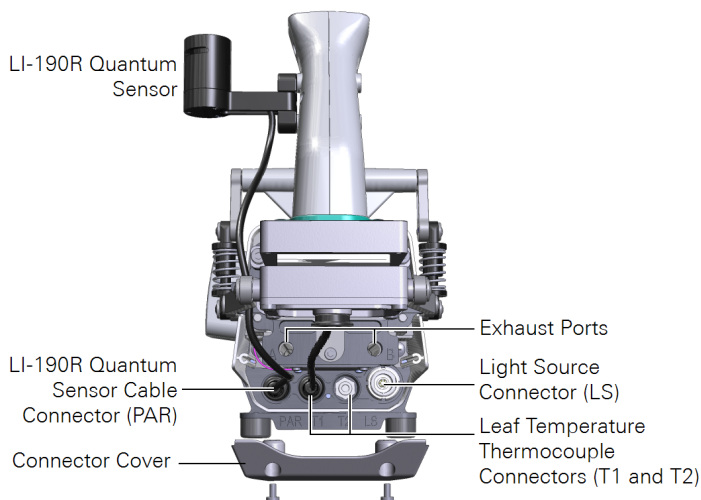


Figure 1-7. The connectors on the front of the head.

Head cable assembly

Part Number
9968-092

The cable assembly consists of a cable for communication and power, and one Beva-line tube for conditioned air. To install the cable, orient the red dot on the cable toward the top of the head or console and press the cable in place.

Accessories

The following components are available for the LI-6800.

External quantum sensor

Part Number
190R-6800

An LI-190R quantum sensor mounts on the sensor head. The entire assembly, including the sensor, mounting bracket, and cable is part number 190R-6800. If reordering, LI-190R is the sensor by itself.

Rechargeable batteries

Part Number
442-11807

The instrument includes three batteries. Batteries are shipped in a partially charged state. They should be fully charged prior to the first use. Leaving them discharged can result in damage to the batteries. One battery will provide approximately six hours of use, but this will vary depending on how you use the system.

Gas conditioning components

Three compounds—a CO₂ scrubber (soda lime), a desiccant (Sorbead®), and humidifier substrate (Stuttgarter Masse) are included with the instrument, as well as several boxes of CO₂ cartridges. In the table below, quantity indicates the number included with the instrument.

Description	Quantity	Part Number
Soda Lime	2 jars	9964-090
Sorbead® Orange CHAMELEON® silica gel beads	0.45 kg bag	622-16951
Pall Stuttgarter Masse	1 jar	9968-165
8-gram CO ₂ cartridges (Box of 25)	3 boxes	9968-227

AC power supply and cord

Part Number
9968-232

The power supply delivers power to the instrument and charges batteries in the console any time the device is plugged into an AC power supply. It is compatible with 110 VAC 60 Hz and 240 VAC 50 Hz power. The power supply can accept a C13 coupler (called an IEC cord). The instrument will include one of the following:

Description	Part Number
For USA, Canada, Mexico, and Japan; NEMA 5-15; Type B	390-00787
For Europe, South America, and Asia; CEE 7/7; Type C	390-01486
For United Kingdom; Type G	390-03089
For Israel; SI32; Type H	390-03059
For Australia; Type I	390-03095



Warning: Use the safety-approved power cord supplied with the instrument. The power supply is equipped with a 3-wire grounding plug. This plug will only fit into a grounded outlet. Do not defeat the purpose of the grounding-type plug. Do not place the power cord where it will be walked on or exposed to water or chemical spills.

Avertissement: Utilisez le cordon d'alimentation à sécurité –approuvée fourni avec cet instrument. Le cordon d'alimentation LI-6800 est équipé d'une fiche à 3 fils avec mise à terre. Cette fiche ne s'adapte qu'à une prise de courant avec mise à terre. Ceci constitue un dispositif de sécurité nécessaire. Ne délaissiez pas le but de ce type de fiche avec mise à terre. Ne laissez pas le LI-6800 où l'on va marcher sur le cordon d'alimentation ou l'exposer aux flaques d'eau ou des produits chimiques.

Power supply adapter

Part Number
9968-242

This optional component allows you to power the LI-6800 from an external power supply, such as a deep cycle battery.

Ethernet cable

Part Number
616-06116

Standard category 5e (Cat5e) network cable to connect the LI-6800 to a computer or network. Length of 2.1 meters (7 feet). The network port is labeled **LAN** on the back of the console.

Caution: Do not use network cables other than the one provided. Other network cables may result in improper electrical performance.

Instrument case

Part Number
9968-222

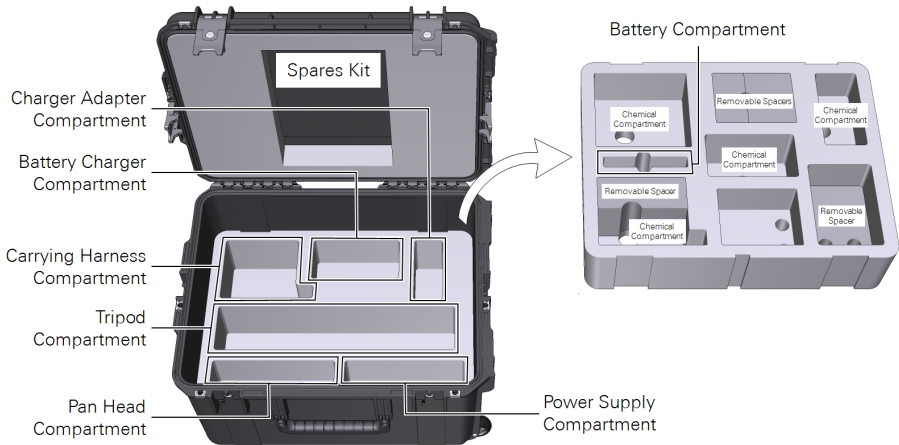
The hard-shell, foam padded case can hold the console, head, and a variety of accessories. Store the instrument in the case when it is not in use. Always use the case to protect the instrument during shipping (see *Preparation for shipping* on page 2-23).



Accessory case

Part Number
9968-223

An accessory case is included with some configurations. The accessory case has two levels. If you are unable to find something, be sure to lift off the upper foam pad to access the second level.



Tripod

Part Number
609-15790

Optional stand for the console or head.

Monopod

Part Number
609-15792

Optional stand for the head.

Panhead

Part Number
609-15791

Optional attachment for the head. Securely holds the head to a tripod or monopod while allowing adjustment to a full range of positions.

Carrying harness

Part Number
9968-221

Optional harness to position the console in front of carrier's torso for easy access to controls. Includes shoulder straps, a waist strap, and hardware.

Single-bay battery charger

Part Number
590-11830

Dock to charge a single battery. Includes power cord for outlet power and 12 VDC adapter for automobile power supplies.

Subsample kit

Part Number
9968-210

The optional subsample kit makes it possible to sample the air stream before and after the leaf chamber. It has hose barb connectors for the air streams, T-fittings, and needle valves.

Description	Quantity	Part Number
Hose barbs (metric; M5×0.8 to 1/8" ID)	4	300-15712
Hose barbs (U.S.; 5/16"-24 × 1/4")	2	300-14479
T-fittings	4	300-07385
Needle valves	2	300-10471

CO₂ tank adapter kit

Part Number
9968-109

An optional adapter to connect a tank of pure CO₂ to the injector. Includes both English and metric hardware.

Zero calibration kit

Part Number
9968-258

The zero calibration kit includes two chemical columns that can be installed (one at a time) on the tube between the console and head to ensure that the air is scrubbed of CO₂ and H₂O during a zero calibration. See *Zeroing with chemicals* on page 8-5 for details.

Internal head chemical bottles

Part Number
9968-094

The internal head chemical bottles ensure that the gas analyzers themselves are free of CO₂ and H₂O vapor. We recommend replacing the head chemicals every three years.

Instrument spare parts kit

Part Number
9968-173

This box contains spare and replacement parts for your LI-6800 console and head. Contact your sales representative for replacement parts. The spare parts kit fits into the top of the accessories case.

Description	Quantity	Part Number
Quick-Connect Coupler	1	300-07125
Bev-a-line Tubing (4.5 m; 15 ft length)	1	222-01824
Low Pressure Chamber Springs	2	9968-240

Description	Quantity	Part Number
Leaf Temperature Thermocouple Assembly	1	9968-245
Console Intake Air Filter with O-rings	1	9968-211
Scrub Tube/Head Filter (10-micron)	12	300-14319
Scrub Tube Filter Seal	6	6368-225
Propafilm™ Kit; Includes double-sided tape and Propafilm	1	9964-151
Flexible Chamber Air Ducts	2	6368-345
O-ring (6×1.5 mm) for Heat Exchanger Tubes	8	192-14314
O-ring (10×1.5 mm) for Leaf Thermocouple	6	192-14435
O-ring (55×2 mm) for Chamber Bottom and Heat Exchanger	3	192-14438
O-ring (44×2 mm) for Chemical Column Cap	6	192-14541
Urethane O-ring AS 005 for CO ₂ cartridge	6	192-14864
Chamber Electronics Gasket	2	6568-327
Leaf Chamber Air Filter Gasket	3	6568-485
Leaf Chamber Air Filter Retention Plate	1	9868-484
Chamber Latch Grommet	2	196-14475
Reflector O-rings	4	192-15947
Reflector Cap Screws	6	151-15742
Chamber Latch Shoulder Screws (M3)	6	146-14474
Bulkhead Screws (M4; 16 mm length; Phillips)	6	150-14431
Chamber Bottom and Air Duct Screws (M3; 6 mm length; Phillips)	6	150-14477
Captive Chamber Mounting Screws	2	144-15344
Mixing Fan Shield and Leaf Chamber Air Filter Screws	6	150-15422

Tools

The spare parts kit includes several tools that are used for maintenance.

Description	Quantity	Part Number
#1 Phillips Screwdriver; 60 mm Blade	1	611-15867
#2 Phillips Screwdriver; 150 mm Blade	1	611-15837
1.5 mm Hex Key	1	611-15953
Large Foam-Tipped Swabs	8	610-05315
Small Foam-Tipped Swabs	8	610-16093
Lint-free Soft Cotton Glove, Small	1 pair	229-10716
Lint-free Soft Cotton Glove, Large	1 pair	229-10715
Anti-static Wrist Strap	1	619-05291
Screen Cleaning Cloth	1	229-16069

Chambers, light sources, and spare parts

Your LI-6800 will include one or more of the following chambers and light sources. Chamber spare parts kits are listed below.

Multiphase Flash™ Fluorometer

Part Number
6800-01A

The Multiphase Flash™ Fluorometer is a combined chamber and light source that induces and measures fluorescence from a leaf. It includes leaf aperture inserts to limit the opening to 6 cm² or 2 cm². See *Installing the fluorometer chamber* on page 5-5 for details. The fluorometer includes a spares kit (part number: 9968-270) with the following items:

Description	Quantity	Part Number
Aperture O-rings; 35×1.5 mm	2	192-16851
O-ring; 6×1.5 mm	1	192-14314
Fluorometer Gasket Kit		9968-208
6 cm ² Aperture AP Gaskets (white upper/lower)	2	6568-512
2 cm ² Aperture AP Gaskets (white upper/lower)	2	6568-566
6 cm ² Aperture Polyethylene Upper Gasket (white)	4	6568-436
6 cm ² Aperture Neoprene Lower Gasket (black)	4	6568-413
2 cm ² Aperture Polyethylene Upper Gasket (white)	4	6564-202
2 cm ² Aperture Neoprene Lower Gasket (black)	4	6564-196

One-sided leaf measurement kit

Part Number
9968-013

The one-sided leaf kit is available to isolate one side of a leaf for one-sided leaf measurements using the fluorometer. The leaf kit is described in licor.com/env/support/LI-6800/topics/one-sided-leaf-measurements.html.

Clear-top leaf chamber (3×3 cm)

Part Number
6800-12A

A small chamber with a transparent Propafilm™ top window. It accommodates one fine wire thermocouple to measure leaf temperature. A GaAsP light sensor measures PAR in the chamber. It is compatible with the 3×3 chamber light source. Aperture inserts limit the opening to 3×3 cm, 2×3 cm, or 1×3 cm. See *Installing the clear-top leaf chamber* on page 5-10 for details. The chamber includes a spares kit (part number 9968-269) with the following items:

Description	Quantity	Part Number
Aperture O-rings	2	192-16680
Light Source Gaskets	3	6568-071
3×3 cm Aperture AP Gaskets (white upper/lower)	2	6568-511
2×3 cm Aperture AP Gaskets (white upper/lower)	2	6568-562
1×3 cm Aperture AP Gaskets (white upper/lower)	2	6568-560
3×3 cm Aperture Polyethylene Upper Gasket (white)	4	6568-429
3×3 cm Aperture Neoprene Lower Gasket (black)	4	6568-384
2×3 cm Aperture Polyethylene Upper Gasket (white)	4	6564-156
2×3 cm Aperture Neoprene Lower Gasket (black)	4	6564-028
1×3 cm Aperture Polyethylene Upper Gasket (white)	4	6568-584
1×3 cm Aperture Neoprene Lower Gasket (black)	4	6568-583

3×3 cm light source

Part Number
6800-02

The 3×3 cm light source mounts directly on top of the clear-top leaf chamber. Includes the light source spares kit (part number 9968-206). 3×3 cm light source gaskets are included with the clear-top chamber spares kit. See *Connecting the 3×3 light source* on page 5-15.

6×6 cm clear-top chamber

Part Number
6800-13

The 6×6 cm chamber features a clear Propafilm top window. It holds two fine wire thermocouples to measure the leaf temperature. An internal, upward facing GaAsP light sensor measures PAR in the chamber. It is compatible with the 6×6 cm light source. See *Installing the 6×6 cm chamber* on page 5-18 for details. The chamber includes a spares kit (part number 9968-207).

Description	Quantity	Part Number
O-Ring AS-154 Viton 75	1	192-12205
Leaf Gasket, 6×6 cm Lower (Black)	3	6568-210
Leaf Gasket, 6×6 cm Upper (White)	6	6568-463
6×6 cm Advanced Polymer Gasket	2	6568-513
Thermocouple Hole Plug	2	6568-523
Propafilm Kit	1	9964-151

6×6 cm light source

Part Number
6800-03

The 6×6 cm light source mounts directly on top of the 6×6 clear-top chamber, Small Plant Chamber, or the Bryophyte chamber. See *Connecting the 6×6 cm light source* on page 5-20 for details. The light source includes one spares kit (part number 9968-249), which includes gaskets and other spare parts:

Description	Quantity	Part Number
6 mm Flat Screwdriver	1	611-16424
Light Source Gasket	1	6568-199
Mounting Screws; M3 × 0.5; 8 mm	4	150-07475
Retaining Washers	2	144-15345
Adapter Kit		9968-189
Adapter Plate	2	9868-209
Mounting Screws; M3 × 0.5; 8 mm	4	150-07475

Sprig adapter blocks

Part Number
9968-271

The Sprig Adapter Blocks attach to the 6×6 cm clear-top chamber, increasing the volume to accommodate sprigs and shoots with whorled leaves. The blocks can be used in ambient lighting conditions and with one or two large light sources. See *Using the Conifer Sprig Kit* on page 5-41 for details. The sprig kit includes the following parts:

Description	Quantity	Part Number
Sprig adapter block with film (upper)	1	9868-586
Sprig adapter block (lower)	1	9868-585
O-rings; AS-154 Viton 175	2	192-12205
Block screws (Panhead M3×30)	8	150-16889
Light source screws (Countersunk M3×30)	1	150-06115

Light source extension cable

Part Number
9968-243

The Light Source Extension Cable is used to connect and operate a light source from the LI-6800 console **ACCESSORY** connector, which makes it possible to operate two light sources simultaneously. See *Light source extension cable* on page 5-32 for details.

Small plant chamber

Part Number
6800-17

The Small Plant Chamber is to measure gas exchange from whole *Arabidopsis thaliana* plants or other small, prostrate plants in 65 mm (2.5 inch) pots or 38 mm (1.5 inch) Conetainers™. The Small Plant Chamber features a transparent Propafilm® cover for measurements under ambient light. It is compatible with the 6×6 cm Light Source, which enables measurements under controlled lighting with any combination of red, green and, blue light. See *Small plant chamber* on page 5-27 for details. The chamber includes a spares kit (part number 9968-231) with the following items:

Description	Quantity	Part Number
Bottom plate for 38 mm Cone-tainers	1	9868-555
38 mm (1.5") Cone-tainers	5	610-09645
Valve assembly for Cone-tainers	2	9968-261
Bottom plate for 65 mm pots	1	9868-554
65 mm (2.5") diameter flower pots	6	610-09646
Valve assembly for 65 mm pots	2	9968-262
Propafilm™ kit with adhesive	1	9964-151
Gasket for 65 mm pots	1	6564-279
AS-150 O-rings	1	192-09262
AS-041 O-rings	1	192-05062
AS-128 O-rings	1	192-12880
6×0.5 mm O-rings	1	192-14314
4-40 Screws	1	122-00004
Large Light Source Chamber Adapter Kit	1	9968-190
Adapter Plate	1	9868-449
Mounting Screws M3×0.5 8 mm	4	150-07475

Bryophyte chamber

Part Number
6800-24

The Bryophyte chamber is used to measure CO₂ and H₂O gas exchange from mosses, hornworts, liverworts, and lichens. The chamber holds loose material in a shallow well during the measurement. It features a clear Propafilm® top for measurements under ambient lighting conditions. It is compatible with the 6×6 cm light source for measurements under controlled lighting with any combination of red, green and, blue light. See *Installing the bryophyte chamber* on page 5-24 for details. The bryophyte chamber includes the following items:

Description	Quantity	Part Number
Mesh Screen (1)	1	197-09025
Short Risers (4)	4	170-04629
Short Riser Screws; M3-8 (4)	4	150-13642
Tall Risers (4)	4	170-16503
Tall Riser Screws; M3-25 (4)	4	150-15454
Chamber Bottom Plate (1)	1	9868-550
Chamber Bottom Screws; M4-12 (4)	4	150-14476
Flat Washers; #4 5/16 (8)	8	167-00155

Custom chamber adapter

Part Number
6800-19

The Custom Chamber Interface is used to attach a custom-made chamber to the LI-6800 gas exchange system. The interface has a power supply connector for one powered peripheral, such as a mixing fan. See *Installing the chamber adapter* on page 5-50 for details. The custom chamber adapter spares kit (part number 9968-254) includes the following items:

Description	Quantity	Part Number
Custom Chamber Mounting Screws	6	150-14476
Swagelok 1/4" Fittings; 7/16 - 20	2	300-16268
Swagelok 1/4" Vinyl Ferrule Set	2	300-05375
Stainless Steel Plugs with O-ring	3	300-16271
1/4" 90° Elbow Connectors; 5/16-24	2	300-16515
Bev-a-line tubing, 1/4" (4.5 meters)	1	220-01824
Swagelok 3/8" Fittings; 7/16 - 20	2	300-16269
Swagelok 3/8" Vinyl Ferrule Set	5	300-16272
Auxiliary Power Cable	1	9968-260
Thermocouple Adapter Cable	1	9968-162
O-ring; 15×1.5 mm	2	192-16274
O-ring; 10×1.5 mm	1	192-14435
O-ring; 6×1.5 mm	1	192-14314

Insect respiration chamber

Part Number
6800-89

The insect respiration chamber is used to measure gas exchange from insects or small mammals. See *Insect respiration chamber* on page 5-47 for details.

Description	Quantity	Part Number
Custom Chamber Adapter	1	9968-182
Sample Column	1	9960-093
Plugs with O-ring	3	300-16271
Swagelok Ferrule	5	300-16272
Swagelok Tube Fitting	2	300-16268
Swagelok ¼" Vinyl Ferrule Set	2	300-05375
Bev-a-line tubing, ¼" (4.5 meters)	1	220-01824
Quick-connect union	2	300-03123
Filter paper with double-sided tape		

Chamber springs

Three types of chamber closure springs are available to close chambers with different levels of tension. In general, the springs that are installed on the chamber at the factory will work with that chamber under most circumstances. If, however, you are measuring delicate plant material, you may want to use the gentler springs. Be aware that the low pressure springs may result in more leaks around the leaf chamber gaskets.



Figure 1-8. Low pressure springs (white; part number 9968-240) are included in the instrument spares kit. Use these springs if measuring delicate leaves that are damaged by the standard springs. Medium pressure springs (silver; part number 9968-131) are suitable for most leaves. High pressure springs (black; part number 9968-239) are used with the 6×6 cm chamber.

Soil CO₂ flux chamber

Part Number
6800-09

The LI-6800 can measure CO₂ efflux from soils when equipped with the soil chamber. The soil chamber is described in licor.com/documents/zoespy2szykxb4ei3xs6kgjaoir1v7ye

Section 2.

Assembling the LI-6800

Putting it all together

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Assembling the LI-6800

This chapter guides you through the assembly and preparations necessary to operate the LI-6800, as well as the preparations that you should take prior to storing the instrument for extended periods.

Preparing the instrument

If you are unpacking the LI-6800 for the first time, be sure to remove any shipping and packaging materials from the instrument. You'll want to gather the console, head, and cable. You'll also want one CO₂ cartridge, desiccant, soda lime, and Pall Stuttgarter Masse. Go through the following pages to put everything together.

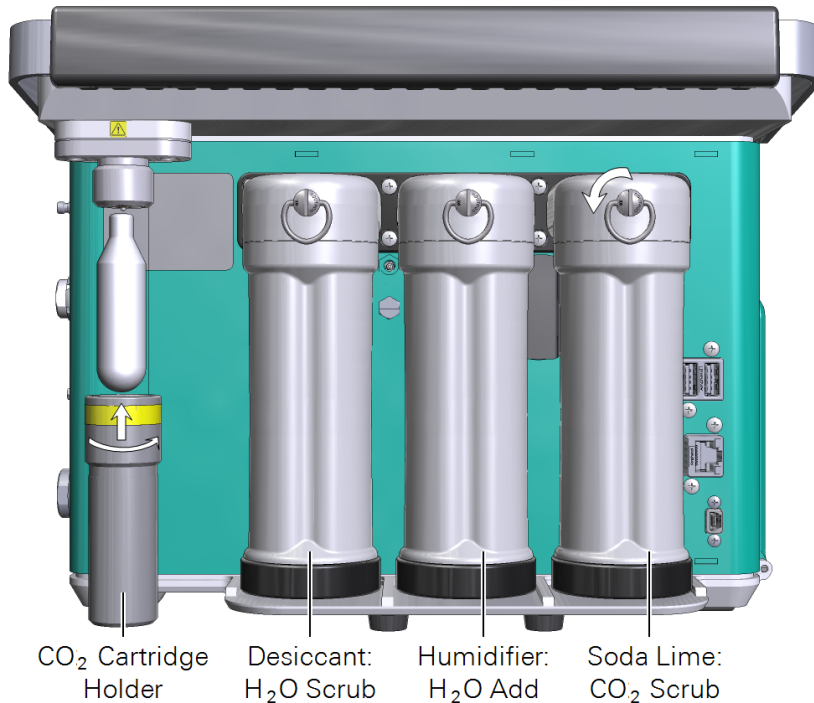


Figure 2-1. The LI-6800 uses an 8-gram CO₂ cartridge for the CO₂ supply. Chemical columns are removed by turning the clasp ¼ turn counterclockwise. See Filling the chemical columns on page 2-5 for tips.

Carbon dioxide injector

The CO₂ injector consists of an internal controller and an external connection that accepts an 8-gram cartridge. One cartridge will last about 8 hours from the time it is pierced (regardless of whether you use it or not). You can use the optional tank adapter kit (part number 9968-109) rather than cartridges for a larger supply. When you install a new cartridge, you can record this in the instrument (see *Fresh cartridge or soda lime?* on page 3-29).



Warning: *The CO₂ cartridges contain high-pressure CO₂. Follow the handling precautions provided by the cartridge manufacturer. Use extreme caution if removing the cartridge while it is pressurized. Doing so will cause the compressed gas to vent quickly. If you need to remove a charged CO₂ cartridge for any reason, slowly loosen the cartridge holder until the gas vents, then the cartridge can be removed. The cartridge may become extremely cold, so be careful. Keep out of sunlight, and under 50 °C. Never dispose of a full cartridge. Keep out of reach of children.*

Avertissement: *Les cylindres de CO₂ contiennent du CO₂ sous haute pression. Suivez les précautions sur la manutention fournies par le fabricant du cylindre. Soyez extrêmement prudent si vous enlevez le cylindre lorsqu'il est sous pression. Faisant cela, vous feriez dégager rapidement le gaz comprimé. Si vous devez à tout prix enlever un cylindre de CO₂ chargé, desserrez lentement le support de la cartouche jusqu'à ce que le gaz se dégage, et alors le cylindre peut être enlevé. Le cylindre peut devenir extrêmement froid, soyez donc prudent. A garder à l'ombre et en dessous de 50 °C. Ne jamais jeter une cartouche pleine. A garder hors de portée des enfants.*

8-gram cartridges

Insert the cartridge, large end first, into the holder (*Figure 2-1* on the previous page) and turn the holder clockwise to tighten it. *Tighten it until you encounter resistance, then quickly twist the holder about ½ turn further to pierce the tip of the cartridge.*

Caution: If you buy CO₂ cartridges from a supplier other than LI-COR, select oil-free cartridges. If oil is drawn into the CO₂ injector, the instrument will have to make a trip back to LI-COR for repairs. We recommend ISI brand: <http://www.isi.com/en/culinary/products/isi-chargers/soda-chargers/>. Store cartridges with the tip pointing up.

Tank adapter kit

A CO₂ tank adapter kit (part number 9968-109) can replace the 8-gram cartridge adapter. It is useful in situations where a tank or other source of pure CO₂ is available. The adapter is designed for pressures from 500 to 2000 kPa (73 to 290 psi).

Tank Adapter Components	Quantity	Part Number
Injector Plate	1	9868-228
Screws (M3; 8 mm length; Phillips)	6	150-14419
Injector O-ring (AS-106)	3	192-14863
Copper Fitting for 4 mm Tube	1	300-14454
Stainless Steel Fitting for 1/8" Tube	1	300-14455



Warning: Remove the CO₂ cartridge before attempting to remove the injector assembly. Failure to do so can cause the cartridge to discharge quickly, which may lead to personal injury.

Avertissement: Enlevez le cylindre de CO₂ avant d'enlever l'assemblage d'injecteur, sans quoi le cylindre peut rapidement se décharger, ce qui peut résulter en une blessure personnelle.

- 1 Remove the cartridge injector assembly.

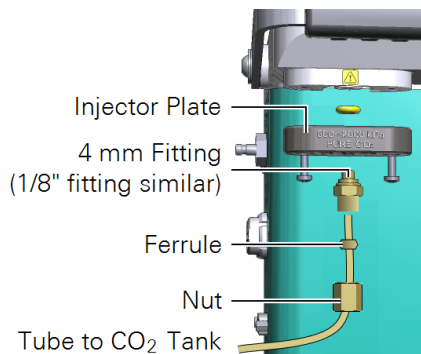
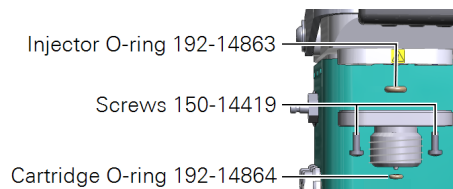
It is held on by two screws.

- 2 Assemble the adapter kit.

The adapter kit has two fittings: A stainless steel fitting for 1/8" tubes and a copper fitting for 4 mm tubes. Install the adapter that works for you.

- 3 Test the injector.

Adjust the pressure regulator to about 1000 kPa (150 psi). Run the system test under **Start Up > System Tests > Soda Lime-CO₂** to verify that the injector is working as expected.



Filling the chemical columns

Three chemical columns on the back of the instrument hold chemicals that condition the air stream. The columns—which are desiccant, humidifier, and soda lime—should remain attached to the console at all times, except when changing the chemicals. During transportation or storage, remove the humidifier column to ensure that water does not enter the console.

1 Inspect the filters.

There are two filters in the top of each column. Be sure they are in good condition and free of contaminants. Replace if needed (see *Chemical columns* on page 8-26).

2 Fill at least to the threads.

Leave a little room to move when shaken. Then you can easily break up channeling, which is the tendency for air to find a dominant passage through the chemicals, thereby saturating the chemicals unevenly in the column.

3 Threads and rim clean and dry.

Wipe the threads, rim, and O-ring with a moist paper towel to remove any solid material. This will help keep the column and cap from being damaged, and ensure that the columns do not develop leaks.

4 Cap O-ring slightly compressed.

Tighten the cap snugly, until the O-ring in the cap is slightly compressed.

5 Clip it in place.

The attachment clip slides into the manifold. Press in and turn the clasp $\frac{1}{4}$ turn clockwise to attach.



The desiccant column

The desiccant column holds Sorbead[®] Orange CHAMELEON[®] or Drierite[®] desiccant. The desiccant removes water vapor from the air. When filling the column for the first time or replacing spent desiccant, you can record this in the instrument (see *Fresh Desiccant or wet Stuttgarter Masse?* on page 3-25).

LI-COR now recommends Sorbead Orange CHAMELEON for the LI-6800 because indicating Drierite has been banned in some regions. Sorbead is an indicating aluminate silicate gel. However, indicating and non-indicating Drierite remain suitable options for the desiccant.

To regenerate Drierite or Sorbead

Both indicating Drierite and Sorbead can be regenerated. Indicating Drierite is blue when dry and pink when it needs to be regenerated. Sorbead Orange CHAMELEON is orange when dry and colorless when it needs to be regenerated.



- 1 Place in an oven.
 - Drierite: 60 minutes at 210 °C (425 °F).
 - Sorbead: 60 to 120 minutes at 120 °C (248 °F).

Preheat the oven. Spread a single layer of the granules in a shallow pan and heat for an hour. Less heat for a longer time will not work, nor will more heat for less time.

- 2 Seal in a glass container while still hot.

After the dry color is restored (blue for Drierite, orange for Sorbead), place the hot regenerated material into a glass container and close the lid while it is still hot. Do not place hot beads into a plastic jar. Store the regenerated desiccant in a sealed glass jar. Avoid plastic containers.

Note: The color of the indicating Drierite may become less distinct after successive regenerations. If it turns black, you've overheated it. Sorbead beads may break over time but the performance is not affected.

The humidifier column

The LI-6800 humidifies the air stream by drawing air through wet Stuttgarter Masse (Pall Corporation). Stuttgarter Masse is a porous ceramic substrate that holds liquid water. Water evaporates into the air stream as air passes through the substrate. When you wet the Stuttgarter Masse, you can record this in the instrument (see *Water vapor control: Who'll stop the rain* on page 3-24).

Caution: Do not add too much water to the column! If there is standing water in the column, if the air creates bubbles as it passes through the column, or if the granules form a clumpy, aqueous mess, it has too much. This may damage the instrument, requiring costly repairs. If there is too much water, pour the substrate onto a dry paper towel to soak up the extra water. After allowing the towel to absorb some water, pour the substrate back into the column.



Warning: *Stuttgarter Masse is safe and nontoxic. Nevertheless, do not breathe dust. If inhaled, move to fresh air and continue breathing normally.*

The safe way

Here we describe a way to moisten the Stuttgarter Masse that will dependably saturate the substrate without adding too much water. You can follow this procedure with new (completely dry) or used (already moist) Stuttgarter Masse substrate.

- 1** Pour about 150 ml of Stuttgarter Masse into a beaker.
- 2** Add enough water to cover the substrate. We recommend filtered water. Tap water will work, but minerals may build up in the substrate.
- 3** Pour off the excess water—through your fingers or a screen—so the moistened substrate remains in the beaker.
- 4** Dump the moistened substrate onto a stack of 3 or 4 paper towels. Allow the towels to soak up some of the water from the substrate.
- 5** Pour the moistened substrate into the humidifier column.
- 6** Wipe the threads on the cap and the column to remove any dry granules, then tighten the cap.

The fast way

This method may save a few minutes over the previous method, at the risk of over-wetting the substrate. It can be used to moisten new or used substrate.

- 1 Fill the humidifier column with Stuttgarter Masse.
- 2 If the substrate is completely dry, add a total of 30 ml water, distributed over both sides of the column.

We recommend softened, distilled, deionized, or reverse-osmosis filtered water. Avoid using hard water because minerals may accumulate in the substrate.



- 3 Wipe the threads on the cap and the column to remove any dry granules, then tighten the cap.

Re-wetting the substrate

When to add water? Add water when you use new Stuttgarter Masse, and periodically afterward. This is how to tell if the substrate needs more water:

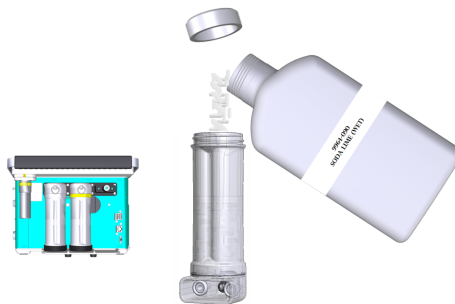
- Look at the substrate. When it is wet, it will have a slightly grayish-white color. When it is dry, it will be pure white. Add water if it is visibly dry.
- Add water if the instrument fails the humidity test under **Start Up > System Tests > Drier-Humidifier**.
- Add water if there are no water droplets visible on the column walls.

You can either follow the procedure in *The safe way* on the previous page, or add 10 to 15 ml of water total to re-wet the substrate. Gently shake the column to distribute the water evenly.

The carbon dioxide scrub column

The CO₂ scrub column holds soda lime (Ca(OH)₂, H₂O, NaOH, and KOH). Indicating soda lime changes from white to violet as it becomes discharged. How often the soda lime needs to be replaced depends upon how much CO₂ has been removed from the air stream. Loss of capacity to scrub CO₂ can be recognized by an inability to reduce the CO₂ mole fraction to near zero and hold it there.

When you add new soda lime, you can record this in the instrument (see *Fresh cartridge or soda lime?* on page 3-29).



Warning: Soda lime causes severe burns when wet. Wear suitable gloves and eye and face protection. Keep out of reach of children. In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.

Avertissement: La Soude caustique cause de graves brûlures quand elle est humide. Portez des gants et des protège-yeux et face appropriés. A garder hors de portée des enfants. En cas de contact avec les yeux, rincez immédiatement avec beaucoup d'eau et consultez un médecin.

To regenerate soda lime

When used in a dry environment, the scrubbing capacity of soda lime will be greatly diminished. Also, as the soda lime becomes saturated it will have reduced scrubbing capacity. Rather than replacing it with fresh soda lime, you can extend it a bit by adding a small amount of water (about 10 ml) to the soda lime tube.

When adding water to soda lime, do it slowly, letting the chemical absorb the liquid. Then, hold the tube horizontally and shake it to distribute the moistened (and clumpy) pellets. Avoid adding too much water; if liquid water gets out of the tube during operation, downstream parts will become corroded.

Installing the LI-190R on the LI-6800

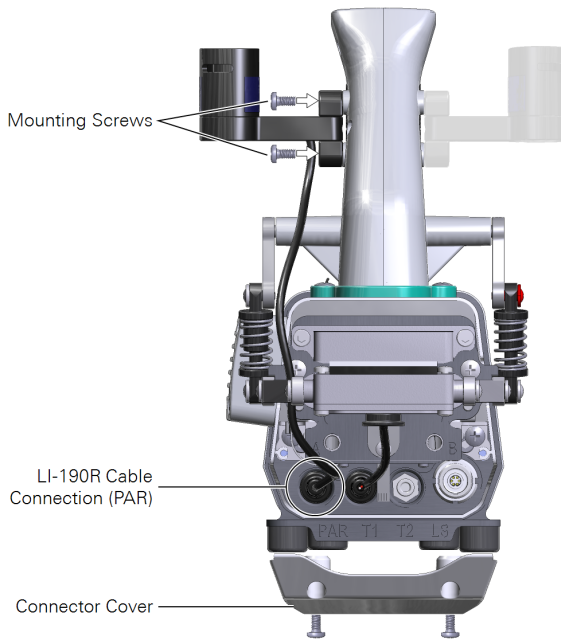
To use the LI-190R-6800 Quantum Sensor, you'll need to install it on the LI-6800 head and enter its calibration information into the software.

Remove the protective red cap from the light sensor before using it. The cap should NOT be in place during measurements. It is to protect the sensor during shipping and storage.

Remove cap before measurements



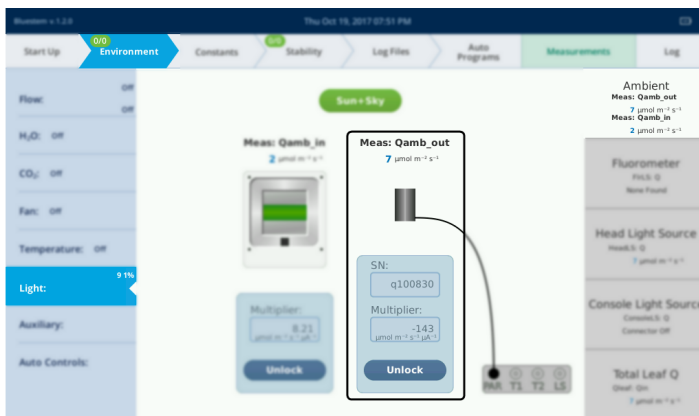
- 1 Install two screws to attach the sensor to the handle.
The quantum sensor can be mounted on either side of the sensor head.



- 2 Remove the head cable connector cover.
- 3 Plug the cable into the connection labeled PAR, then reinstall the connection cover.
- 4 Enter the calibration data.

The LI-190R calibration certificate is included with the instrument's documents. You can also retrieve it from www.licor.com/env/support.

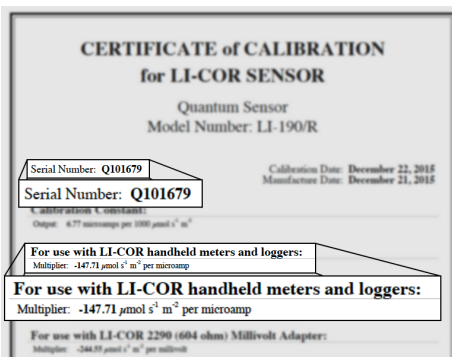
- 5 Power on the LI-6800.
- 6 Go to **Environment > Light > Ambient**.



You're interested in the settings under **Meas: Qamb_out**. Tap **Unlock** to access the controls.

- 7 Enter the serial number and multiplier.

The unique multiplier for the light sensor is on the calibration sheet under **For use with LI-COR handheld meters and loggers**. It will be a negative number (probably different from the value in the example below).



- 8 Verify the light sensor response.

Under **Start Up > Data Dictionary**, select the variable **Meas > Qamb_out**. If you're in an office, expect a value less than 10. Cover the light sensor with your hand. The value should drop in response to the shade.

Connecting the head cable

The cable and tube bundle connects the console and head. The cable is symmetric—either end of the cable can connect to either the console or head.

Caution: Do not connect or disconnect the head cable while the instrument is powered on. Doing so can cause damage to the internal circuitry, leading to costly repairs. Save money and your reputation: *Always power off the console when installing or removing the head cable.*

One end of the cable connects to either the **HEAD (1)** or **HEAD (2)** connector (either one is suitable). The other end connects to the connector on the head. Be sure that the *red dot on the cable connector faces up*, then press the cable into the connector.

The tube connects to the air outlet labeled **AIR OUT** on the console and the connector on the head. To remove the cable or tube, grasp the collar and pull straight out.

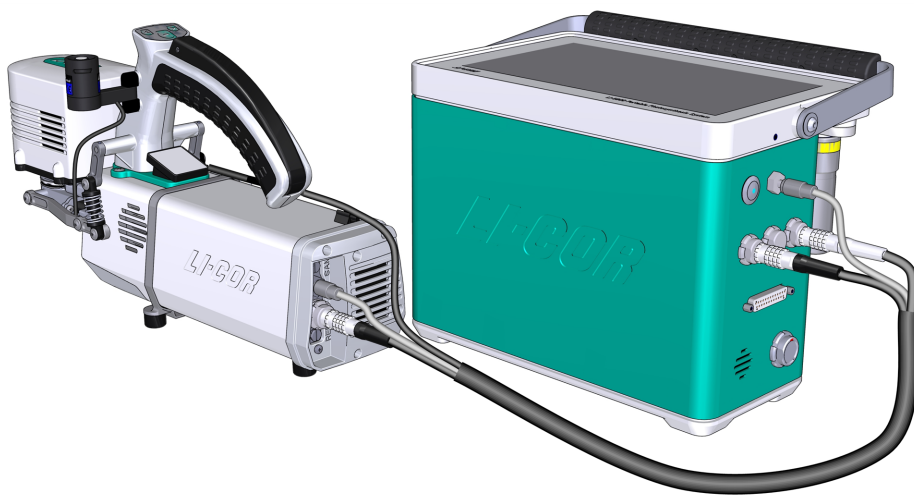


Figure 2-2. Cable and air supply connections on the head and console. Red dots face up.

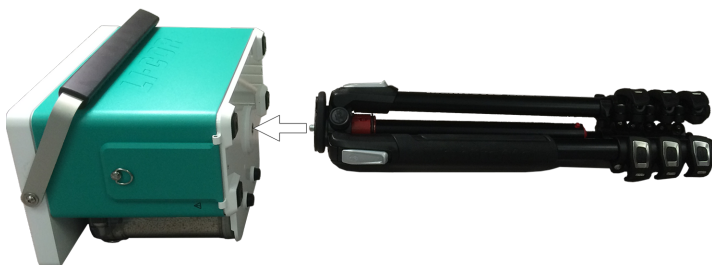
Using the tripod, monopod, panhead, and harness

The tripod, monopod, panhead and harness available from LI-COR are durable and suitable for field work. But, you can mount the head and console on nearly any camera tripod or monopod (3/8-16 threads). *We recommend using a sturdy tripod that has a broad stance.*

Photosynthesis system tripod

The LI-6800 may include a tripod (part number 609-15790), depending upon the configuration ordered. The tripod screws to the bottom of the console or head.

Caution: Take some time to get acquainted with the tripod. Read all of the manufacturer's instructions that are provided with it. Take extra care to set it up securely, as an out-of-balance load may topple over.



- 1 Remove the threaded cap from the top of the tripod.
- 2 With the tripod legs collapsed, screw it into the bottom of the console. Or, attach the panhead to the tripod for the head.
- 3 Extend the legs.

Each leg has three clasps. To extend a leg, open a clasp, pull the leg to the desired length, and then close the clasp to lock the leg in position.



4 Expand the stance.

The stance can be expanded to the first stop without pressing the button. Press it to expand the stance further. A broad stance is more stable than a narrow one. So, extend the legs as far as practical and expand the stance.



5 Extend the mast.

The mast can extend vertically or horizontally. To move the mast into a horizontal position, extend the mast fully in the vertical direction. Press the button on the bottom of the mast while pulling to extend it further so that the mast can rotate to a horizontal position. **Important:** *Be sure that the tripod is stable! Do not let it fall over.* Read the instructions provided with the tripod.



Panhead

The panhead gives the ability to adjust the position of the sensor head to nearly any orientation. The panhead (optional; part number 609-15791) includes a detailed instruction sheet.

Caution: Read all of the manufacturer's instructions that are provided with the panhead. Failure to follow the instructions can lead to improper use of the panhead.

When you use the panhead:

- 1 Remove the fabric strap that is around the handle. Squeeze the handle, then remove the strap.



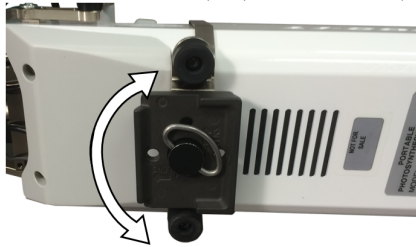
- 2 Detach the mounting plate.

Hold the safety latch away from the panhead, then rotate the latch about 90° until the mounting plate is loose. The latch will lock in the open position.



3 Attach the mounting plate to the head.

The base can be in nearly any orientation, except...



...this one, because the feet interfere with the panhead.



Caution: Do not remove the feet from the head. They are required to reduce shock.

- 4** Press the base plate into place on the panhead. The latch will snap closed.
Carefully check to be sure the head is firmly attached.

Monopod

Your instrument may also include a monopod (part number 609-15792). The monopod is a single-foot platform used to support the head during a measurement¹.

The panhead mounts to the head to provide full rotation of the head on the horizontal and vertical axes. This enables you to easily position the head at nearly any angle during measurements. It can be installed on either the monopod or tripod. Refer to the manufacturer's instructions for details.

Harness

The optional harness (part number 9968-221) positions the console in front of the holder's torso, providing easy access to the controls. The harness kit includes a shoulder strap, waist strap, D-rings (part number 193-15728), and screws (part number 150-15736; M5, 10 mm length) to attach the D-rings. The screws have a thread locking compound applied. If you provide your own screw, apply a thread lock to it.

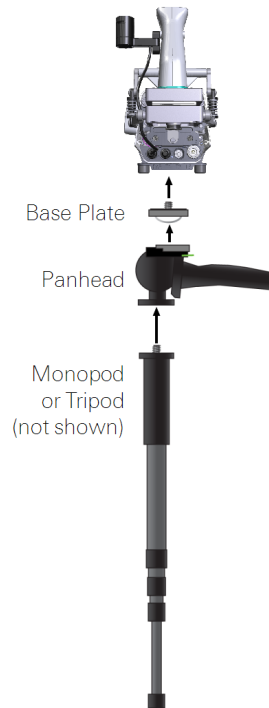


Figure 2-3. Panhead and monopod. The base plate is part of the panhead. The sensor head mounts to the plate and panhead, which mounts atop the monopod or a tripod.

¹Some call this combination the cephalopod.

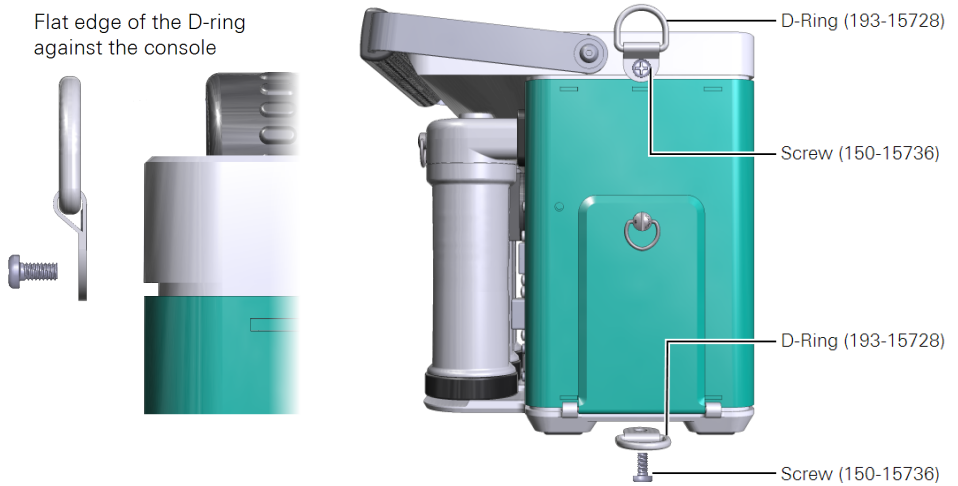
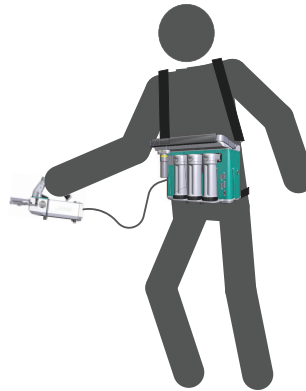


Figure 2-4. Install the D-rings with the flat edge against the console.

The shoulder straps attach to the upper D-rings and the waist strap to the lower D-rings. The waist strap clasps on the wearer's back side.



Powering the LI-6800

The LI-6800 can be powered from the included batteries, from the power adapter, or from a user-supplied power source.

Power adapter

The AC power supply is compatible with 100-240 VAC power sources of either 50 or 60 Hz. The LI-6800 can be powered from the adapter. The adapter also charges the batteries that are installed in the instrument. Use the safety-approved power cord supplied with the instrument.

Batteries

Three batteries (part number 442-11807) are provided with the instrument. Two batteries can be installed behind the battery cover, but only one battery needs to be installed to power the device.



Figure 2-5. Insert the batteries with the groove toward the columns and the tab away from the columns.

The batteries are 14.4 V lithium ion battery packs with 6.8 amp-hour capacity. Two batteries should provide up to 12 hours of operation, depending upon the configuration. Batteries will be drawn down quickly when you run the light source at high intensities and control temperature far from ambient. Keep this in mind when you plan your field work.

Charging batteries in the console

When batteries (one or two) are installed in the console, they will charge any time the LI-6800 is powered with the adapter. The LI-6800 supports hot-swapping, meaning that you can take one battery out and replace it with a freshly charged battery and measurements will not be interrupted—so long as there is a charged battery installed in the instrument.

The console displays the remaining charge, either as a percent of time. The batteries also have a **Charge Gauge** indicator, which shows the charge level of the battery.

Charge Gauge



Charging batteries with the external charger

The single-bay charger (part number 590-11830; Inspired Energy CH7000A) can charge one battery at a time. To use the charger, plug it into a wall socket and place a battery in the dock. The charger is compatible with 100 to 240 VAC power at 50 to 60 Hz. When charging, you'll see a blinking LED. When fully charged, you'll see a solid green LED.

Note: You can use the automobile adapter kit to charge the battery from an automobile. Leave the vehicle running when charging the LI-6800 battery to prevent excessive discharge of the car battery.



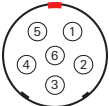
Warning: Charge batteries only with a SMBUS compliant level 2 or 3 charger. Do not heat above 80 °C. Do not open battery, dispose of in fire, or short circuit—may ignite, explode, leak, or get hot, causing personal injury. Replace battery with same part number only. Use of another battery may present a risk of fire or explosion. Keep away from children.

Avertissement: Ne rechargez les batteries qu'avec un chargeur conforme au SMBUS niveau 2 ou 3. Ne chauffez pas au-dessus de 80 °C. Ne pas ouvrir la batterie, ni ne la jeter au feu ou court-circuiter—elle peut mettre à feu, exploser, couler, ou s'échauffer, causant des blessures personnelles. Remplacez les batteries seulement avec le même numéro de pièce. L'utilisation d'une autre batterie peut présenter un risque, ou un incendie ou une explosion. A garder hors de portée des enfants.

External power source

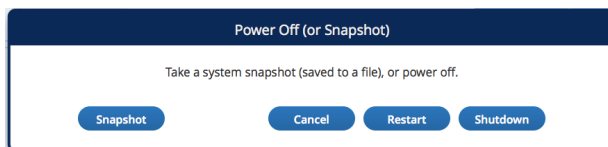
You can power the LI-6800 with any 12 to 18 VDC power source. A deep-cycle marine battery with at least a 20 amp-hour capacity will work fine. To power it, you'll need the power cable adapter (part number 9968-242).

Table 2-1. Power inputs for the auxiliary power supply connector.

Connector	Pins	Description
	1 and 5	⎓ 24 VDC, 8.4 A; LI-COR power source
	3 and 4	⎓ 12-18 VDC, 14.3 A; custom power source
	2 and 6	Ground

Power on and off

To turn on the LI-6800 briefly press the power button. To turn it off, press and release the power button, or go to **Start Up > Standby/Power Off > Power Off**. You'll be prompted to cancel, restart, shutdown, or snapshot. If, for some reason the instrument will not shut off through the normal means, press and hold the power button for 5 seconds to do a hard shutoff.



In the **Power Off** window, you have the option to take a system **Snapshot**. A snapshot is a file that shows the state of the system at that moment. This is helpful for troubleshooting and diagnostic purposes when the system seems weird. It is also a quick way to capture some strange special moment. See *Snapshots* on page 6-30 for details about the snapshot.

If you are logging to an Excel file, the LI-6800 writes the Excel file after you close the log file. The LI-6800 will warn you that it is writing the Excel file and wait to shut down until the file is written. If you press Restart or Shutdown anyway, the Excel file will likely be corrupted, so it is best to wait until the file is written.

Putting it away for the season? See *Storing the LI-6800* below.

Storing the LI-6800

The instrument can be stored for a few weeks without any special preparation. For short-term storage, park the chamber (not open, but not closed).

For longer-term storage (a month or more), we recommend that you remove the batteries from the console and clean and dry the instrument with a soft damp (not

dripping wet) cloth and mild detergent if needed. Clean the screen with common glass cleaners. Spray the cloth, not the glass.

The LI-6800 should always be stored in the case. The instrument storage temperature range is -45 to 50 °C, <85% relative humidity. The instrument will draw a small current when it is off. A fully charged battery will drain in about 7 months in an instrument that is off.

You can store the instrument with the desiccant and soda lime in the columns; however, you may need to replace the chemicals prior to using it again. Be sure the instrument stays below 50 °C during storage. We recommend the following before storing or shipping the instrument:

- Remove the CO₂ cylinder. Vent the gas to depressurize the cylinder, and then remove it. This prevents CO₂ adsorption to internal console components. This is especially important when storing the instrument in the closed case, which will trap the vented CO₂ gas.
- Empty the Stuttgarter Masse column or remove it from the console. This will ensure that the valve manifold stays free of water in the event that the case becomes inverted.
- Remove any plant material from the leaf chamber.
- Park the chamber (do not close it) to prevent compressing the gaskets.

Battery storage

Store the batteries with a 30-50% charge. Replenish the charge every 6 months of storage. The batteries do not have a "memory," and there is no need to fully discharge a battery routinely.

The battery pack is not user serviceable. If it exceeds a temperature or load threshold, a non-resettable fuse will open, rendering the battery non-functional. If the batteries become excessively discharged in storage, the charger will supply a 3-minute wake-up charge in an attempt to activate the electronics.

Battery removal

To remove the batteries, open the battery cover (see *Figure 2-5* on page 2-19). Grasp the tab on the battery and pull.



Battery disposal

Lithium ion batteries must be disposed of safely. The batteries may cause environmental damage or damage to human health if they are disposed of improperly. Do not dispose of the batteries in unsorted municipal waste or an incinerator. Many localities have battery recycling facilities that will accept lithium ion batteries. Check your local regulations to determine how to dispose of the batteries.

This product uses CR Lithium coin cells to preserve memory. Perchlorate material: Special handling may apply. In the U.S. State of California, see www.dtsc.ca.gov/hazardouswaste/perchlorate.

Preparation for shipping

When you need to ship the LI-6800, we recommend that you observe the following tips:

- Empty the chemical columns.
This will reduce shipping weight and strain on the console; and ensure that water from the humidifier column does not enter the console.
- Remove the Stuttgarter Masse column and store it in the case.
- Remove the CO₂ cartridge from the holder.
Do this even if it is empty. If you remove a charged cartridge, it will be noisy when the gas vents.
- Leave the chamber parked.
This will prevent the leaf chamber gaskets from becoming compressed.
- Console in the case.
Disconnect the cables and tube. The scrub tubes should be toward the back of the case and handle folded down.
- Head placed in the case.
Disconnect the head cable and tube. The chamber can stay attached to the head.
- Secure the case latches.
Use a secondary closure method to ensure that the case stays closed. For example, use zip ties to secure the lid.

Shipping to LI-COR?

In addition to everything mentioned in *Preparation for shipping* on the previous page, be sure to:

- Contact us for an RMA (Return Materials Authorization) number so we know to expect your instrument.
 - Phone: 402-467-3576
 - In the U.S and Canada: 800-447-3576
 - Email: envsupport@licor.com
 - Or contact your local representative.
- Be sure to include the console, the head, the head cable, and any components that need a calibration, such as light sources and the light sensor.
- Don't bother sending batteries, other cables, power supplies, battery chargers, or extra chambers unless you want us to examine them.
- Back up your data.

We do not save a copy of the data before we start working on the instrument, nor do we delete files from a customer's instrument. But if you send us a console with priceless data stored on it and nowhere else, then you may be in for some abnormally bad luck. So please, send us only files you can afford to lose.

Section 3.

Guided tours

Learning how to make it work

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Configuration	3-8	Logging options	3-83
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Leaf temperature constants	3-76		
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Guided tours

The purpose of this section is to help you gain familiarity with the LI-6800 interface. We do this with a number of guided tours. We recommend that you follow along on your instrument. You won't need plant material for these tours. That will come in a later section.

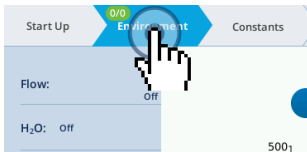
Overview of the operating system

Here are a few things to remember as you use the instrument.

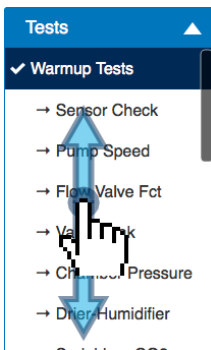
Touch gestures

The LI-6800 touch screen supports tap and swipe gestures, which can be initiated with a finger or touch-screen stylus.

Note: Do not press with excessive force or with sharp objects, such as mechanical pencils. Doing so can damage the display. The touch screen will not respond to wet hands or hands that are covered with gloves.



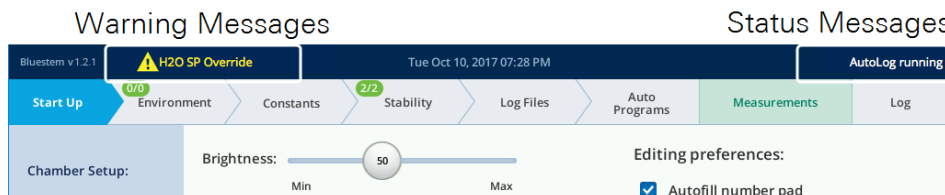
To **Tap**, gently touch the display and then quickly remove your finger from the screen. This action will select an option, open a menu, activate a button, or open a text-entry field.



To **Swipe**, touch your finger to the display, then move the finger in the direction that you want to scroll. This action is available for menus and screens that have scroll bars visible.

Messages: Warnings and status indicators

As you use the instrument, make a habit of training your eyes toward the upper left of the display. Here the instrument will display caution and warning **Messages**. In the upper right, the instrument will display **Status** indicators.



It is wise to become familiar with the messages, what they indicate, and whether any corrective action is necessary on your part. For example, a message like **Flow_s Low** may indicate a leak, configuration issue, or simply that the chamber is open. See *Messages* on page 7-32 for details.

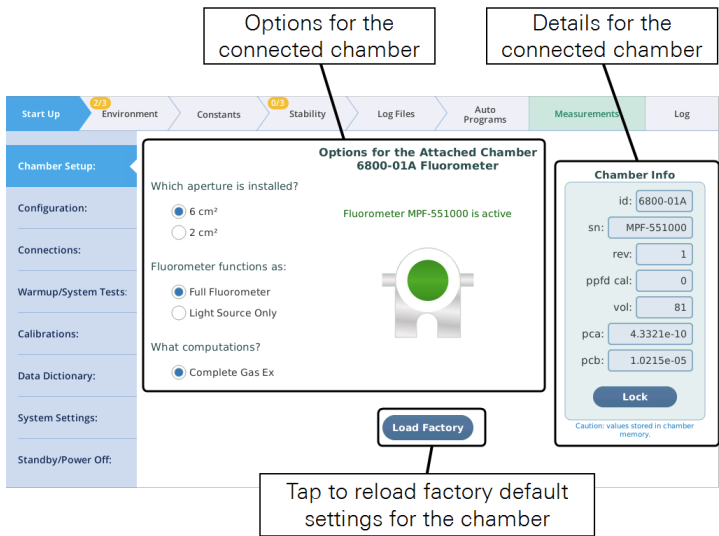
The Start Up tab

The information in this section will help you become familiar with some of the basic features of the instrument and how to change settings.

Chamber setup

Under **Start Up > Chamber Setup**, you'll see the options available for the attached chamber on the left and the chamber details on the right. The chamber-related calibration values should be automatically populated when you connect a light source.

Note: The controls for this page are disabled when a log file is open.



When the attached chamber is the 6800-01A and the fluorometer is detected (i.e., the cable is plugged in), you have a choice of **Function: Full Fluorometer** or **Light Source Only**; as well as choices for the aperture. If you choose Light Source Only, all of the fluorometer-related screens will be hidden.

Note: Toggling between the two modes (Full Fluorometer and Light Source Only) will automatically cause grids and graphs to reset to the factory default appropriate for the mode. Also, the group Flr will not be shown in variable pickers while in Light Source Only mode.

The button labeled **Load Factory** is a quick way to set the LI-6800 for the factory defaults appropriate for the attached chamber.

What computations?

There are four possible computation options, although not all are available for all chambers. These are **Complete Gas Ex**, **Area-based Fluxes**, **Mass-based Fluxes**, and **Insect Respiration**.

Note: Toggling between computation options will automatically cause grids and graphs to reset to the appropriate factory default.

Complete gas exchange encompasses photosynthesis A , transpiration E , conductance g , and all of the usual computations that derive from them. Non-fluorometer chambers also provide for just A and E , on a leaf area basis (mol or $\text{mol m}^{-2} \text{s}^{-1}$) or mass basis (mol or $\text{mol kg}^{-1} \text{s}^{-1}$). With mass-based fluxes, the only gas exchange constant (under the Constants tab) is Mass in grams (gm). Area S and stomatal ratio K are not available (*Figure 3-1* below). The two flux options are the only options available for any chamber without provisions for measuring leaf temperature (e.g., 6800-17).

Table 3-1. Details for the flux computation options.

Name	Quantity	Units	Description
Area based fluxes	A	$\mu\text{mol m}^{-2} \text{s}^{-1}$	CO ₂ flux
	E	$\text{mol m}^{-2} \text{s}^{-1}$	H ₂ O flux
	S	cm	Leaf area
Mass based fluxes	A	$\mu\text{mol kg}^{-1} \text{s}^{-1}$	CO ₂ flux
	E	$\text{mol kg}^{-1} \text{s}^{-1}$	H ₂ O flux
	Mass	gm	Plant mass
Insect respiration	A	$\mu\text{g g}^{-1} \text{min}^{-1}$	Insect respiration
	E	$\text{mol kg}^{-1} \text{min}^{-1}$	Insect "transpiration"
	Mass	gm	Insect mass

The screenshot shows the 'Constants' tab in the software interface. The 'Gas Exchange constants' section is active, displaying the following settings:

- S:** 9.00 cm² (Leaf area)
- K:** 0.5 (Stomatal ratio)
- Geometry:** 0: Broadleaf, 1: Needles, 2: Custom
- Custom:** 2.00

Figure 3-1. Gas exchange constants. The options available will differ, depending on whether the instrument is configured for area-based or mass-based measurements.

Chamber info

These fields present the information saved in a chamber's EPROM memory. They are programmed at the factory. They can be edited.

- **id** - The model number will be one of these: 6800-01, 6800-01A, 6800-12, 6800-12A, 6800-13, 6800-17, or 6800-24.
- **sn** - The serial number should match the number on the chamber label.

- **rev** - A revision number. Not used currently.
- **ppfd cal** - The light sensor calibration factor is used for the 6800-12, 6800-12A, and 6800-13 chambers. For the other chambers, this value can be missing or zero.
- **vol** - The chamber volume (cm^3) is used to tune the response time of control loops when controlling the sample CO_2 or water vapor. See *Table 3-2* below.
- **pca, pcb** - Two coefficients used to correct measured chamber over pressure ΔP (kPa) for the effects of fan speed (rpm) R and flow rate F ($\mu\text{mol s}^{-1}$). Not all chambers require this. See *Table 3-2* below for *pca* and *pcb* values.

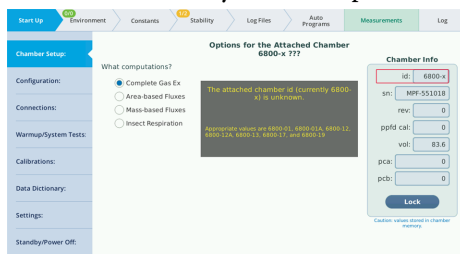
Table 3-2. Values for volume and pressure correction coefficients for each chamber.

Chamber	Volume (cm^3)	pca	pcb
6800-01 Fluorometer	83.6	0	0
6800-01A Fluorometer	87.3	4.3321E-10	1.0215E-5
6800-12 3×3	101.8	0	0
6800-12A 3×3	109	2.6520E-10	1.3064E-5
6800-13 6×6	193.7	4.3998E-11	1.4792E-5
6800-17 Small Plant	193.2	0	0
6800-19	? (possibly ~200)	0	0

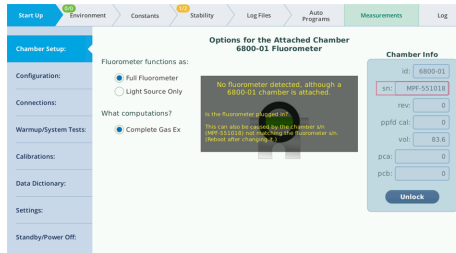
Warning messages

There are warning messages that will be triggered if there is a mismatch between that information and the information stored on the chamber. This can be caused by manually entering incorrect chamber info or if the chamber memory becomes corrupted somehow.

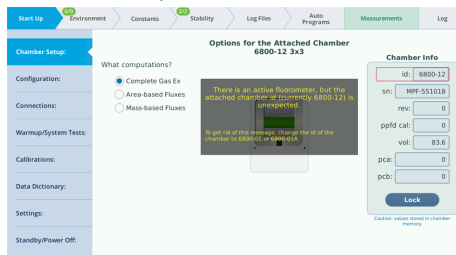
- **Unknown chamber warning:** If triggered, the chamber id box is highlighted in red. That is where you fix the problem.



- **Missing fluorometer:** The chamber sn box is highlighted in red since this is often the cause of the mismatch.



- **Unexpected fluorometer:** If this happens, the chamber id box is highlighted in red. It is likely because the chamber id is not correct.



If you encounter one of these messages, follow the on-screen directions to correct the issue.

Factory defaults

Tapping the **Load Factory** button will reset the indicated configuration groups to factory defaults appropriate for the chamber and options you have selected. Here are some examples:

If selected, the **EnvLight** configuration group implements factory default settings for each currently attached light source. If the attached chamber is compatible with the light source (as in, clear-top 3×3 cm chamber and small light source), the LI-6800 assumes that the light source is attached to the chamber.

In the **GasEx** group, leaf temperature constants are set for measuring leaf temperature using connector T1. Gas exchange constants are set for Complete Gas Exchange, and leaf area depends on the attached chamber. If the chamber supports apertures, it selects the default aperture.

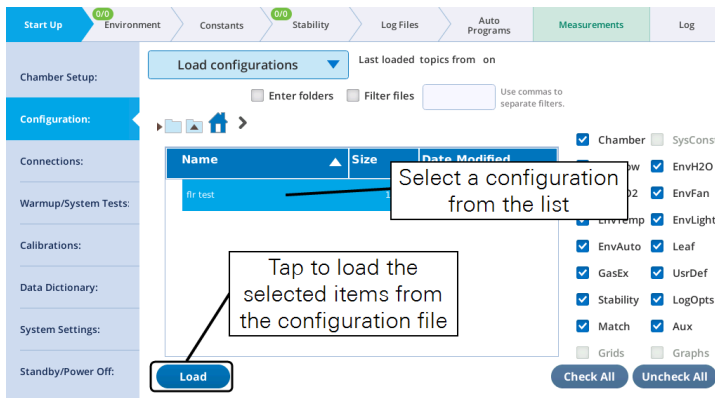
In the **EnvFlow** group, the flow rate is set according to the attached chamber.

Configuration

The **Configuration** tab is where you load, save, or transfer a configuration file to an external USB storage device. A configuration file is a collection of settings that tells the instrument what to do.

Load configurations

Here, you can load an existing configuration file. If there are no saved configurations, the list will be empty. To load a configuration, select the file and tap **Load**. If there are parameters that you *do not* want loaded, clear the check box, then tap **Load**.



Save configurations

Under **Start Up > Configuration**, you can save the configuration by selecting **Save configurations** from the menu. When saving, you can choose the settings to include in the saved configuration. The settings available to save and load are:

- **Chamber:** Chamber-related settings.
- **EnvFlow:** Flow control settings.
- **EnvH2O:** Water vapor control settings.
- **EnvCO2:** CO₂ control settings.
- **EnvFan:** Mixing fan controls.
- **EnvTemp:** Temperature control settings.
- **EnvLight:** Light settings.
- **EnvAuto:** Auto Controls settings.

- **Flr**: Fluorometer settings.
- **Leaf**: Parameters relating to light source and leaf absorption constants. See *Table 6-10* on page 6-34.
- **GasEX**: The system constants.
- **UsrDefCon**: User defined constants.
- **Stability**: Stability criteria.
- **LogOpts**: Logging options, settings under **Log Files > Logging Options**.
- **Match**: Matching configuration information.
- **Aux**: The state of Auxiliary on the 25-pin connector
- **Grids**: The configuration of variables under the **Measurements** tab.
- **Graphs**: The configuration of graphs under the **Measurements** tab.

To save the configuration, tap **New Config**, enter a name, then tap **OK**. You can also save configurations to a folder structure that you create.

USB Utilities (Start Up)

Under **Start Up > Configuration > USB Utilities**, you'll find options for copying files between the LI-6800 internal memory and an attached USB storage device.

Inserting a USB drive will, after several seconds, automatically bring up the USB file system in either the source or destination views, depending on how the To/From radio buttons are set (*Figure 3-2* on the next page). Inserting a second USB drive will produce a radio button selection for which USB drive you wish to use (*Figure 3-3* on page 3-11).

To copy one or more files or folders, select them (tap and drag for multiple, contiguous selection) in the source view, select a folder in the destination view using the **Browse** button or typing in the edit box, then tap the **Copy** button. A dialog confirming the copy or reporting a failure is displayed after the copy (*Figure 3-4* on page 3-11).

To remove a USB drive safely, tap **Eject** to verify it is safe to remove (*Figure 3-5* on page 3-12). Note you can continue to use the USB drive after tapping **Eject** if you choose - it remains functional until you physically remove the drive from the console.

If a USB drive has a space character in the volume name (allowed, but never a good idea), the name in the edit box for the destination volume will show a `nx20` instead of a space. For example, a drive named `DISK IMG` will be shown as

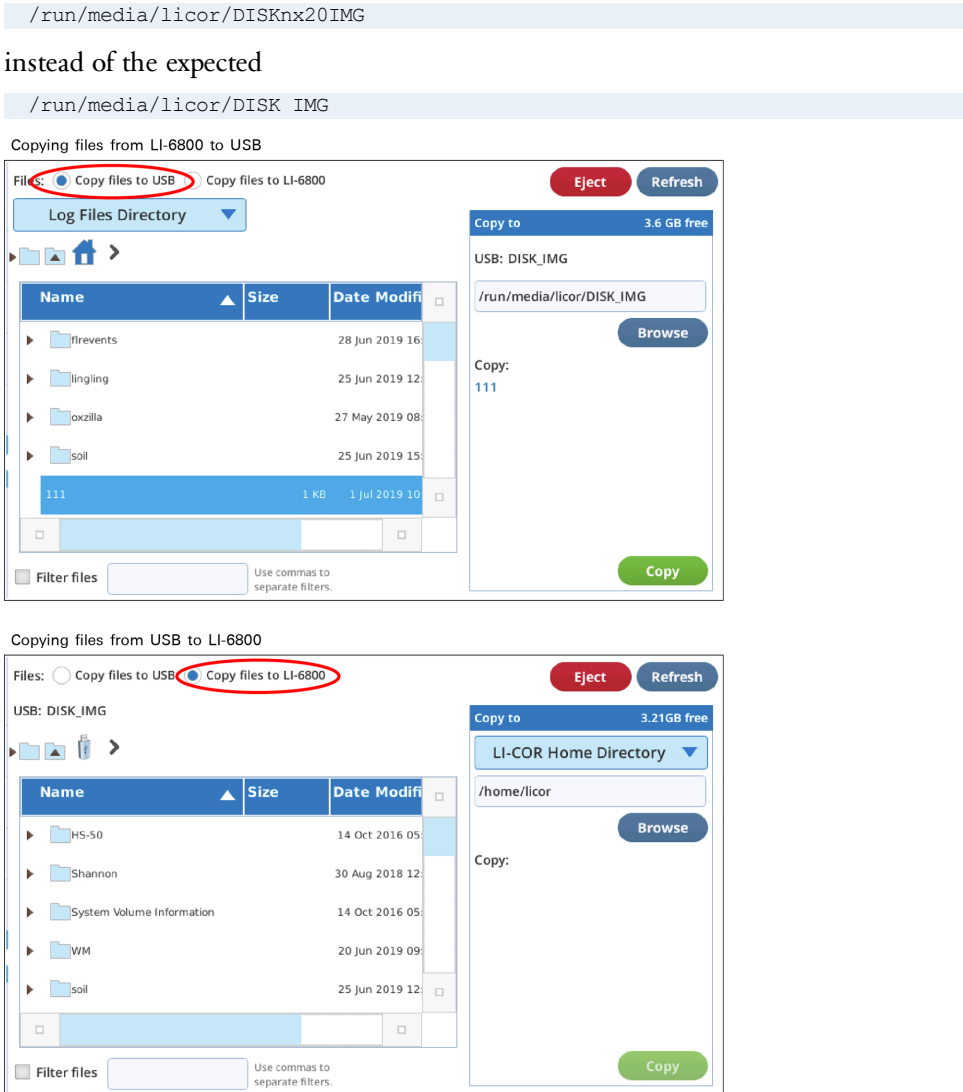


Figure 3-2. USB Utilities displays the source on the left and the destination on the right.

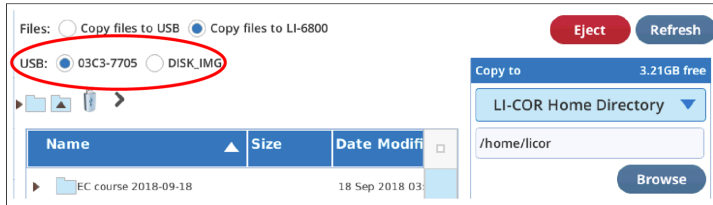


Figure 3-3. If a second USB drive is inserted, radio buttons are provided to select which to use.

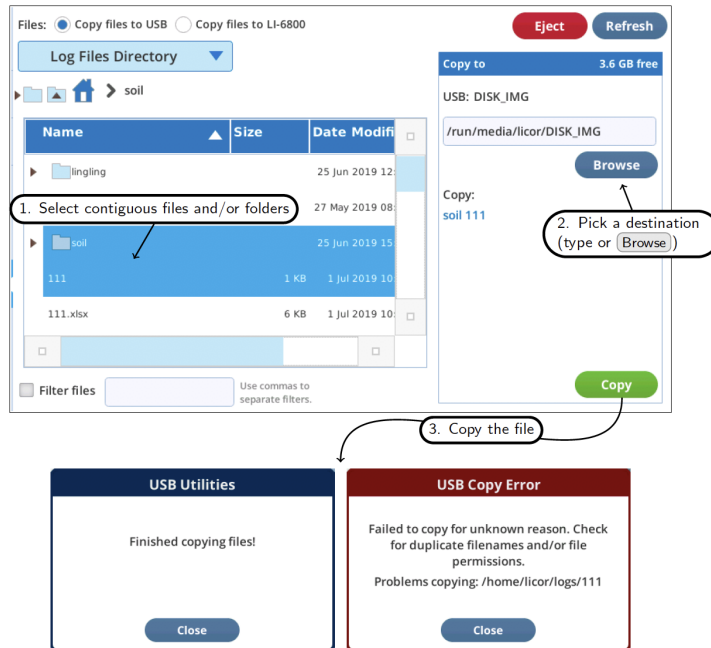


Figure 3-4. Copying an item: 1. Select it in the source view, 2. Select a destination, 3. Tap **Copy**. Existing files will not be overwritten on the destination, but will bring up the error dialog.

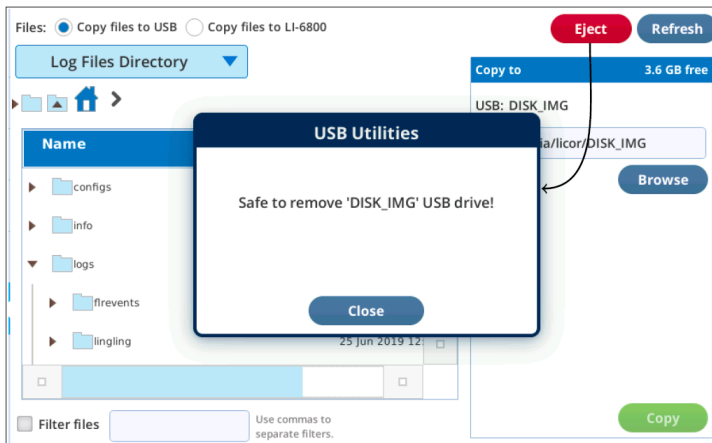
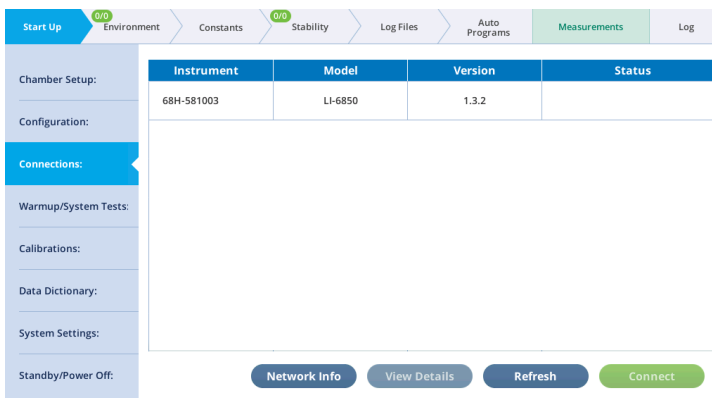


Figure 3-5. Tap **Eject** to verify it is safe to remove a USB drive.

Connections

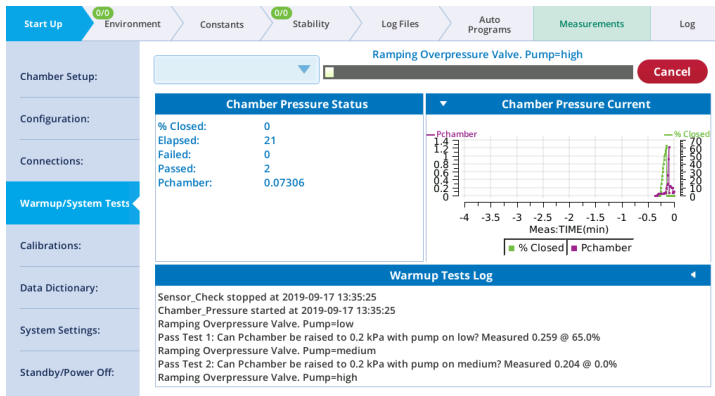
Here you can connect to any head (digitally) that is attached to the console; or on the same network as the console, assuming that the console is connected to your network with the RJ-45 Ethernet cable. The latter is a good way to wreak havoc on the photosynthesis lab next door, but remember, if you can see them, they can see you.



Warmup and system tests

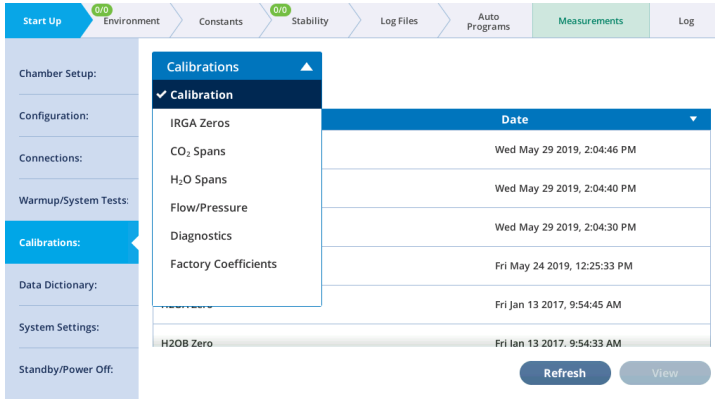
The warmup tests are designed for daily use, to catch errors and faults that might compromise your measurements. If everything passes, you can be reasonably sure (but not guaranteed) that the system is working as expected. Common failures are due to worn out desiccant or an empty CO₂ cylinder. Other failures may be simply because a setting or measurement is just outside the expected range, in which case you have to decide if it needs attention or can be ignored.

The tests are a convenient way to diagnose issues with the instrument. There are groups of tests: Warmup Tests, Controller Tests, Fluorometer Tests, and Tech Tests. You can run tests as a group or individually. For details on the tests, see *Warmup and system tests* on page 7-2.



Calibrations

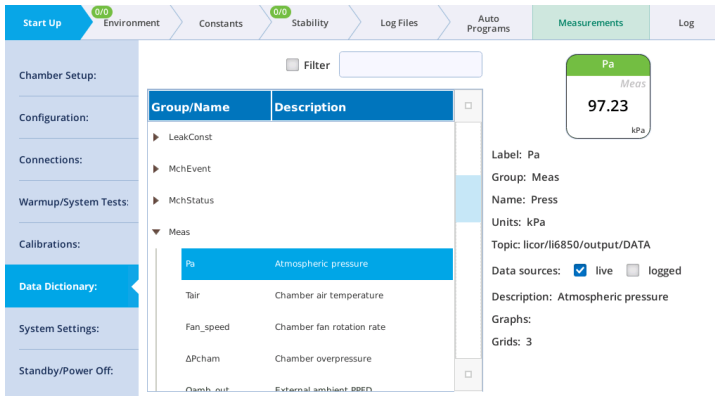
The **Calibrations** tab gives you six options: The calibration History, IRGAs, CO₂ Spans, H₂O Spans, Flow/Pressure, Diagnostics, and Factory Coefficients. This is where you view current calibration values, review the calibration history, and perform user calibrations. See *User calibration* on page 8-2 for details.



Under the **Calibrations > Diagnostics** tab, you'll see values related to the CO₂ and H₂O analyzers, including zeros, spans, absorbances, and more.

Data dictionary

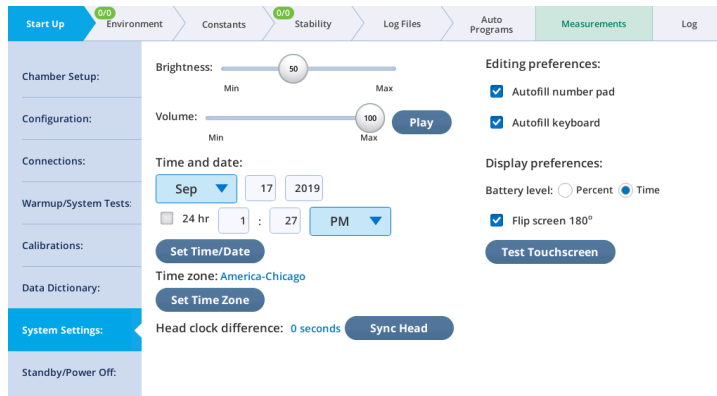
The **Data Dictionary** shows terms that are used by the LI-6800. Terms are grouped, as described in *Summary of symbols* on page 6-31. For each variable, it will show the **Label, Group, Name, Units, Description, Data sources**, and the value for that variable. By toggling live or logged, you can see the current value or the last value logged.



System settings

The **System Settings** tab is where you configure basic instrument settings, such as display brightness and volume level. The date and time settings can be set by a user, but the setting will automatically reset to the network time when the instrument is connected to a local area network or the internet. The changes to the time zone, however, will be stored. You can check for any discrepancies between the clock in the console and head, and correct it by tapping **Sync Head**.

You can set the **Autofill** behavior for the number pad and keyboard. So, if you're annoyed by the numbers that automatically populate the number pad entry, you can change the setting here. Or, if you would prefer that the number pad always display the current setting when opened, change it here too.



You can change the battery level displayed, flip the screen 180° so your coworker on the other side of the table can see it, or launch the touch screen test.

If you suspect a "dead" region on the touchscreen, you can use the **Test Touchscreen** button to test the screen. Drawing a big X from corner to corner will quickly find any inactive columns or rows. A dead band will be a vertical or horizontal band all the way across the display in which you cannot make a mark.

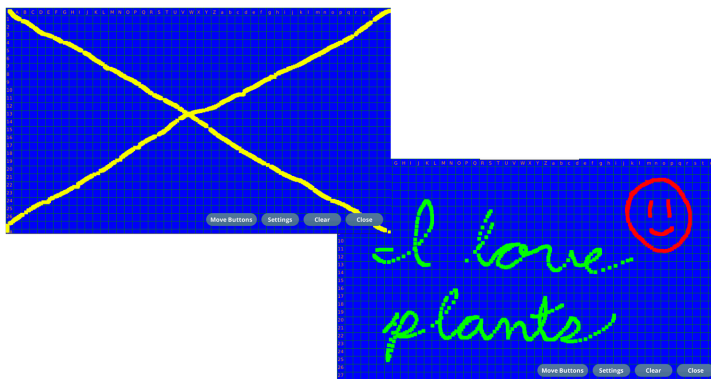
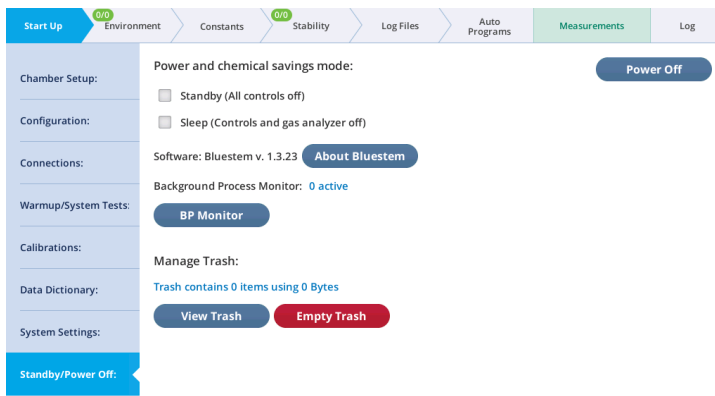


Figure 3-6. Test the touchscreen by drawing on it with your finger. A large X from corner to corner is a good test. If you have more time, you can make a more fun drawing.

Standby and power off

When you're in the middle of a field day, or if you just got everything going and now it is time to take a break, you may want to keep all the current settings, but not consume the chemicals and battery power while you take a break. There are two settings for this: **Standby (All controls off)** and **Sleep (Controls and gas analyzer off)**.



Standby (All controls off), formerly called Conserve Chemicals, will shut down all environmental controls and light sources. Disabling this setting will bring them back to their previous values. When disabled, turning on Flow, H₂O, CO₂, Fan, or Temperature, will end **Chemical Saver** mode.

Sleep (Controls and gas analyzer off), formerly **Conserve Power and Chemicals**, will shut down all power-consuming components in the head, including IRGAs, temperature controls, and fans. Data values will stop updating. Disabling this setting will not disable the chemical saver—so you can warm up the IRGAs without consuming chemicals.

The BP Monitor is where you can observe the status of any Background Programs that are running.

For details on the **Trash**, see *Trash* on page 6-4.

The Environment tab

The leaf chamber environment is controlled from the **Environment** tab. The available controls are **Flow**, **H₂O**, **CO₂**, **Fan**, **Temperature**, **Light**, and **Auto Controls**. In this section we describe the settings under these tabs. Each of the tabs on the left will indicate whether the setting is **Off** or **On**, and if on, it will indicate the setpoint and whether the setpoint is achieved.

Flow control: The answer is blowing in the wind

The **Environment > Flow** screen shows—and lets you interact with—the flow control system.

With software version 1.1, we introduce a method for dealing with leaks. This is described mathematically in *Mass balance derivation using a two-compartment model* on page C-38. Here, we describe information that is displayed on the **Flow** screen.

Pump speed and flow partition

Air flow originates from the console pump, which has settings of **Auto**, **High**, **Medium**, **Low**, **Minimum**, and **Off**. The **Auto** setting automatically selects the proper pump speed based on the requested flow rate. The flow splitter divides the flow coming into the sensor head into two paths: one path goes to the leaf chamber and sample gas analyzer, and the other path goes to the reference analyzer.

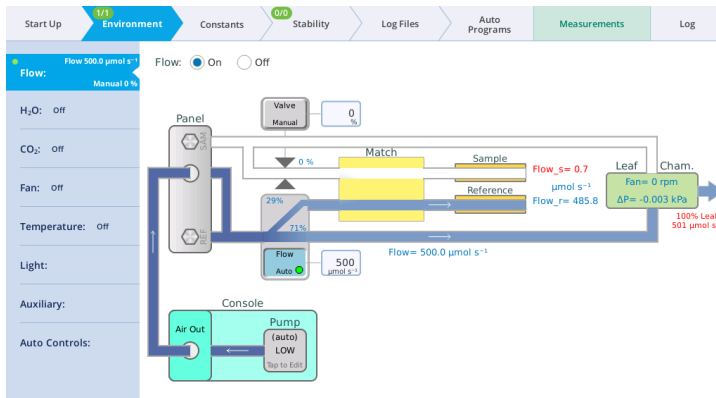
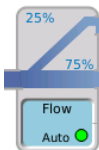


Figure 3-7. The **Flow** tab is where you turn flow **On** or **Off**, set **Pump** speed, proportion flow between reference and sample, and set the chamber pressure.

Flow: On Off The **Flow** option turns the pump on or off.



Pump controls the pump speed setting, and thus, the total flow through the system. Tap the button to change the setting. Auto is a safe choice.



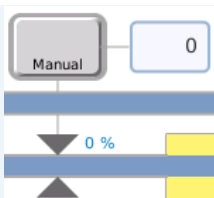
Tap the top of the **Flow** button to switch the chamber flow control between % and $\mu\text{mol s}^{-1}$. You'll use $\mu\text{mol s}^{-1}$ during measurements. Tap the upper part of the graphic to open the one-tap scale.



Flow rate setting (or flow split if % is selected), controls the amount of flow through the chamber, and thus the sample gas analyzer. The remainder is delivered to the reference gas analyzer.



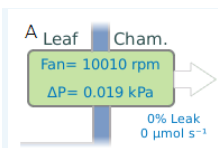
Flow rate, as measured by the mass flow meter. It should be very close to the flow rate setting.



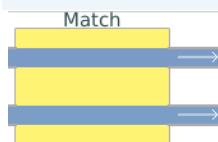
Press. Valve setting is used to create a small overpressure in the chamber to counteract the effect of diffusion leaks. Tap the triangles to open the one-tap scale. Tap the bottom to switch between automatic and manual setting. The recommended setting is 0.1 kPa.



Flow_r and **Flow_s** ($\mu\text{mol s}^{-1}$) are flow through the gas analyzers. These flows are not measured with great accuracy. There will be differences between these measurements and flow measured by the mass flow meter. In this image, Flow_s is near 0 because the chamber is open.



Fan is the measured mixing fan speed. **ΔP** is the difference between chamber pressure and ambient.



Match valve position. The match valve position will change when the instrument is matching the IRGAs.

Leak rate

The flow tab shows a real-time estimate of the leak rate from the leaf chamber. This value is logged in a variable group called **Leak**. When a leak gets large enough that the correction becomes more than 2% at very low fan speeds, the fan control diagram will clearly show you the relationship between fan speed and the leak correction factor, allowing you to select a fan speed that keeps the correction factor at a negligible level. If, for some reason, you have large leaks, and you need a low fan speed, then you will clearly see the degree to which your results will depend on the leak correction.

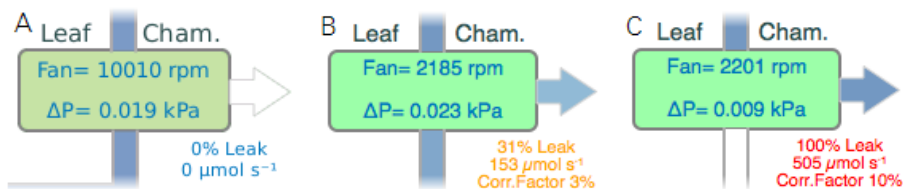


Figure 3-8. **A:** There is a leak, but the fan speed is high enough, so no correction is applied. **B:** There is a leak and a low fan speed. The instrument warns you that the correction factor is 3%. **C:** With the chamber open, the leak is 100%.

The mixing fan controller provides guidance for selecting a suitable fan speed. See *Fan speed control: All mixed up* on page 3-31 for details.

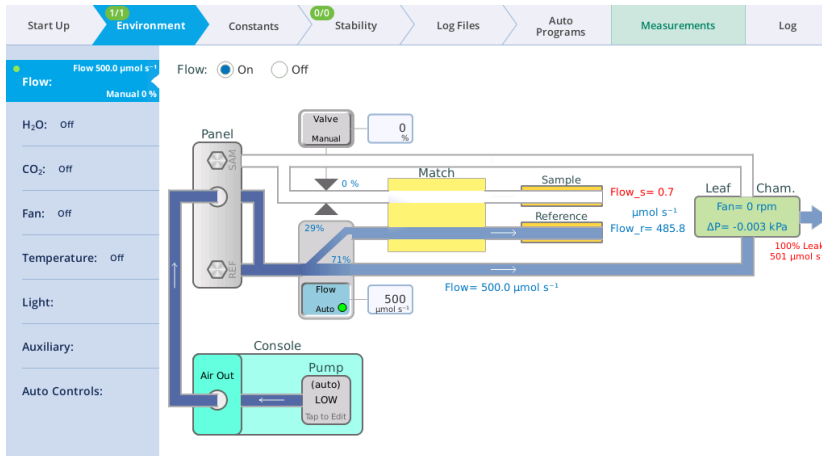
Manual flow control

- 1 Start with the chamber open.
- 2 Set Flow to On.

Tap the button below **Pump** and set the pump speed to **Low**.

- 3 Set the **Flow** to the chamber.

You want to set flow as $\mu\text{mol s}^{-1}$ so tap the **Flow** button if needed, then enter a **Flow rate** of 500.



- 4 Be sure the chamber overpressure set to 0 kPa.

Close the chamber and notice what happens: Paths with airflow are now blue—a shade that is proportional to flow rate. In the example above, the flow meter tells us how much flow is going to the leaf chamber. The remainder is going to the reference analyzer. These values will be slightly different for your instrument.

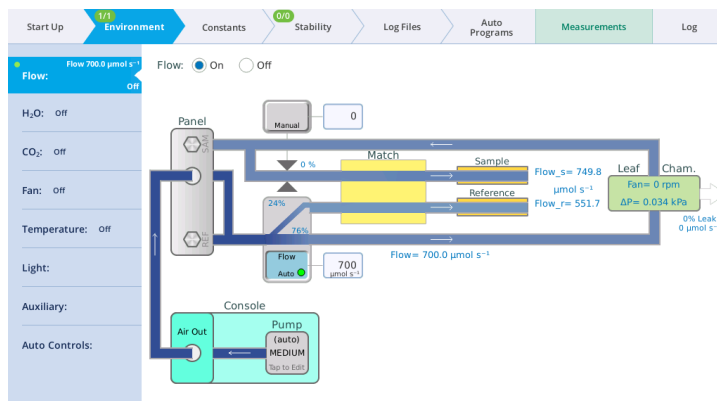
- 5 Now, tap the flow split graphic and select about 800 $\mu\text{mol s}^{-1}$. See how this alters the **Flow_r** and **Flow_s** values.

Automatic flow control

In normal operation, you don't need to set the flow splitter manually. You can let the instrument do it based on a target value you enter.

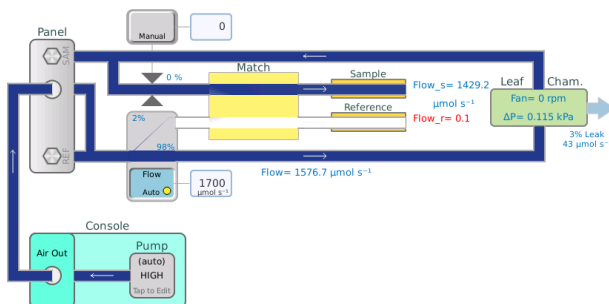
- 1 Set the **Pump** to **Auto**.
- 2 Close the chamber and enter a target flow of 700 $\mu\text{mol s}^{-1}$.

The flow controller will very quickly bring the flow to the target value. Also, you have about $760 \mu\text{mol s}^{-1}$ left over for the reference cell (this number will be different for your instrument). The dot on the **Flow** tab turns green and the 0/1 on the Environment tab becomes 1/1, indicating that the setpoint has been achieved.



3 Set the target very high, such as $1700 \mu\text{mol s}^{-1}$.

Notice you have very little flow left over for the reference cell, and its flow dropped to a value that is too low (indicated by **Flow_r Low** warning message and the white reference flow path).



Balanced Flow

Having sample and reference flow rates balanced can be advantageous for eliminating potential errors, and it is now easier to achieve. For every pump speed, there is one flow rate that will match sample and reference flows (to within $5 \mu\text{mol s}^{-1}$). To quickly get to that flow rate, simply tap the **Pump** button on the **Flow** control

page to bring up the **Pump Speed** dialog (*Figure 3-9* below). Tap one of the four flow rate possibilities presented on the right side.

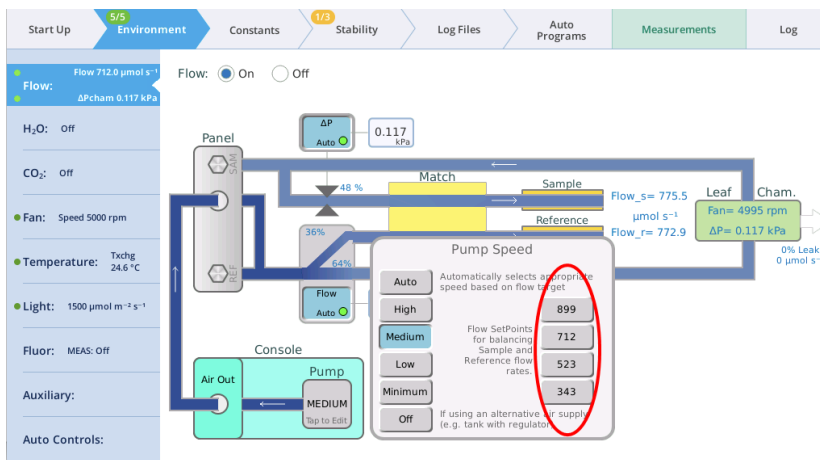


Figure 3-9. Balanced flow.

There are two Background Programs that deal with balanced flow:

- When you tap one of the balanced flow setpoint buttons, a BP (`/home/utility/apps/system/balance_flow.py`) is launched (no opening dialog) a few seconds later to check on the result, and fine-tune the flow if needed. That fine-tuning will automatically update the balanced flow setpoint for that pump speed. (Note: the chamber must be closed, of course, for the flows to match. If the chamber is open, then the BP won't attempt to adjust the flow rate.)
- The BP `/home/utility/apps/utilities/Balance_Flow.py` has an opening dialog that lets you pick what pump speed to use (*Figure 3-10* on the facing page), then balances the flow for that speed. (Cycling through all four pump speeds is also an option.) Running this program also updates the balanced flow setpoints shown in *Figure 3-9* above.

There is also a hidden shortcut: If you tap the flow rate set point edit box (the one you normally use to enter a new flow rate), and enter an E instead of a value, the `balance_flow.py` BP will be launched. If the pump is in Auto mode, it will change to the non-automatic version of its current speed, and the flows will be balanced.

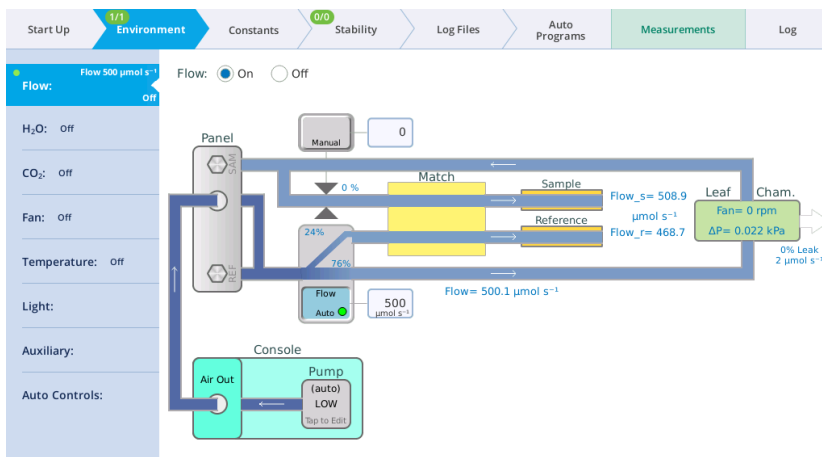


Figure 3-10. The BalanceFlow utility program can be run manually to determine (and automatically update) balanced flow set points.

Chamber overpressure

Now you can move on to the rest of the flow system.

- 1 Set up the instrument for automatic flow control at $500 \mu\text{mol s}^{-1}$, and close the chamber. Things should look like this:



2 Tap the button beneath the words **Press. Valve** so it reads ΔP kPa.

For **Chamber overpressure**, enter 0.2 kPa. The instrument limits pressure to 0.2 kPa to prevent damage to itself.

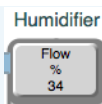
If you are measuring irregular leaves and the chamber does not seal very well, you can increase this setting to force leaks out and minimize inward diffusion of ambient air. Change the setting back to 0 kPa.

Water vapor control: Who'll stop the rain

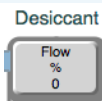
Water vapor is set under the **Environment > H₂O** tab. Here you can alter the humidity in the air stream by adding and removing water vapor. Remember, leaf chamber humidity is a combination of humidity coming from the console and from the leaf.

H₂O: On Off

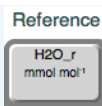
The H₂O option turns humidity control On or Off.



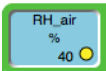
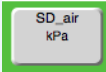
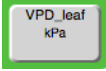
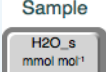
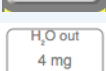
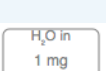
Tap this button to control flow through the humidifier manually. You can enter a setpoint when this option is selected.



Tap this button to control flow through the desiccant manually. You can enter a setpoint when this option is selected.



Tap this button to control water vapor in the reference analyzer.

	Tap this button to control relative humidity in the leaf chamber.
	Tap this button to control on the saturation deficit of the air.
	Tap this button to control vapor pressure deficit in the leaf chamber.
	Tap this button to control water vapor in the sample analyzer.
	Fresh Desiccant or wet Stuttgarter Masse? Tap these buttons when you replenish the humidifier and desiccant.
	

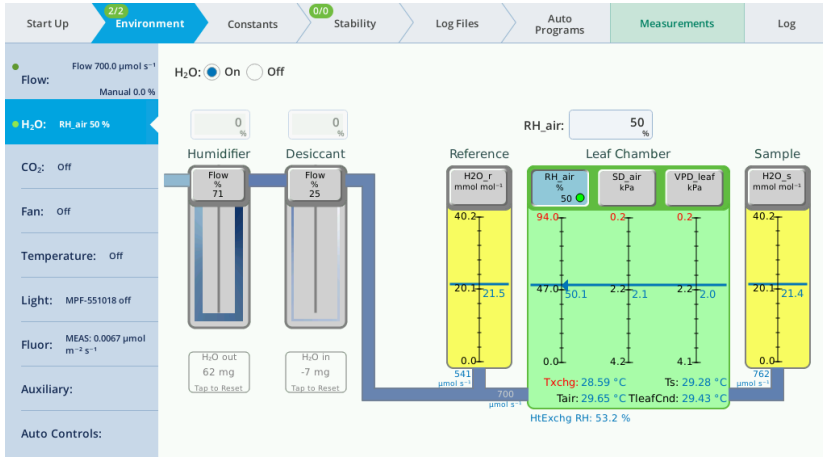
In addition to the buttons, you can tap the tube graphics to open the one-tap scale.

Controlling the H₂O concentration

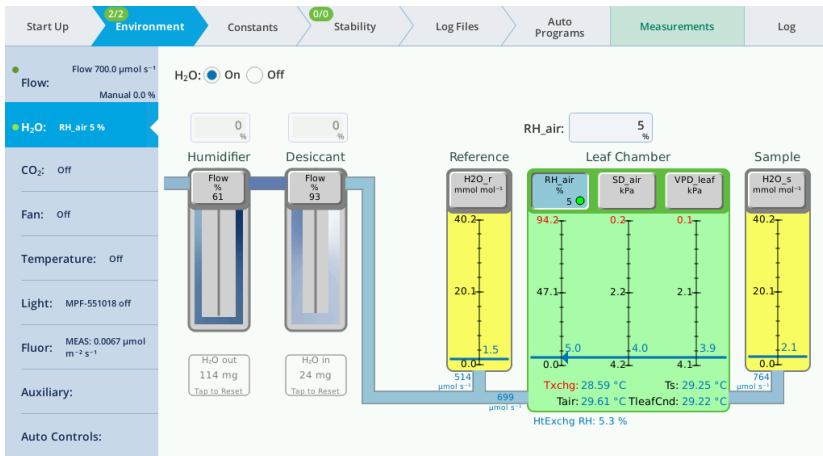
This exercise shows the range of water vapor concentrations that the instrument can establish. Start with a closed chamber.

- 1** Configure flow.
 - Under **Environment > Flow**.
 - Turn **Flow**: On
 - Set the pump speed to **Auto**
 - Set the flow rate to $700 \mu\text{mol s}^{-1}$.
- 2** Under **Environment > H₂O**, set **H₂O** to **On**.
- 3** Tap the **RH_air** button, then enter 50% in the **RH_air** box.

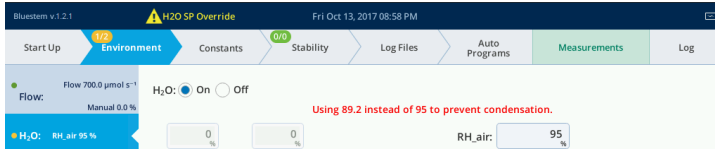
Watch the display as the instrument adjusts the settings to achieve the relative humidity setpoint.



4 Set the RH_{air} setpoint to 5% and observe the display as the instrument achieves the setpoint.



5 Set the RH_{air} setting to 95% and observe the display as the chamber relative humidity approaches the setpoint. (95% is too high, but we are forcing the instrument to override the setting.)



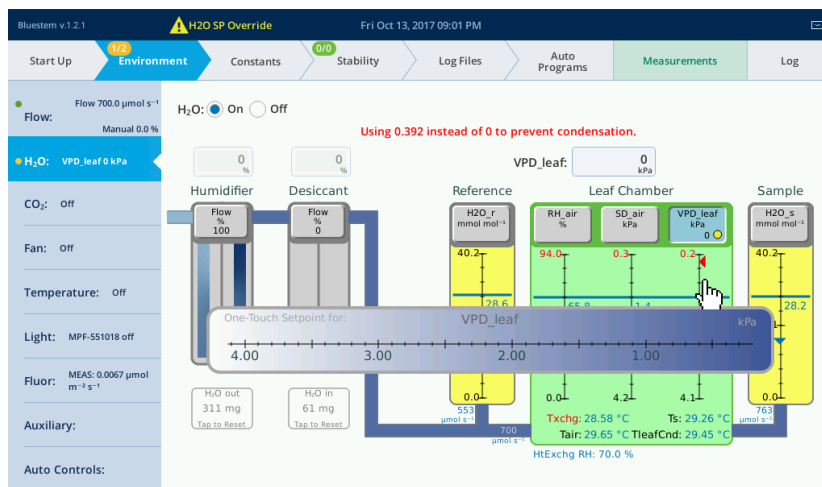
Notice that the instrument will not achieve the setpoint and it displays **H2O SP Override**. The instrument has limited the RH to prevent condensation in the chamber, gas analyzers, or heat exchanger.

- Now reset the **RH_{air}** setting to 50%.

Controlling vapor pressure difference

In this exercise we'll control leaf vapor pressure difference (VPD) in the leaf chamber. It is expressed as kPa.

- Tap **VPD_{leaf}**, then tap the scale and tap 0 to select 0 kPa.



The instrument will achieve the highest humidity allowed by adjusting the humidifier and drier. Observe the **Sample** and **Reference** RH values. It will override the setpoint.

- Change the setpoint to 2 kPa.

Observe the changes in the flow path and the subsequent changes to the **Sample** and **Reference** RH values.

- Change the setpoint to 3 kPa.

Again, observe the changes in the flow path and changes to the **Sample** and **Reference** RH values.

Selecting a temperature for the condensation check

Software version 1.1 introduces the ability to measure both *Tleaf* and *Tleaf2*. With two thermocouples, you can use one or the other, inside or out of the chamber, and averaging, which adds to the complexity. The LI-6800 uses *TleafCnd* for the temperature, but what if we are averaging, or using energy balance, and want to communicate that one of the thermocouples is the limiting temperature, even though the average isn't? And there's one more thing: now that we have the ability to do flux-only computations, *TleafCnd* may not even exist. In the words of an old commercial, What's a mother to do? Make rules, of course, and here they are:

- 1 If *Tleaf* or *Tleaf2* (if in the chamber) become the humidity-limiting values, the label and value will reflect that (Figure 3-11 below, A).
- 2 If *TleafCnd* is being computed, and any in-chamber thermocouple is NOT the limiting temperature, then *TleafCnd* will be used (Figure 3-11 below, B).
- 3 If *TleafCnd* is not computed, the label and value will be (in order of priority) *Tleaf*, *Tleaf2*, or *Tair* (Figure 3-11 below, C).

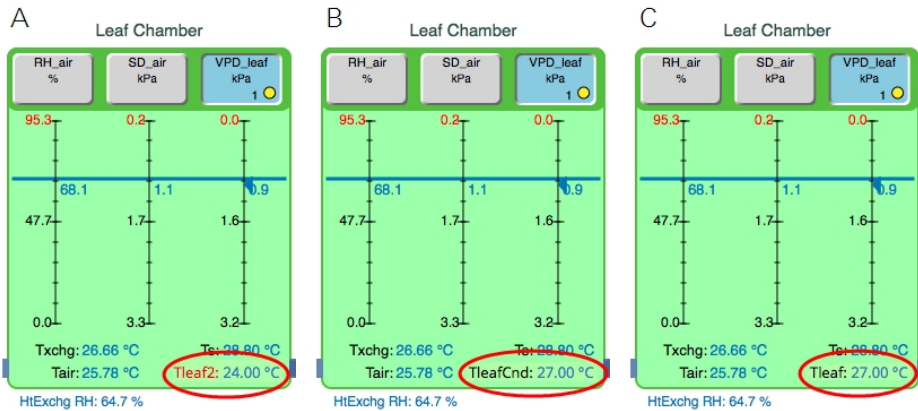
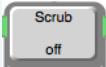
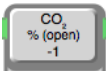
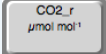
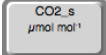


Figure 3-11. The leaf temp label now reflects the same temperature used in computations (including when energy balance is used), unless *Tleaf* or *Tleaf2* becomes the limiting temperature. **A:** *Tleaf2* is in the chamber, and it is the limiting temperature. It doesn't matter if there is a *TleafCnd* value or not. **B:** Wherever leaf temperature is coming from (energy balance, average, it doesn't matter), it is not the limiting temperature. **C:** There is no *TleafCnd* (flux only), but if you want VPD leaf, it will use *Tleaf* (or *Tleaf2*, or *Tair*) to compute one.

Carbon dioxide control: Full of hot air

CO₂ control is set under **Environment** > **CO₂**. The instrument removes CO₂ from the air stream using the soda lime scrubber and adds a controlled amount of CO₂ with the mixer. Be sure there is a charged CO₂ cartridge installed for these exercises.

CO₂ injector: On Off The **CO₂ injector** option turns the CO₂ control on or off.

<p>Soda Lime</p> 	<p>Tap this button to control soda lime (CO₂ scrubber). The options are Auto, On, and Off. You'll use Auto during normal operation.</p>
<p>Injector</p> 	<p>Tap this button to manually control the CO₂ injector. Normally this will be controlled by the instrument when you control the concentration in the reference of sample gas analyzer.</p>
<p>Reference</p> 	<p>Tap this button to control the concentration in the reference gas analyzer, then enter the target concentration.</p>
<p>Sample</p> 	<p>Tap this button to control the concentration in the sample gas analyzer, then enter the target concentration.</p>
<p>CO₂ in 0 mg Tap to Reset</p>	<p>Time 0.1 hr Tap to Reset</p> <p>Fresh cartridge or soda lime? Tap these buttons when you replenish the soda lime or CO₂ cartridge.</p>

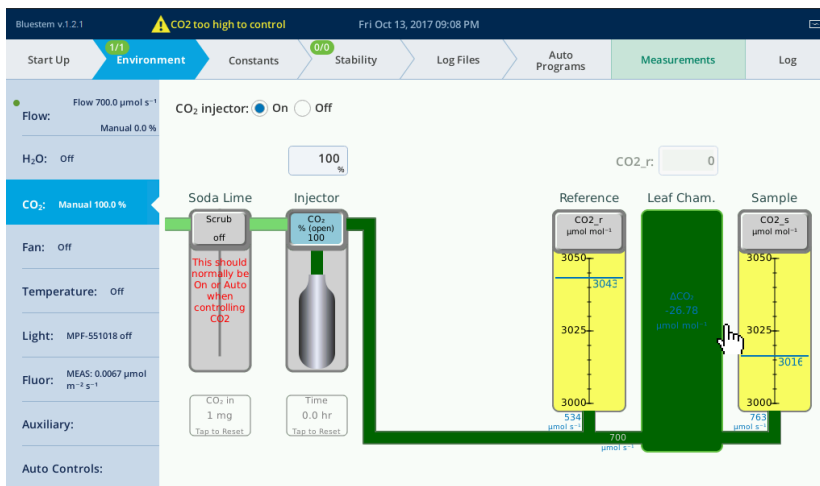
In addition to the buttons, you can tap the graphics for any activated control to open the one-tap scale.

Manually control the CO₂ concentration

You can control the CO₂ injector and scrubber (soda lime) independently of each other. We'll begin by just operating the injector. You can control the soda lime and injector both manually, but normally you'll let the instrument control them automatically based on target value for the sample or reference gas analyzer or the leaf chamber.

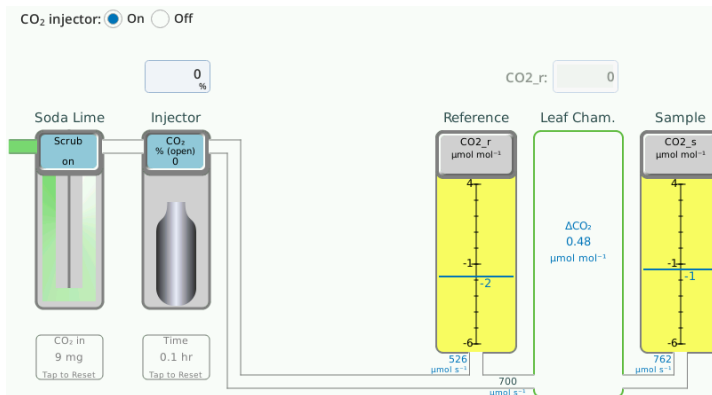
- 1 Set the flow to 700 μmol s⁻¹.
- 2 Turn the **CO₂ Injector On**.
- 3 Set the **Soda Lime to Auto**. Tap the button and select Auto, if needed.

- 4 Tap the **Injector** button and then tap the cartridge graphic. Tap 100% on the graphic to open the injector all the way.



The instrument will open the CO₂ injector all the way to achieve the highest CO₂ possible. Observe the concentrations of CO_{2_r} and CO_{2_s}.

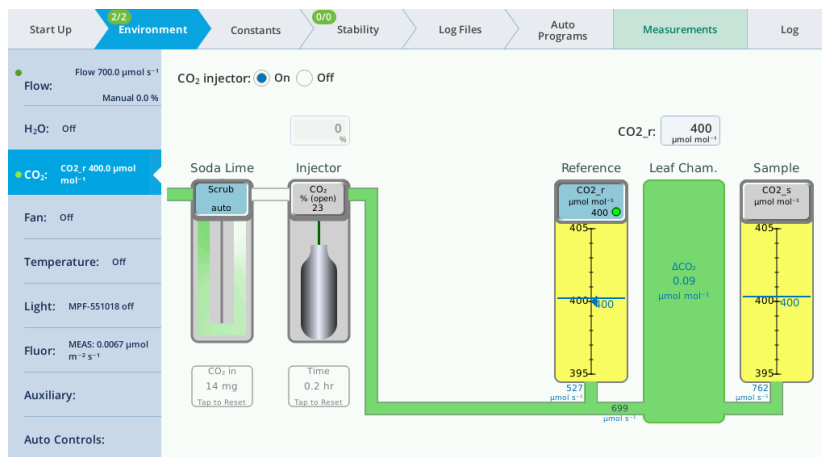
- 5 Tap the **Soda Lime** button and select **On**. Set the **Injector** to 0%. Observe the CO_{2_r} and CO_{2_s} values. They should approach zero quickly.



Automatically control the concentration

- 1 Tap the **Soda Lime Scrub** button and select **Auto**.
- 2 Tap the **Reference (CO_{2_r})** button, then enter a target for CO_{2_r} of 400 $\mu\text{mol mol}^{-1}$.

Observe the concentrations of CO_2_r and CO_2_s . It may take a minute for the values to stabilize at the setpoint. Since there is no leaf in the chamber, the ΔCO_2 should be very close to zero. If the Δ is not close to zero, you may need to match the IRGAs.



3 Enter a setpoint of $5 \mu\text{mol mol}^{-1}$ in the reference cell.

Observe how quickly it adjusts to the setpoint.

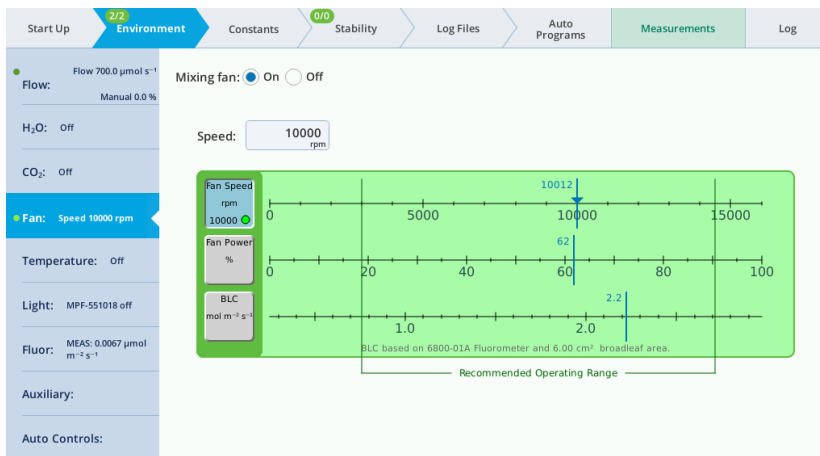
4 Change the setpoint to 150, 500, and then $1000 \mu\text{mol mol}^{-1}$.

Wait for the system to adjust after you've changed the setting. Observe the measurements in the reference and sample IRGAs, and the value for ΔCO_2 .

5 Now turn the CO_2 injector off.

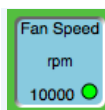
Fan speed control: All mixed up

The mixing fan speed is controlled under **Environment > Fan**. It can be controlled to target a fan speed (rpm), power (%), or a boundary layer conductance ($\text{mol m}^{-2} \text{s}^{-1}$).

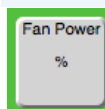


Mixing fan: On Off

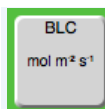
The **Mixing Fan** option turns the mixing fan On or Off.



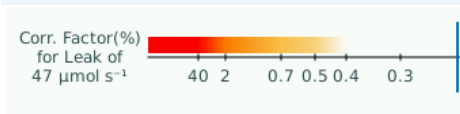
The **Fan Speed** button is to control the mixing fan on a target RPM.



The **Fan Power** button is to control the mixing fan on a power %.



The **BLC button** is to control the mixing fan speed on a target Boundary Layer Conductance. This is not shown in any configuration in which the BLC is not computed.



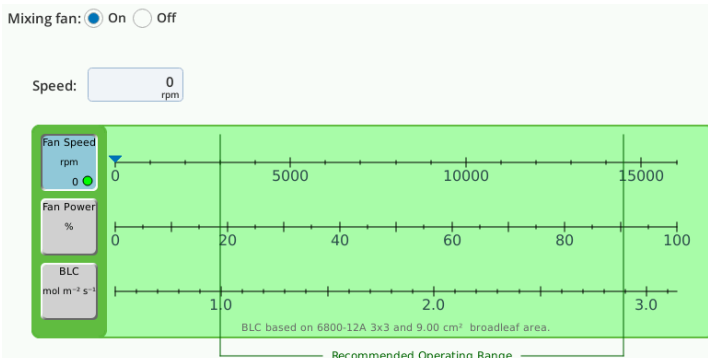
For the early-model chambers (6800-01, 6800-12) a leak correction factor may be applied at low flow rates and low fan speeds. This is only shown when a leak correction factor is needed.

Control fan speed

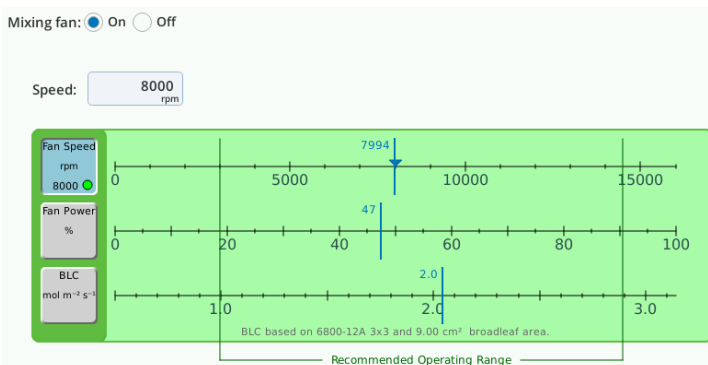
When controlling the fan speed manually, tap the slider or simply enter a value.

- 1 Set the **Mixing Fan** to **On**, then select **Fan Speed**.

Tap the scale at 0 rpm or enter 0 rpm in the text box labeled **Speed**. Observe the fan speed as it drops to 0.



- 2 Tap the scale at 8000 rpm or enter 8000 rpm in the box. The fan will spin up to about 8000 rpm.

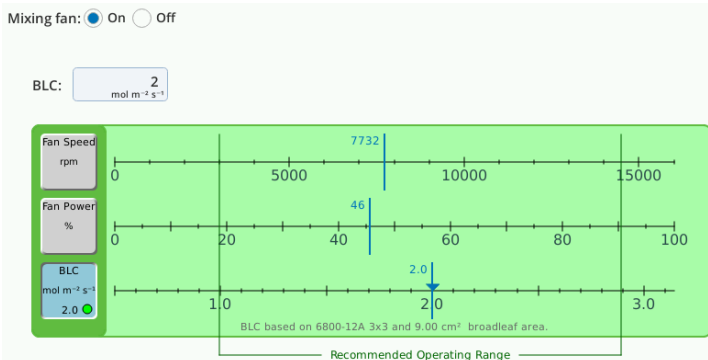


- 3 Try a few other settings. Let the fan speed stabilize before selecting the next setting.

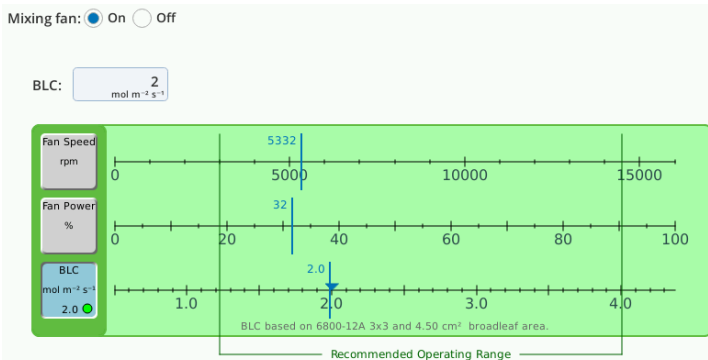
Maintain constant boundary layer conductance

The fan can be set to maintain a target boundary layer conductance. This setting is useful when you want to maintain BLC the same on successive samples of varying area.

- 1 Set the leaf type and area. If you're using an aperture, select it under **Start Up > Chamber Setup**. Then, under the **Constants > Gas Exchange** tab, select **Geometry: 0: Broadleaf**. For **S**: enter the full chamber leaf area.
- 2 Turn the fan **On**, enter a **BLC** of $2 \mu\text{mol m}^{-2} \text{s}^{-1}$. Note the fan speed.

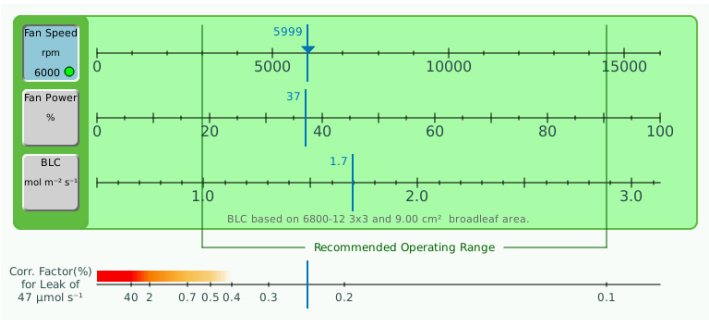


- 3** Go back to the **Constants > Gas Exchange** tab and set the leaf area to $\frac{1}{2}$ the previous setting. The instrument will adjust the fan speed to maintain the specified conductance. Observe the values as the instrument achieves the setpoint.



- 4** Turn the mixing fan off.

Note: A Corr. Factor (%) for Leak of $X \mu\text{mol s}^{-1}$ will be visible when using the early 6800-01 and 6800-12 chambers. It is larger when the fan speed and flow rate are both low. Generally speaking, you'll want your fan speed and flow rate to be high enough that there is no correction factor. The desired range is indicated in this screen. We recommend a minimum fan speed of 10,000 rpm during normal operation.



Temperature control: The heat is on

The LI-6800 can achieve temperatures of 10 °C above and below ambient temperatures under many conditions. Solar loading, light control, and temperature extremes may limit the range of temperature control.

Temperature: On Off

The **Temperature** option turns the heat exchanger On or Off.

Console

Temperature measured in the console valve manifold.

Console_T
(manifold)

IRGA

Temperature measured in the gas analyzers.

Tr
(Reference) Ts
(Sample)

Txchg

°C

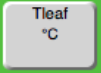
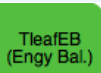
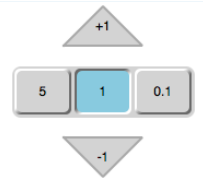

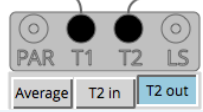
25.00

Select this to control the heat exchanger temperature, then enter the setpoint.

Tair

°C

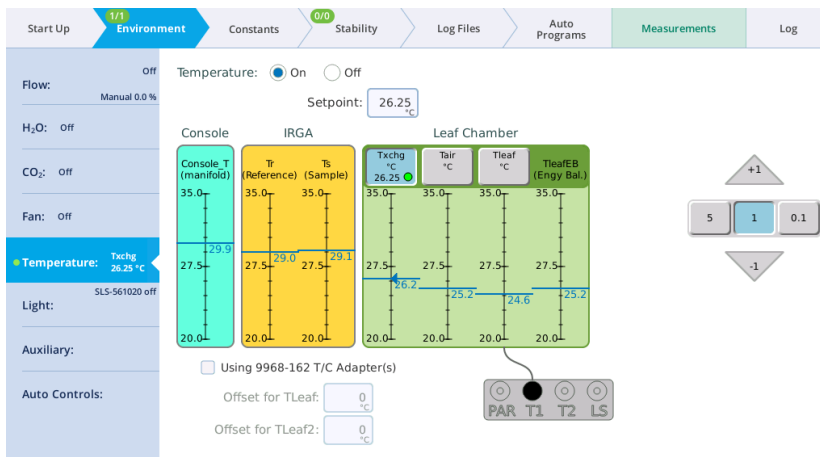
Select this to control air temperature, then enter the setpoint.

	<p>Select this to control leaf temperature, then enter the setpoint.</p>
	<p>Leaf temperature computed by energy balance.</p>
	<p>Quick adjust: Choose an increment (5, 1, or 0.1), then tap an arrow to adjust up or down by the selected increment.</p>
	<p>Representation of the head connector panel. The temperature source is T1 in this case.</p>
	<p>The Latching Buttons are visible when a second thermocouple is detected. These are described in greater detail in <i>Leaf temperature source</i> on the facing page.</p>

Adjusting the temperature

In this exercise we'll go through the settings to see the instrument respond.

- 1 Turn **Temperature: On** and then tap **Txchg**.
Observe the **Txchg** and **Tchamber** values in the display.
- 2 Enter a **Setpoint** of 25 °C or something that is a few degrees different from the current measured values.
The heat exchanger fan will turn on and the instrument will adjust to the new setpoint.
- 3 Next, tap **Tair** and enter a setpoint of 27 °C.



The instrument will adjust the heat exchanger until the air temperature is at the setpoint.

4 Now, set the Temperature Control to Leaf.

The instrument will adjust the heat exchanger until the leaf temperature measurement is at the setpoint.

5 Turn the temperature control off.

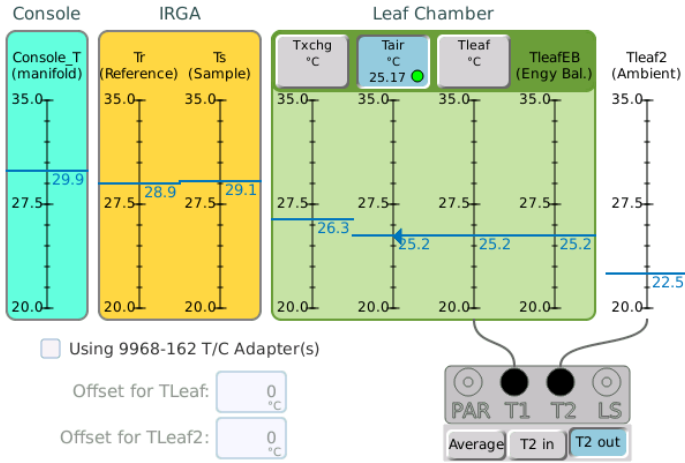
Leaf temperature source

The leaf temperature can be measured directly from one thermocouple, two thermocouples, or be computed with an energy balance equation. Temperature calculations are described in *Additional calculations* on page C-20. Any time there are two thermocouples plugged in, there is a question of what to do with the second value: Average the two? Use it for an external temperature?

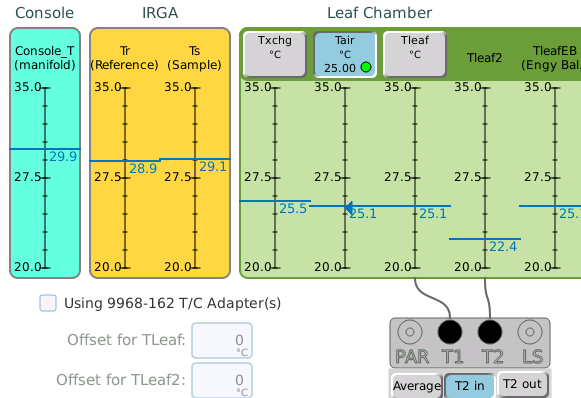
The instrument selects temperature based on the following:

- If one thermocouple is plugged into T1 or T2, the LI-6800 uses the thermocouple.
- If you want to use energy balance, select it in the **System Constants** page.
- If no thermocouples are plugged into T1 or T2, the instrument uses energy balance.
- If two thermocouples are plugged in, you can specify the role of the thermocouples:

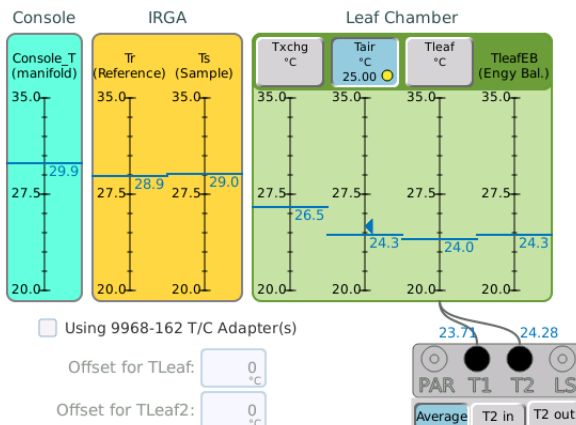
If **T2 out** is selected, T_{leaf2} is treated as external, so it won't be considered for high humidity or condensation warnings.



If **T2 in** is selected, T_{leaf2} is internal, but is only used for high humidity or condensation warnings.



If **Average** is selected, T_{leaf2} and T_{leaf} are averaged for computations and control, but still considered individually for high humidity warnings.



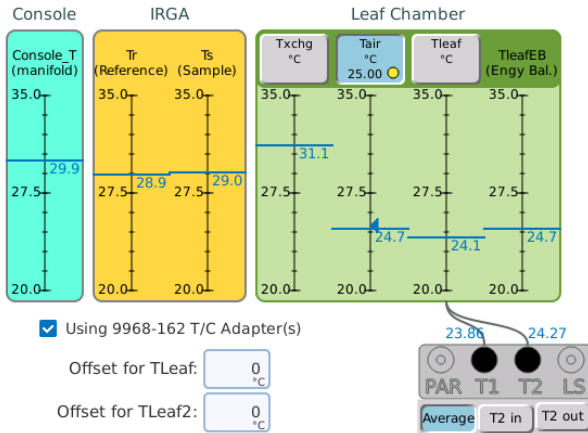
The variable that shows the source of the leaf temperature is $T_{leafCnd}$.

What happens when averaging is enabled and one or more thermocouple is removed? For temperature control, the temperature controller will continue to control temperature using whichever thermocouple remains. If both are removed, the controller will use chamber air temperature for its target. The value for $T_{leafCnd}$ works the same way, except that if no thermocouple are connected, the energy balance is used. Plugging the thermocouples back in will restore the use of T2 (averaging, in, or out) to what it was when it was unplugged.

For the Excel file, the provisions for averaging via the weighting factors for getting from T_{leaf} , T_{leaf2} , and T_{leafEB} , to the temperature used for gas exchange computations ($T_{leafCnd}$) are unchanged.

Note: The structure of the equations in the Excel files is such that you can change what you use for leaf temperature after recording the data.

When using the 9968-162 Thermocouple Adapters, there is the potential for offset errors when using thermocouple with long leads. The offset should be measured by comparing the thermocouple's reading with a standard (e.g., ice bath), and entering the correction factor in **Environment > Temperature** under **Offset for TLeaf** and **Offset for TLeaf2**.



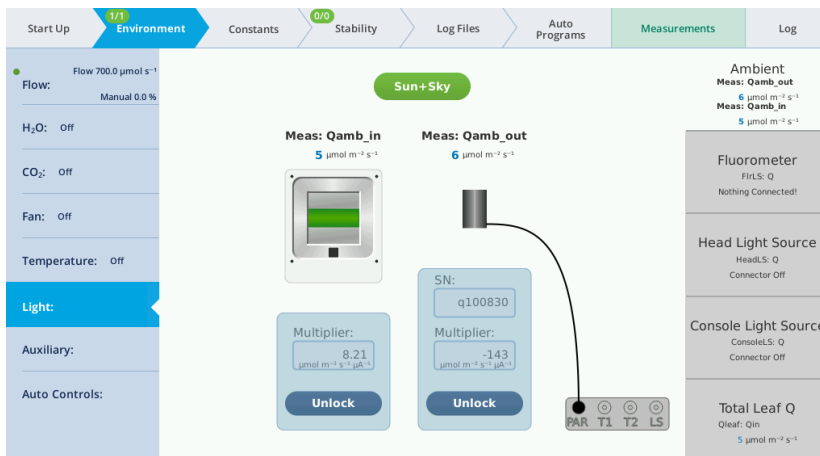
Light: Here comes the sun

In this section, we describe the **Environment > Light** tab. The options available to you depend on whether you have a light source attached, and what combination of chamber and light source you are using.

Ambient light

With a clear-top chamber, you use ambient light to illuminate the leaf. Light can be the sun, office lights, growth chamber light, or whatever light is driving photosynthesis in the sample plant. If your instrument has a light source installed, remove it for this exercise. Skip this exercise if you only have the fluorometer.

- 1 With the instrument powered on, observe the values of **Meas: Qamb_in** and **Qamb_out**. These are the measured ambient light in and out of the chamber, respectively.



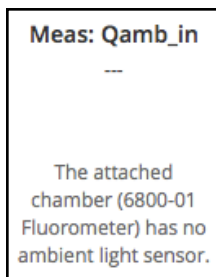
2 Select a light source.

Tap the button (labeled **Sun + Sky** in the example), then choose the option that best represents your lighting conditions.



3 Place your hand over the clear chamber top.

Observe the value of **Meas: Qamb_in** on the display. It will drop to near $0 \mu\text{mol m}^{-2} \text{s}^{-1}$ when the chamber is covered. The chambers with ambient light sensors are the 6800-12, 6800-12A, and 6800-13. If your chamber does not have a built-in light sensor, this box will be empty.



4 Now place your hand over the LI-190R sensor.

Observe the value of **Meas: Qamb_out** on the display. It will drop to near $0 \mu\text{mol m}^{-2} \text{s}^{-1}$ when the sensor is covered. You may also see **Meas: Qamb_in** drop if your hand shades the chamber.

Light source

Under **Environment > Light**, you'll find 5 subtabs where you can configure light settings. You may see some blank tabs depending on which light source is connected. For example, if you have a fluorometer connected, the Head Light Source and Console Light Source tabs will be blank.

The **Environment > Light > Head Light Source** tab is where you configure the light source that is attached to the **LS** connector underneath the sensor head. The tab called **Console Light Source** is to configure a light source connected to the **ACCESSORY** connector on the console.

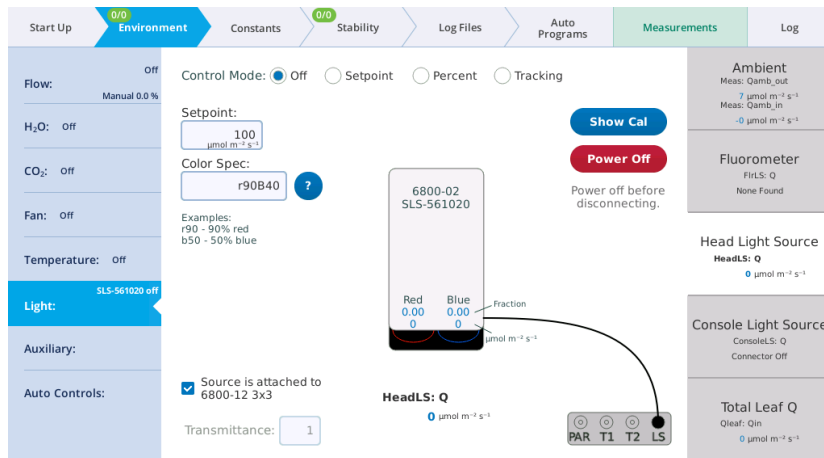


Figure 3-12. The **Environment > Light > Head Light Source** tab when the $3 \times 3 \text{ cm}$ light source is installed.

You specify the **Control Mode** for light sources that are connected to the head or console connector. The options are:

- **Off:** Disables the light source LEDs.
- **Setpoint:** Lets you enter the quantity of photons (**Q**) in $\mu\text{mol m}^{-2} \text{s}^{-1}$ in the Setpoint field.

- **Percent:** Controls the light source as a percentage of maximum drive voltage to the LEDs.
- **Tracking (3×3 and 6×6 cm light sources only):** Generally this is set to track **Qamb_out**, as measured by the LI-190R. When tracking is selected, the **Setpoint** field will continually come from the external quantum sensor (editing is disabled). To track ambient light with the fluorometer, see *Control light by tracking ambient (fluorometer)* on page 3-46.

In any **Control Mode**, you can set the **Color Spec** using a simple shorthand, which is described in *Color spec* on page A-1.

Show Cal is where you set the calibrations of the light source. These fields are populated automatically, so you will not need to edit these fields under normal circumstances.

Power On/Off is so you can connect or disconnect a light source without turning the instrument off. Always power off a light source before connecting or disconnecting the cable.

Transmittance: A transmittance factor to account for any losses between the lamp window and the leaf.

Light source exercises

In this exercise, you'll need a light source to be installed. In this exercise, we describe the 3×3 cm light source. The 6×6 cm light source is configured similarly, with more options for the green and white light. For this exercise, open the chamber so you can see the light that shines out of it. ***Remember, never look directly into the light source.***

Control light on a setpoint

With the **Setpoint** control, you can set the light intensity (**Setpoint**; $\mu\text{mol m}^{-2} \text{s}^{-1}$) and adjust the proportion of red and blue (**Color spec**). The variables **Red** and **Blue** indicate the fraction of each color.

1 Under Environment > Light > Head Light Source, select Setpoint.

The screenshot displays the 'Environment' tab with 'Light' selected. The 'Head Light Source' section is active, showing the 'Setpoint' control mode selected. The setpoint value is 100 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The 'Color Spec' is set to r90B10. A diagram of the 6800-02 SLS-561020 light source is shown with a red filter (0.90) and a blue filter (0.10). The 'HeadLS: Q' is set to 100 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The interface also shows various measurement readouts on the right side.

2 For the Setpoint, enter 100 $\mu\text{mol m}^{-2} \text{s}^{-1}$.

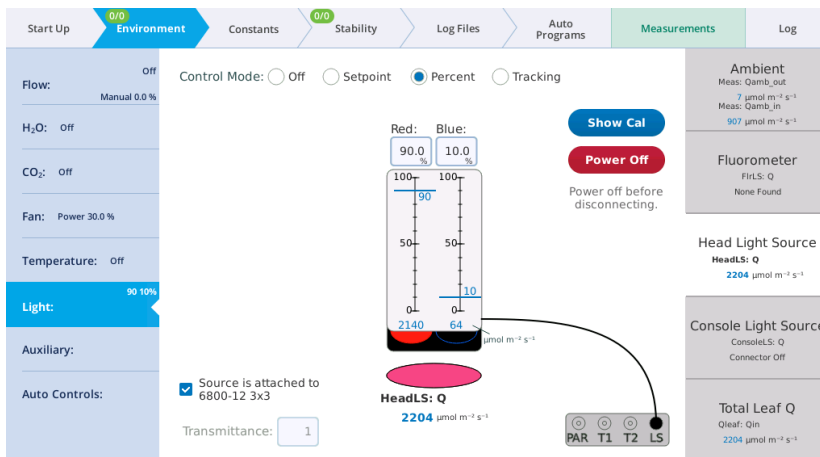
Observe the light that shines out of the chamber.

3 If it is not already set, enter r90 for the Color Spec.

This sets the ratio to red: 0.9 and blue: 0.1, which is a common ratio for photosynthesis measurements.

Control light on a percent

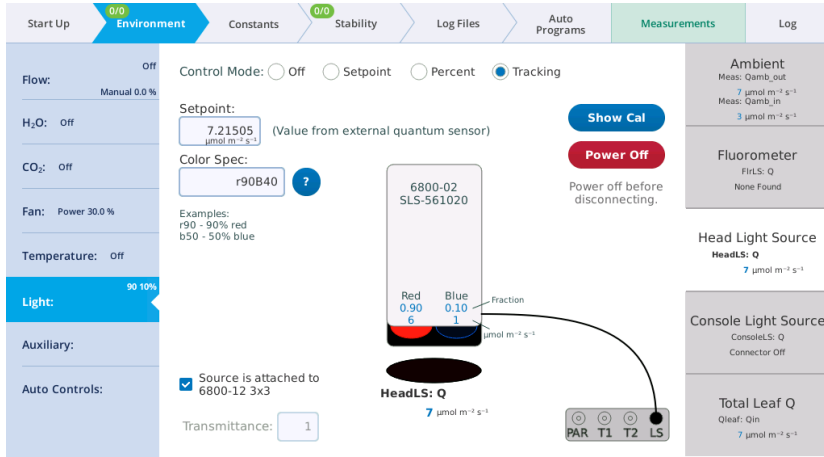
With the **Percent** control, you can set the red and blue as a percentage of power available. Maximum irradiance is achieved when you enter 100 for both red or blue.



Observe the relationship between the percentages entered and the measured light. Powering the red LEDs at 75% and the blue LEDs at 25% gives a **Head LS: Q** of about $2575 \mu\text{mol m}^{-2} \text{s}^{-1}$ in this example. The exact values for your instrument will be different.

Control light by tracking ambient (3×3 or 6×6 cm light sources)

When using an 3×3 cm or 6×6 cm light source, the **Tracking** control sets the light source brightness to track ambient light, as measured by the LI-190R quantum sensor. With the 3×3 cm light source, you can control red and blue. A typical Red:Blue ratio is 9:1, which is established by a **Color Spec** of r90. The 6×6 cm light source and fluorometer provide more color controls.



- 1 Observe the values for **Setpoint** and **HeadLS: Q**.
The **Setpoint** value is driven to measured Q_{amb_out} . In this example it is $7.16 \mu\text{mol m}^{-2} \text{s}^{-1}$ from the fluorescent lights in the LI-COR office.
- 2 Place your hand over the LI-190R to block the light.
Observe **Head LS: Q**. It should quickly drop to near $0 \mu\text{mol m}^{-2} \text{s}^{-1}$.
- 3 Lift your hand off of the light sensor and watch the **Setpoint** and **Head LS: Q** return to the previous value.

Control light by tracking ambient (fluorometer)

With the fluorometer, tracking ambient light is configured with an **Auto Control**. To track ambient, go to **Environment > Auto Controls**, select a parameter (1 to 6), and set the **Target** to control **Env Light Controls > Qin**. Set the **Function F(t)** to tracking. Set **Track this variable** to Q_{amb_out} . Check the **AutoRepeat** box, and then tap **Start**. When tracking ambient with this configuration, the **Environment > Light** tab will indicate that these parameters are under the control of the **Auto Control**.

Transmittance factors

Transmittance factors allow you to think of light source setpoint targets as what the leaf is receiving, which may or may not be what the light source is actually producing. The fluorometer and 3×3 cm light source are calibrated with chamber tops in place, so their transmittance factors are 1.0. This is not necessarily true when apertures are used. The 6×6 cm light source is calibrated without any chamber top,

since there are multiple chambers it fits on. It has transmittance factors for each chamber type.

When you connect a chamber (or specify a chamber ID under **Start Up > Chamber Setup**) the LI-6800 will automatically choose the appropriate transmittance factor for that chamber. If you suspect that the transmittance factor is not correct, you can verify it from *Table 3-3* below.

Table 3-3. Transmittance for attached light sources.

Chamber	Light Source		
	6800-01	6800-02	6800-03
6800-01 fluorometer	1.0	-	-
6800-01A with 6 cm ² aperture	1.0	-	-
6800-01A with 2 cm ² aperture	0.87	-	-
6800-12 3×3 cm chamber	-	1.0	-
6800-12A with 3×3 cm aperture	-	1.0	-
6800-12A with 2×3 cm aperture	-	1.01	-
6800-12A with 1×3 cm aperture	-	0.98	-
6800-13 6×6 cm chamber	-	-	0.81
6800-17 small plant chamber	-	-	0.86

If you have a custom method of illuminating a chamber (for example, using a 6×6, 4 color light source (6800-03) suspended above a 3×3 chamber), you will need to measure the transmittance factor for that configuration. A simple way to estimate this is to set the light source to 1000 mol m⁻² s⁻¹, then compare the reading of a quantum sensor that is pressed against the face of the light source (e.g., 1082 mol m⁻² s⁻¹) with the reading beneath the top of the chamber at leaf level (e.g., 774 mol m⁻² s⁻¹). The transmittance is the ratio: 774/1082 = 0.72. Once that transmittance is entered, setting the light source to 500 mol m⁻² s⁻¹, will cause it to actually put out 500/0.72 = 694 at its face, in order to achieve 500 at the leaf surface.

Light source factory calibration settings

The **Show Cal** button gives access to light source factory default settings and calibration data. The information in this tab is for diagnostic purposes. Do not change it unless in consultation with technical support.

Total leaf Q

The fifth tab, **Total Leaf Q**, is where you specify which light source(s) contribute to the light on the leaf. The **Customize** button allows you to weight each light source's contribution to the leaf, to accommodate nearly any scenario. For example, a 3×3 cm or 6×6 cm chamber with a light source mounted on the top and bottom will be configured this way. You can **Use Default** values for the light sources, or **Customize** them by entering a multiplier.

Light source test program

We have added a **Test** button to light source control screens that launches a background program that checks some of the calibration coefficients (u_1 ; u_2 ; u_3). This test is only available to light sources that attach to the head or console (not the MPF fluorometer).

The values in u_0 are found by determining each color control's on/off threshold. The other values (u_1 ; u_2) relate control setting to color output, and are important for determining color mix to achieve the specified value. These values should be fairly stable with time, but are easily checked. The test that is launched (`/home/licor/apps/tech/LightSourceCal.py`) is best done with the light source in the dark; if the light source is attached to a chamber, then clamping the chamber onto something very unreflective (black felt, for example) is recommended.

The image shows a control interface with several panels. On the left, a 'Factory Calibration' panel displays fields for id (6800-03), sn (181005), f (0.0684 0.0863 0.1094 0.0764), u0 (299 348 999 423), u1 (0.94820 0.61932 0.51449 0.94112), and u2 (-1.7364e-06 -4.947e-06 -1.8674e-06 -5.0974e-06). A red circle highlights the u0, u1, and u2 fields. Below this is an 'Unlock' button. To the right of the calibration panel is a 'Color Spec' section with a dropdown set to 'r90' and a help icon. Below that is a 'Light Source Response Checker (BP#5)' dialog box with the title 'Test a light source response' and the text 'Factory calibration coefficients u0, u1, and u2 are tested. Best done with chamber closed on black felt.' The dialog has a dropdown menu for 'Light source attached to:' set to 'head' and 'Continue' and 'Cancel' buttons. In the background, a main control panel shows tabs for 'Stability', 'Log Files', 'Auto Programs', 'Measurements', and 'Log'. Below the tabs are radio buttons for 'Setpoint', 'Percent', and 'Tracking'. A 'Show Cal' button is highlighted with a red circle, and an arrow points from it to the 'Test' button in the dialog. Below the 'Show Cal' button are 'Test' and 'Power Off' buttons. A 'Power off before disconnecting.' warning is visible. To the right of the main panel are several measurement readouts: 'Ambient' (Meas: Qamb,act 10 μmol m⁻² s⁻¹), 'Fluorometer' (FHL5: Q 500 μmol m⁻² s⁻¹), 'Head Light Source' (HeadL5: Q 0 μmol m⁻² s⁻¹), 'Console Light Source' (ConsoleL5: Q Connector Off), and 'Total Leaf Q' (Qleaf: Qin 500 μmol m⁻² s⁻¹). A 'Light Source' (LS) icon is also visible.

Figure 3-13. The test button will launch a BP that checks a light source.

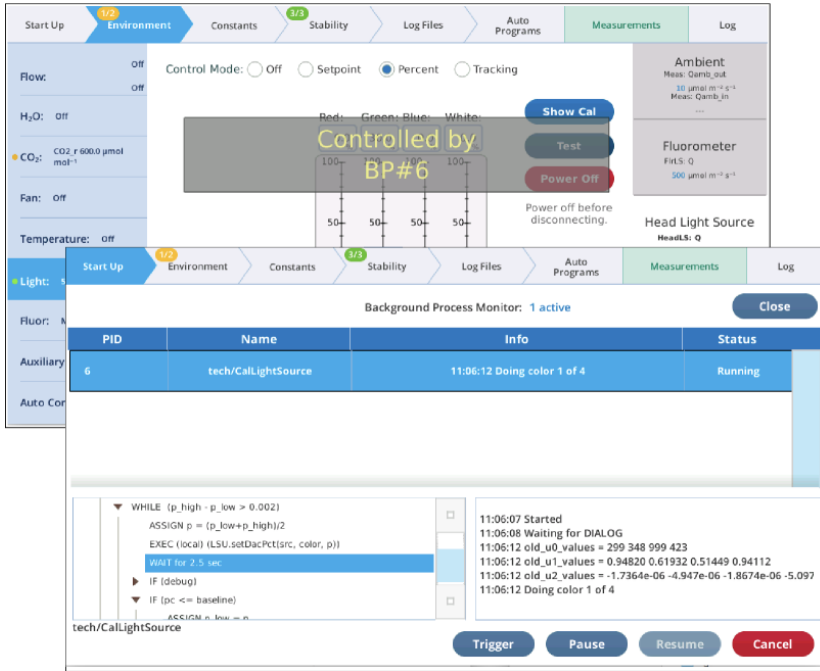


Figure 3-14. While the program is running, progress can be monitored in the Monitor screen, or on the light control panel screen.

When it is done, another dialog is presented showing the results. Tapping **Install** will update the coefficients with those that were just calculated.

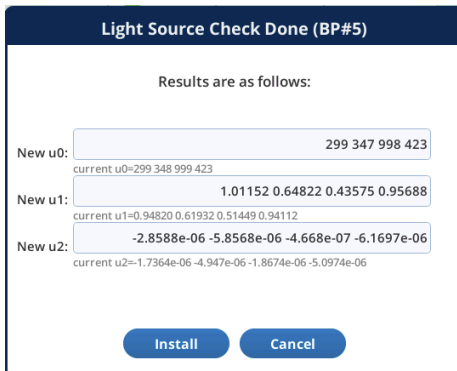


Figure 3-15. LightSourceCal closing dialog

Multiphase Flash™ Fluorometer: Blinded by the light

If you have the fluorometer attached, you'll see the **Fluor** tab on the left. Here you can configure the fluorometer. Some fluorometer settings are configured through the **Environment > Light > Fluorometer** tab, which takes this form when the fluorometer is attached:

The screenshot displays the configuration interface for the Multiphase Flash Fluorometer. The interface is organized into several sections:

- Navigation Tabs:** Start Up, Environment (1/1), Constants (0/0), Stability, Log Files, Auto Programs, Measurements, Log.
- Control Mode:** Radio buttons for Off (selected), Setpoint, Percent, and Test.
- Setpoint:** A text input field containing the value 8, with units $\mu\text{mol m}^{-2} \text{s}^{-1}$.
- Color Spec:** A text input field containing the value r90.
- Recommended color spec:** A note stating: "Recommended color spec: r90B40 when combining gas exchange and fluorescence. This produces 90% red (or more), with a blue upper limit of $40 \mu\text{mol m}^{-2} \text{s}^{-1}$ ".
- Fluorometer Model:** 6800-01 MPF-551018.
- Fluorometer Channels:** A graph showing Red (0.00), Blue (0.00), and Farred (0.00) channels. A 'Fraction' label points to the graph.
- Source Status:** A checked box indicating "Source is attached to 6800-01 Fluorometer".
- Transmittance:** A text input field containing the value 1.
- Show Cal:** A blue button to initiate calibration.
- Measurement Summary (Right Side):**
 - Ambient:** Meas: Qamb_out, Qamb_in (all 0).
 - Fluorometer:** Firs: Q (0).
 - Head Light Source:** HeadLS: Q, Connector OFF.
 - Console Light Source:** ConsoleLS: Q, Connector OFF.
 - Total Leaf Q:** Qleaf: On (0).

The options for **Control Mode**, the **Setpoint** field, the **Color Spec** field, **Transmittance**, and **Show Cal** are all the same as those described in *Light source* on page 3-42.

Setup screens

Each setup screen has a graph that shows the current settings, and also the results of the last event of that type that was made. The time of the fluorescence peak (or minimum, for a dark pulse) is called out on the graph, to aid in setting proper duration times. For flash events, the displayed F data are only those points during the flash. This has the affect of zooming in on the upper part of the response. (For more controlled zooming and other options for fluorescence plots, see the **Files Screen**.)

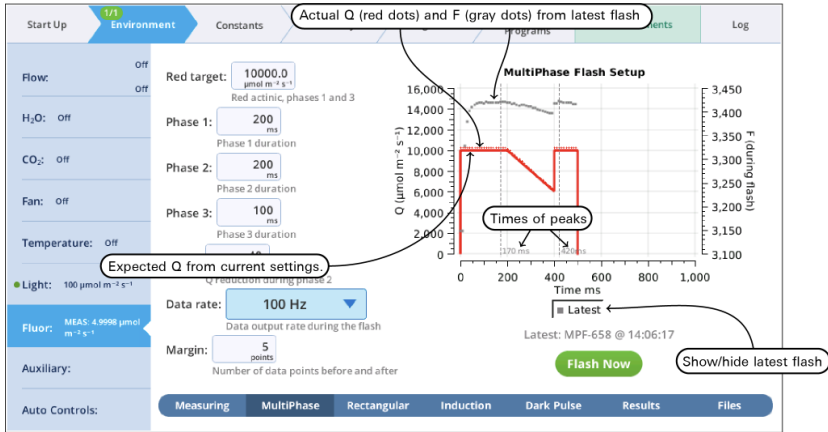


Figure 3-16. An example setup screen for MultiPhase flashes.

Results screen

The **Results** screen shows the current state of the fluorescence computations (Group name = Flr), and the dark adapted, light adapted, and dark pulse events on which they are based.

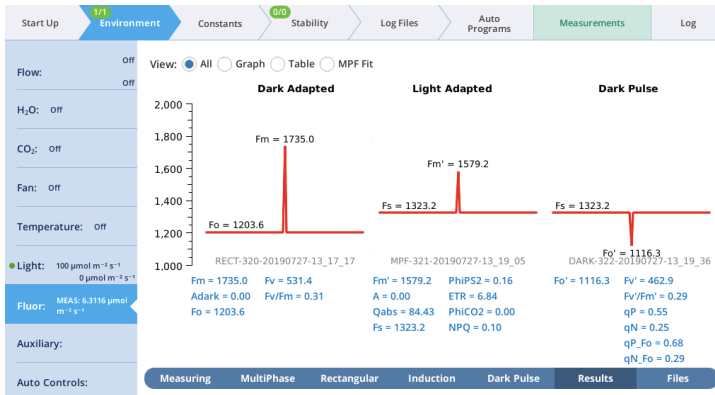


Figure 3-17. The state of the Flr computational group.

When fluorescence events are triggered manually from their setup pages, the dark adapted, light adapted and dark pulse sections are treated independently. When events are triggered as part of logging to a file, the following rules are followed.

- A log that triggers an FoFm will clear the light adapted and dark pulse sections.
- A log that triggers an FsFm' will update the light adapted, but clear the dark pulse section.
- A log that triggers an FsFm'Fo' will update the light adapted and dark pulse sections.

Light adapted vs dark adapted is determined by the following: A flash is dark adapted if the fluorometer actinic is off, or else the measured Q is less than $1 \text{ mol m}^{-2} \text{ s}^{-1}$ at the time the flash occurred.

Files screen

The **Files** screen allows you to view and compare up to 8 fluorescence events, either by loading them from file, or by capturing when they occur.



Figure 3-18. The Files Screen can hold up to 8 events.

Graph

Variables that can be plotted are Q (actinic light), F (modulated fluorescence) and DC=Q (non-modulated fluorescence normalized by non-modulated Q).

- $\text{Log}_{10}(\text{time})$ - Apply a log scale to the time axis, which is useful for looking at an induction flash.

- **Normalize** - Plot each y axis variable as scaled by the maximum value in the data set. In the case of F for a flash event, it is scaled by F_{max} .
- **Zoom up** - Change the scaling of each Y axis so that only the top x% of the data is shown.

Figure 3-19 below illustrates these options.

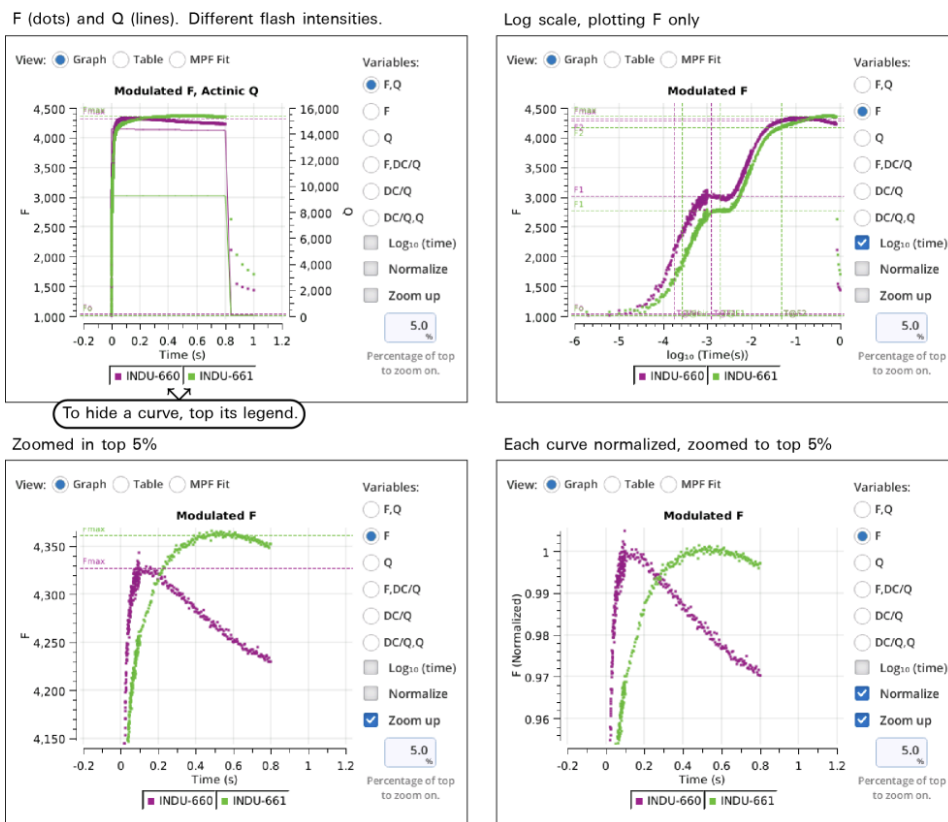


Figure 3-19. Some plot modifiers illustrated using two induction curves.

Table

The **Table** screen shows all events in tabular form. All single-value items from the fluorometer event file are shown in alphabetical order. If more than one type of event file (MPF, Rectangular, Induction, Dark Pulse) is present, some entries will be blank.

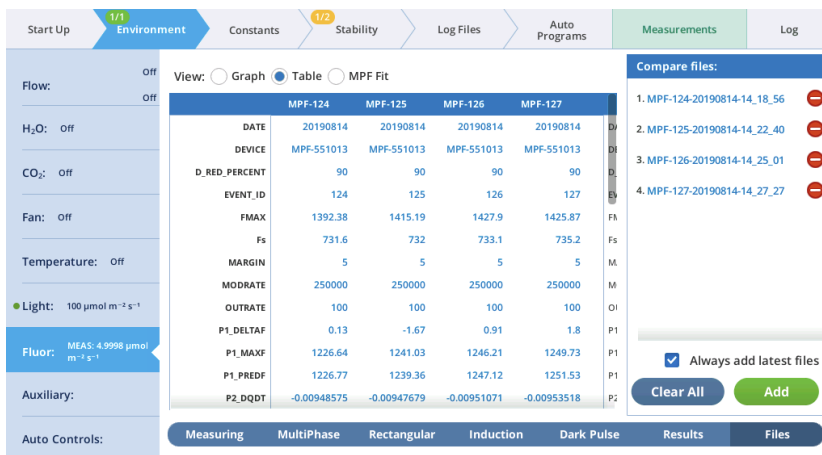


Figure 3-20. The Table view shows all of the single-valued entries in the fluorescence event file.

See *The Constants tab* on page 3-74 for a description of what the row labels mean. The last row (VERSION) will show 1 for new files, or 1* for pre-version 1 files. Such files are temporarily converted to version 1 when loaded so will appear in the table with all the new entries filled in.

MPF fit

MultiPhase Flash events in the Compare files list will appear on the **MPF Fit** screen, whose graph shows the Phase 2 regressions. In addition, the Fmax value of the flash is shown (Fmax is whichever is larger: the extrapolated Phase 2 value (P2 INT), or the Phase 1 max (P1 MAXF)). Two validity indicators are also shown, based on the phase 2 regression: the predicted Phase 1 max (P1 PREDF) and the predicted phase 3 max (P3 PREDF). The closer they are to the line the better the phase 2 regression is in predicting phase 1 and phase 3 fluorescence.

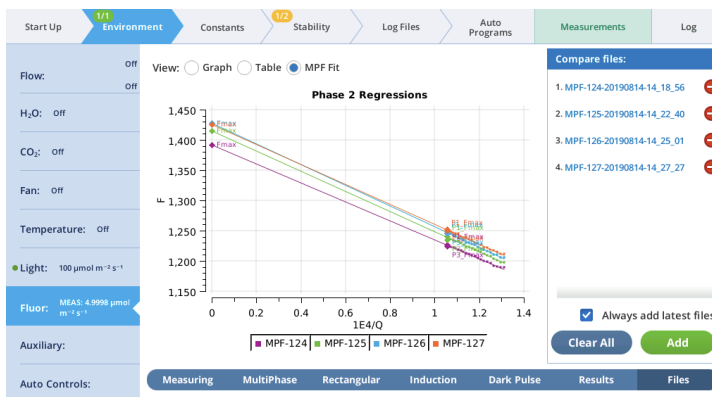


Figure 3-21. The phase 2 regressions of MPF events.

Actinic light exercise

Under **Environment > Light > Fluorometer** you can set the actinic light by setting the Control Mode. Actinic refers to continuous light for purposes of driving photosynthesis, and it can be set to **Off**, **Setpoint**, **Percent**, and **Test**. The LEDs that provide actinic irradiance are the same ones used for saturating flashes, plus several blue LEDs (475 nm). You control the light in one of two ways: Selecting and entering a **Setpoint** and **Color Spec**, or selecting and entering the **Percents**. The options for **Color Spec** are given in *Color spec* on page A-1.

This tutorial steps you through actinic light controls. Open the fluorometer chamber so you can see light shining out of it.

1 Set the Control Mode to Setpoint and then configure the Color Spec:

- **Setpoint:** $100 \mu\text{mol m}^{-2} \text{s}^{-1}$
- **Color Spec:** r90b10

Observe the light shining out of the chamber (never look directly into the chamber). Now look at the measured values for Red, Blue, and Farred.

The screenshot shows the 'Environment' tab with 'Light' selected. Under 'Light', the 'Fluorometer' section is active, showing 'Color Spec: r90b10' and 'Firls: Q 100 μmol m⁻² s⁻¹'. A 'Measured values' callout points to a color calibration chart with the following data:

Color	Fraction
Red	0.90
Blue	0.10
Farrd	0.00

The chart also shows 'Firls: Q 100 μmol m⁻² s⁻¹' and a 'Show Cal' button.

2 Change the Color Spec to r90b40.

Observe the light. It is now 69% red and 31% blue. Check the measured values on the display.

3 Set the Color Spec to r90b40.

With this setting, the LI-6800 provides 90% red with a maximum of 40 μmol m⁻² s⁻¹ blue. This is a typical ratio for combined gas exchange and fluorescence measurements.

The **Show Cal** button under the **Environment > Light > Fluorometer** gives access to factory default settings and calibration data for the fluorometer. The information in this tab is for diagnostic purposes. Do not change it unless in consultation with technical support.

Measuring light exercise

The **Fluor > Measuring** tab is where you configure the pulse amplitude modulation (PAM) rate and intensity. You can control the following:

- **Dark mod rate:** Dark modulation rate, the modulation rate when actinic light is zero. It can be set to 50 Hz, 100 Hz, 200 Hz, 500 Hz, 1 kHz, 2 kHz, and 5 kHz.
- **Light mod rate:** Light modulation rate, the modulation rate when actinic light is greater than zero. Light modulation rates are 1, 2, 5, 10, 20, and 50 kHz.
- **Flash mod rate:** Flash modulation rate, is the modulation rate during a flash.
- **Averaging:** The averaging period for F_o (minimal fluorescence) and F_s (steady-state fluorescence). The default averaging time is 15 seconds.

The screenshot shows the 'Environment' tab of the instrument control software. On the left, there are controls for Flow, H₂O, CO₂, Fan, Temperature, and Light. The 'Light' control is set to 100 μmol m⁻² s⁻¹. The 'Fluor.' control shows a measurement of 0.1000 μmol m⁻² s⁻¹. The 'Measuring Beam' is set to 'On'. The 'Dark mod rate' is 500 Hz, 'Light mod rate' is 1 kHz, and 'Flash mod rate' is 125 kHz. The 'Averaging' is set to 15 s. The 'Record fluorometer trace' section has 'Start' and 'Stop' buttons. The 'Auto Controls' section has tabs for Measuring, MultiPhase, Rectangular, Induction, Dark Pulse, Results, and Files. On the right, there are three measurement displays: 'Q_modavg' (0.1000 μmol m⁻² s⁻¹), 'F' (6.0 FIrLS), and 'F_dc' (108.4 FIrLS).

The displayed variables are:

- **Q_modavg**: The contribution to Q from the modulated light. This value will be non-zero when the measuring light is on.
- **F**: Demodulated fluorescence signal.
- **F_dc**: DC fluorescence.

Note: You can continuously record the fluorescence data into a file by tapping **Start** under **Record fluorescence trace**. This resulting data file is described in *The continuous fluorescence trace* on page 6-28.

This tutorial steps you through the **Light** and **Measuring** light controls. It will show how the modulation rate affects F and the light contributed by the fluorometer. The chamber can be open or closed for this exercise.

- 1 Set up a graph that shows the variable F on the left axis and Q_modavg on the right, with *Time* as the horizontal axis.

Under the **Measurements** tab, select graph **F** to make it easy to remember. Tap **Edit graphs**, then change the first variable to F (under FIrLS). Plot it on the left axis, scaled from -10 to 10. Add a second variable, Q_modavg , and plot it on the right axis, scaled from 0 to 5.

The screenshot shows the 'Edit graphs' interface. At the top, it says 'FIrLS:F vs. Meas:TIME'. There is a 'Show legend' checkbox and a 'Fixed' button. A 'Plot' button is also visible. Below the plot area, there are two rows of variables: 1. F (with a purple square icon) and 2. Q_modavg (with a green square icon). There are also some icons for adjusting the scale and legend. Annotations with arrows point to the 'Plot' button and the 'Fixed' button, with text: 'Tap the switch to plot variables against time' and 'Tap Fixed (or whatever setting is displayed) to adjust the scale'.

- 2 Under **Environment > Light > Fluorometer**, turn **Control Mode** off.
- 3 Under **Environment > Fluor**, turn the **Measuring Beam** to **On** and configure the fluorometer:
 - **Dark mod rate:** 50 Hz
 - **Light mod rate:** 1 kHz
 - **Flash mod rate:** 250 kHz
 - **Averaging:** 15 s

Observe the graph of F and Q_modavg . Expect to see a noisy fluorescence signal and Q_modavg near 0.

- 4 Change the **Dark mod rate** to 500 Hz.
Observe the graph. F will become less noisy and Q_modavg will increase.
- 5 Change the **Dark mod rate** to 5 kHz.
Observe the graph. Again, F will become less noisy and Q_modavg will increase.
- 6 Turn the **Measuring beam** off.
- 7 Under **Environment > Light**, set **Control Mode** to **Setpoint** and enter $200 \mu\text{mol m}^{-2} \text{s}^{-1}$.
This establishes an actinic light level of $200 \mu\text{mol m}^{-2} \text{s}^{-1}$.

- 8 Under **Environment > Fluorometer**, change the measuring beam settings:

- **Dark mod rate:** 50 Hz
- **Light mod rate:** 1 kHz
- **Flash mod rate:** 250 kHz
- **Averaging:** 15 s

Observe the graph. F will be noisier than before and Q_modavg will return to 0.

- 9 Change the **Light mod rate** to 10 kHz.
Observe the graph. F will become less noisy and Q_modavg will remain 0.
- 10 Change the **Light mod rate** to 50 kHz.
Observe the graph. F will become less noisy and Q_modavg will remain 0.
- 11 Now you'll view the **Measuring** and **Actinic light** together. Turn the **Measuring Beam** back **On** and change the **Measuring Beam** settings:
 - **Dark mod rate:** 50 Hz
 - **Light mod rate:** 1 kHz
 - **Flash mod rate:** 250 kHz
 - **Averaging:** 15 s

Turn both the **Measuring** beam and **Actinic** On. Observe the graph. F will be less noisy and Q_modavg will rise to about 0.1.

12 Change the **Light mod rate** to 10 kHz.

Observe the graph. F will become less noisy and Q_modavg will rise to about 1.

13 Change the **Light mod rate** to 50 kHz.

Observe the graph. F will become less noisy and Q_modavg will rise to about 5. The expected values are shown in *Figure 3-22* below.

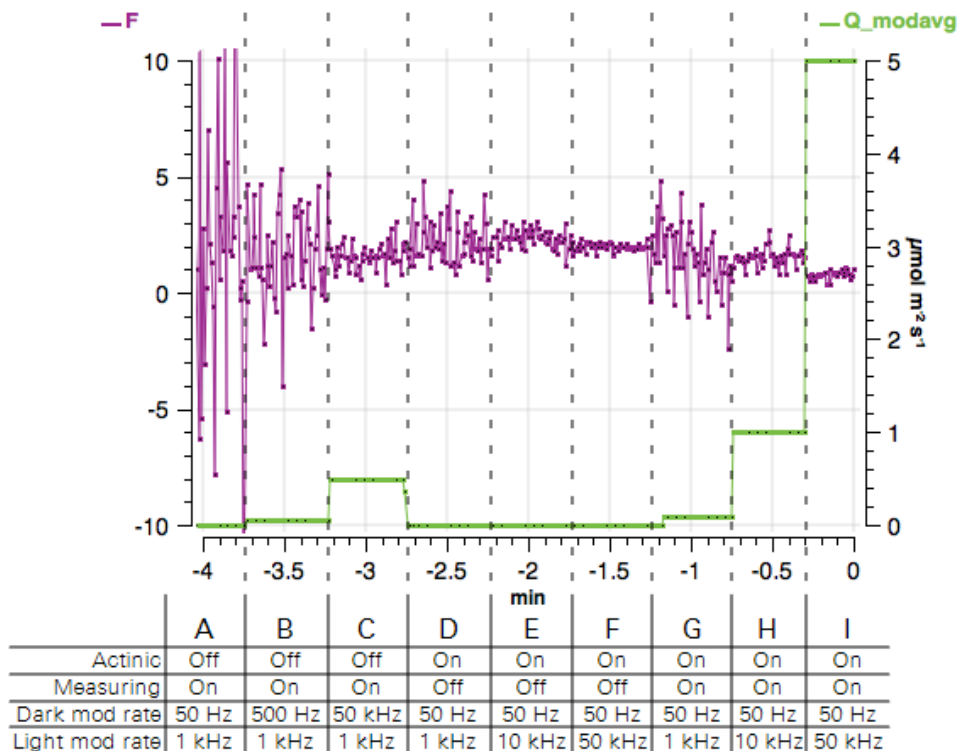


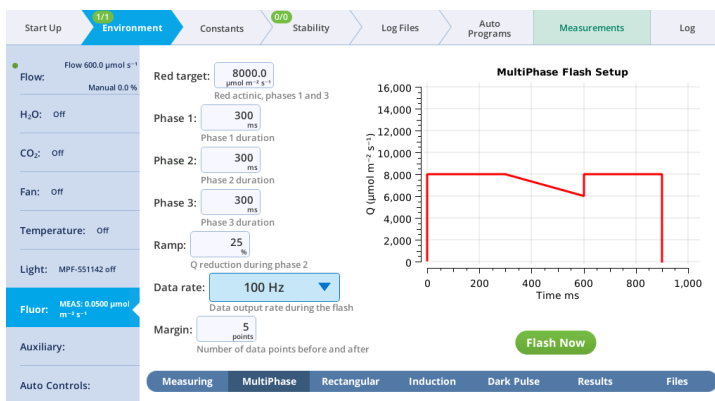
Figure 3-22. The parameters we configured in this exercise. Notice how an increase in the modulation rate decreases the variation in the fluorescence signal (F) but increases the average light (Q_modavg).

Multiphase flash exercise

This tab is where you configure a multiphase flash event. The Multiphase Flash™ Fluorometer uses varying intensities during the flash, and computes the

fluorescence at saturation from the y-intercept of a plot of fluorescence vs. $1/\text{intensity}$. You can control the following:

- **Red target** is the setpoint for red actinic light during phases 1 and 3.
- **Phases 1, 2, and 3** are time (ms) for the initial peak, ramp, and final peak, respectively.
- **Ramp** is the percentage decrease in flash intensity from the red target value during phase 2.
- **Output rate** is the data output rate during the flash.
- **Margin** is the number of points plotted before and after the flash.



1 Make a dark-adapted measurement.

This is how you measure a leaf that is adapted to darkness. Under **Environment > Light > Fluorometer**, set **Control Mode** to **Off**. This will set the LI-6800 to make the dark-adapted measurement. We'll do a light-adapted measurement in a moment.

2 Under Environment > Fluor > Measuring, set Measuring Beam to On.

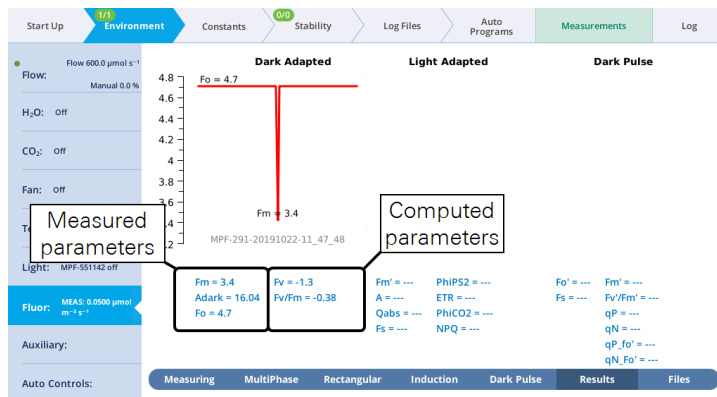
3 Under Environment > Fluor > MultiPhase, set up the multiphase flash. Do this with the chamber open.

- **Red target:** $8000 \mu\text{mol m}^{-2} \text{s}^{-1}$
- **Phase 1:** 300 ms
- **Phase 2:** 300 ms
- **Phase 3:** 300 ms
- **Ramp:** 25%
- **Output rate:** 500 Hz
- **Margin:** 5 points

4 Look toward the chamber (never look directly into the light source), and tap **Flash Now**.
If the chamber is open you'll see the flash.

5 View the **Dark-Adapted** results.

Tap **Results** to see the results from the most recent flash.

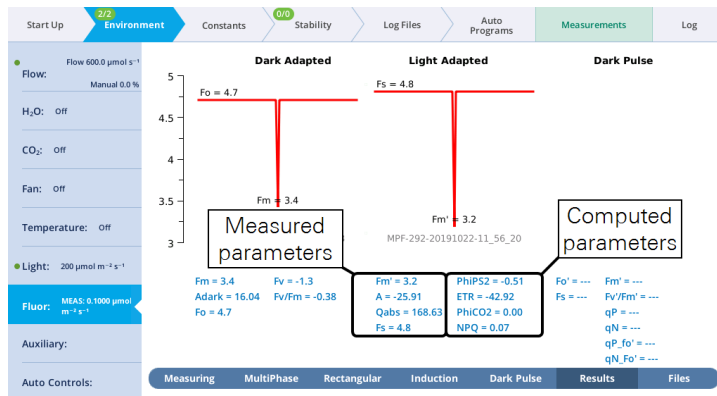


6 Make a **Light-Adapted** measurement.

Under **Environment > Light > Fluorometer**, set the **Control Mode** to a **Setpoint** of 200. This will set the LI-6800 to make the **Light-Adapted** measurement.

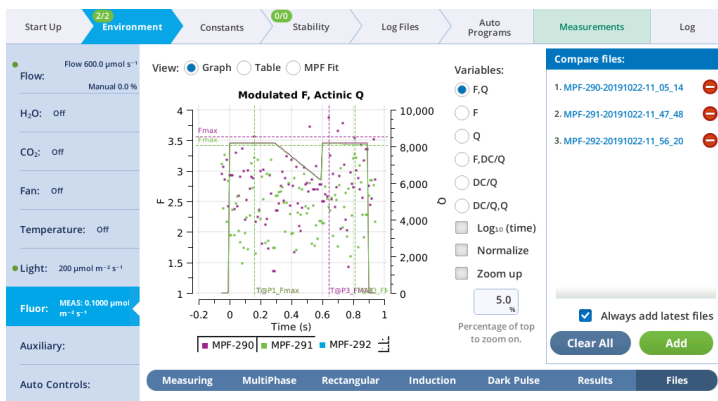
7 With the same **Multiphase Flash** settings set previously under **MultiPhase**, tap **Flash Now**.

8 View the **Light-Adapted** results.



9 Tap **Files** at the bottom right to see the flash data.

The values for Q represent the instrument's measurement of the flash. The fluorescence data will look meaningless because we did not measure a leaf.

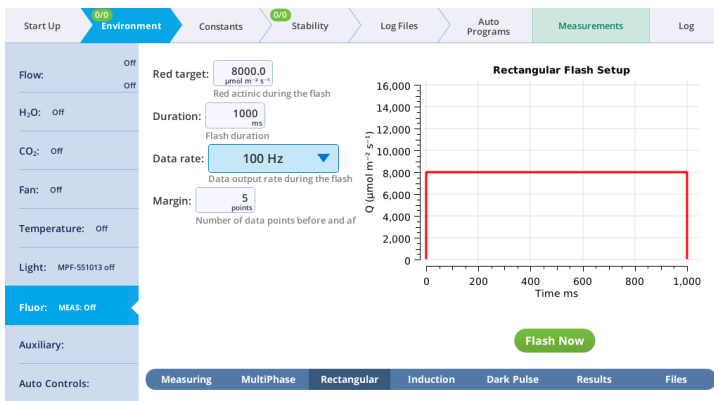


10 Review the other options.

Rectangular flash exercise

The next tab, called **Rectangular**, is where you configure a saturating flash that provides continuous light for the duration of the flash.

- **Red target** is the setpoint for red light during a flash.
- **Duration** is the time of the flash, in milliseconds.
- **Output rate** is the data output rate during a flash, in Hz.
- **Margin** is the number of data points plotted before and after the flash.



1 Make a dark-adapted measurement.

This is how you measure a leaf that is adapted to darkness. Under **Environment > Light > Fluorometer**, set **Control Mode** to **Off**. This sets the LI-6800 to make the dark-adapted measurement. We'll do a **Light-Adapted** measurement in a moment.

2 Under **Environment > Fluor > Measuring**, set **Measuring Beam** to **On**.

3 Set up a rectangular flash. Keep the chamber open.

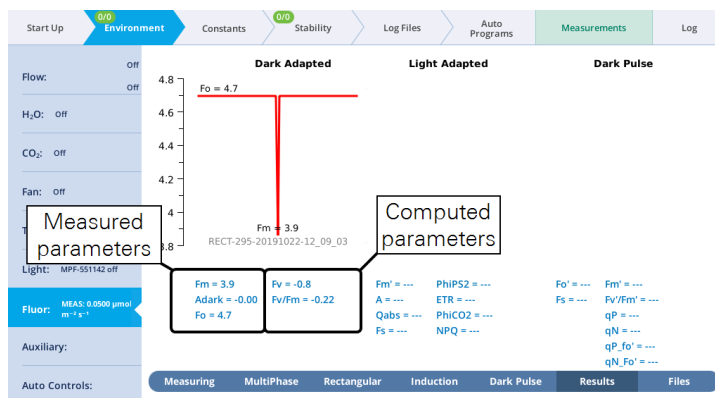
- **Red target:** $8000 \mu\text{mol m}^{-2} \text{s}^{-1}$
- **Duration:** 1000 ms
- **Output rate:** 500 Hz
- **Margin:** 5 points

4 Look toward the chamber (never look directly into the light source), and tap **Flash Now**.

You'll see the red light shine out of the chamber as the fluorometer does the flash.

5 View the **Dark-Adapted** results.

Tap **Results** to see the results from the most recent flash.

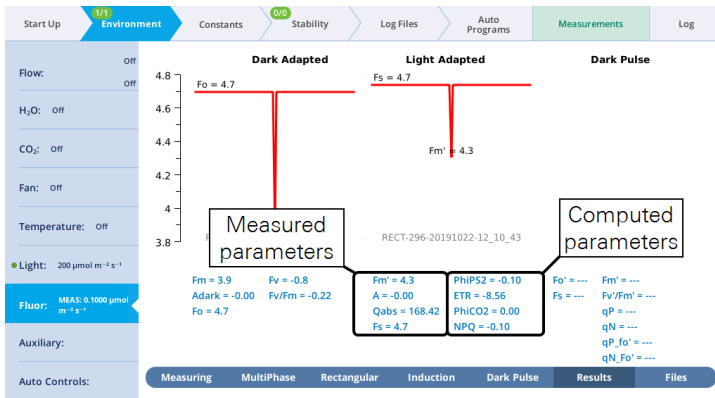


6 Make a **Light-Adapted** measurement.

Under **Environment > Light > Fluorometer**, set **Control Mode** to a **Setpoint** of 200. This sets the LI-6800 to make the **Light-Adapted** measurement.

7 With the same **Rectangular Flash** settings set previously under **Rectangular**, tap **Flash Now**.

8 View the **Light-Adapted** results.

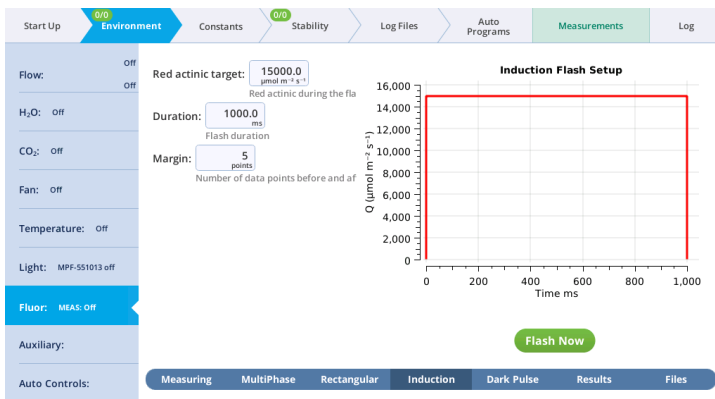


9 Tap View for a graph and details.

Induction flash exercise

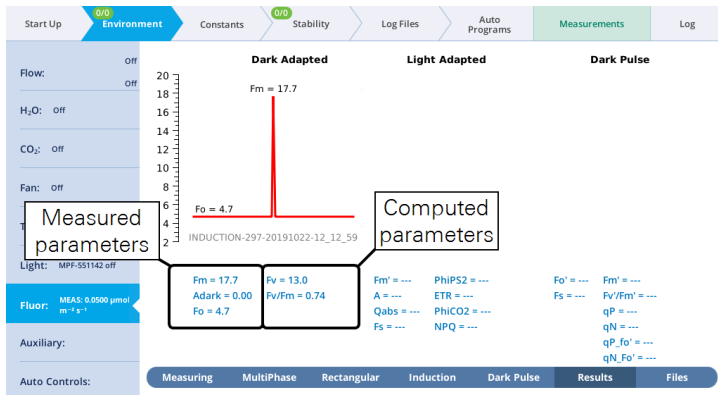
You can control the following:

- **Red target:** Red actinic light during the flash.
- **Duration:** The milliseconds of flash.
- **Margin:** Number of data points before and after the flash.



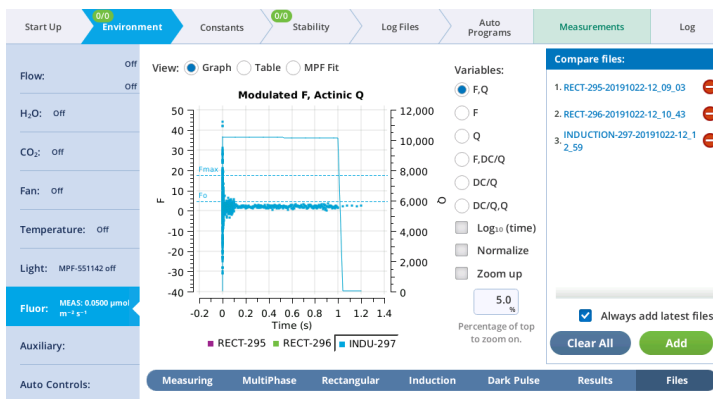
- 1 In this example, you'll make a dark-adapted measurement, so turn Actinic light off. (Environment > Light > Fluorometer, set Control Mode to Off).
- 2 Turn the measuring beam on (Environment > Fluor > Measuring, Measuring beam On).
- 3 Set up an induction flash (Environment > Fluor > Induction). Keep the chamber open.
 - **Red target:** 15000 $\mu\text{mol m}^{-2} \text{s}^{-1}$

- **Duration:** 1000 ms
 - **Margin:** 5 points
- Look toward the chamber (never look directly into the light source), and tap **Flash Now**. You'll see the flash shine out of the chamber.
 - View the induction flash results.
 - Tap **Results** to see the results from the most recent flash.



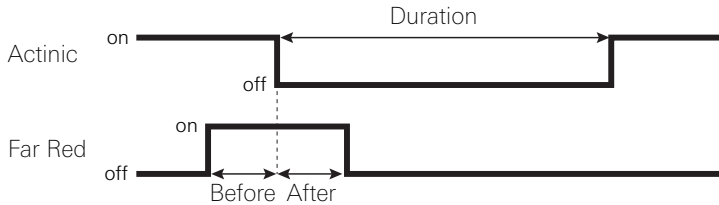
- Tap **View** under **Light-Adapted** to see the flash data.

The values for Q represent the instrument's measurement of the flash. The fluorescence data will look meaningless because we did not measure a leaf.



Dark pulse exercise

A dark pulse (brief dark period) is designed to measure Fo' on a light-adapted leaf. The timing parameters are shown in *Figure 3-23* below. You trigger a dark pulse under **Environment > Fluor > Dark Pulse**.

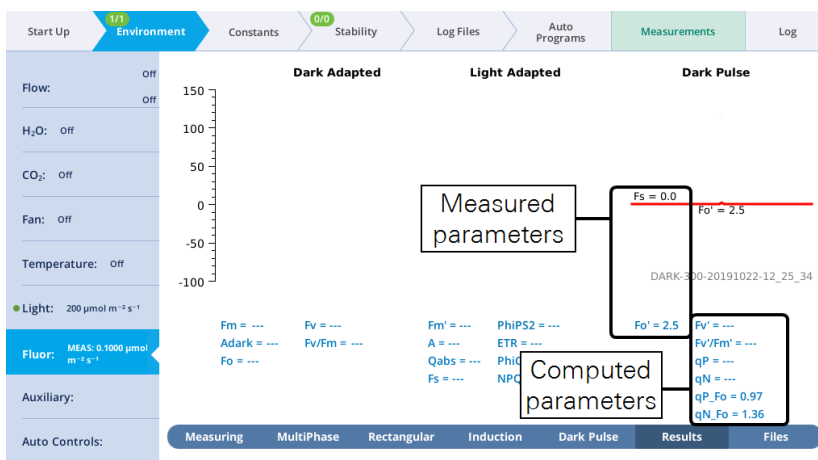


*Figure 3-23. Timing parameters for a dark pulse. The far red LED comes on a number of seconds **Before** the actinic light goes off, and remains on for a number of seconds **After**. The actinic light is off for the **Duration**.*

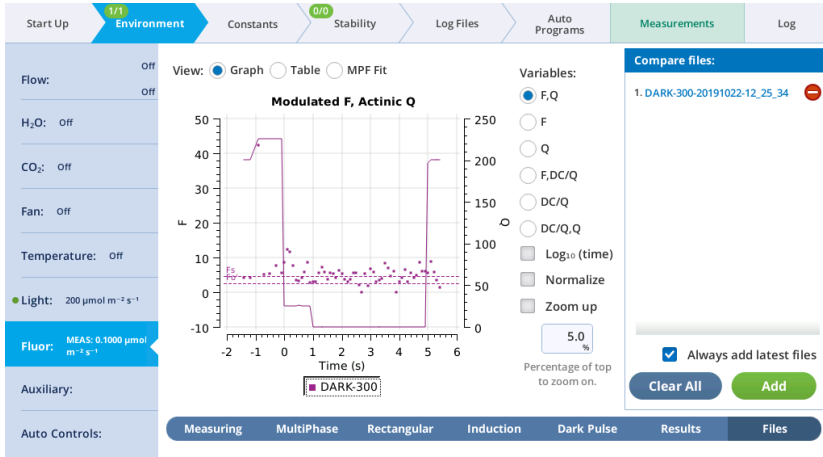
You can control the following:

- **FarRed target:** Far red target for the dark pulse (maximum of $25 \mu\text{mol m}^{-2} \text{s}^{-1}$).
- **Duration:** The milliseconds of dark pulse.
- **Before:** The number of seconds the far red is on before the actinic is off.
- **After:** The number of seconds the far red is off after the actinic is off.
- **Margin:** Number of data points before and after the flash.

- 1 Turn Actinic light on under **Environment > Light > Fluorometer**. Set the **Control Mode** to **Setpoint** of 200.
- 2 Under **Environment > Fluor > Measuring**, set **Measuring Beam** to **On**.
- 3 Under **Environment > Fluor > Dark Pulse**, set up a dark pulse. Keep the chamber open.
 - **FarRed target:** $25 \mu\text{mol m}^{-2} \text{s}^{-1}$
 - **Duration:** 5 s
 - **Before:** 1 s
 - **After:** 1 s
 - **Margin:** 5 points
- 4 Look toward the chamber (never look directly into the light source), and tap **Pulse Now**.
You probably won't see the far red, (you might if you do the flash in a dark room) but you will see the actinic light turn off.
- 5 View the dark pulse results.
Tap **Results** to see the results from the most recent flash.

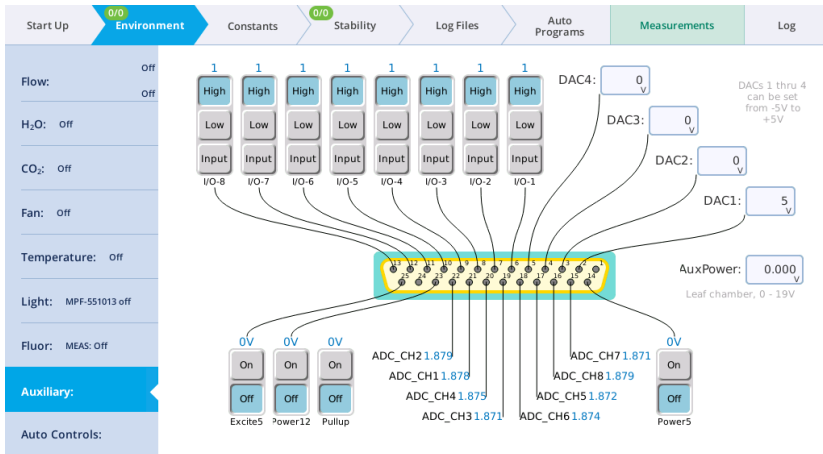


- 6 Tap **View** under **Dark Pulse** to see the flash data.
The values for Q represent the instrument's measurement of the flash. The fluorescence data will look meaningless because we did not measure a leaf.



Auxiliary channels: Release the Kraken

The LI-6800 can control peripheral components (such as mixing fans) and accept data from peripheral sensors (such as thermocouples) through the 25-pin connector. The connector is labeled **USER I/O** on the console. The outputs can also be controlled via the **AutoEnv** controls and from the generic **AutoPrograms**.



This is described in detail in *Configuring auxiliary channels* on page 4-46.

Auto controls: The LI-6800 abides

The **Auto Controls** (under the **Environment** tab) are settings that instruct the LI-6800 to control a variable setpoint based upon parameters that you specify. The auto controls can be configured to change during a prescribed time period. Auto controls will override any setpoint or manual settings on the individual environmental control tabs.

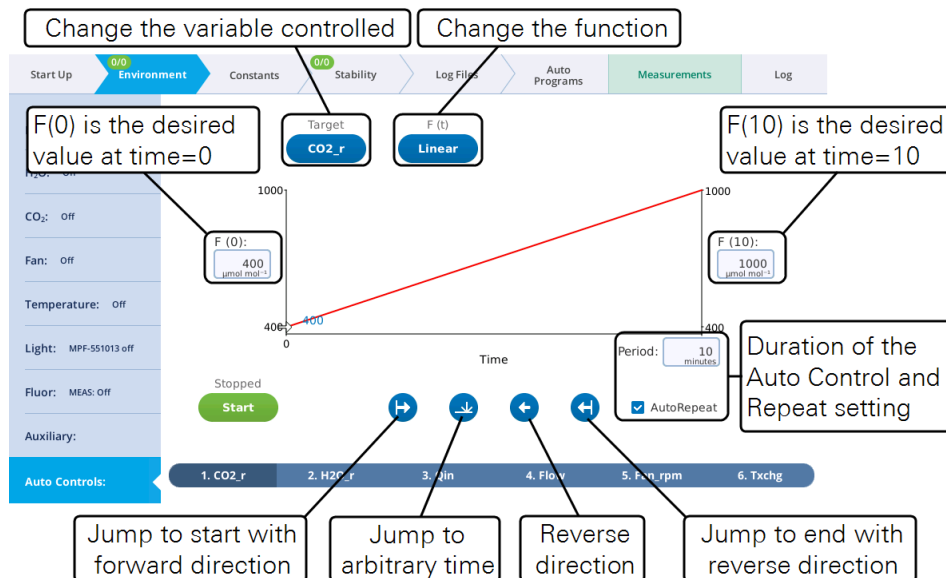


Figure 3-24. Any **Auto Control** can control any variable.

Each control can be configured to be:

- A setpoint at the start time: $F(0)$.
- A setpoint at the stop time: $F(x)$, where x is the time period in minutes.
- The **Period** (minutes) with the option to repeat automatically.
- A shape: $F(t)$, which can be **linear**, **step**, **sine/4**, **sine/2** (half sine), or **tracking**.



As an **Auto Control** is in progress, you can change the timing:



Jump to start with forward direction.



Jump to arbitrary time. Tap to choose a time setpoint.



Reverse direction.



Jump to end with reverse direction.

Auto Controls can be used for tracking. Rather than following a prescribed curve, such as a linear change between two end points, in tracking mode the setpoint comes from some other measurement or computation. The $F(0)$ and $F(x)$ values are meaningless in this mode, but the **Period** still matters, unless **AutoRepeat** is on, in which case tracking will run forever.

The following exercise shows a basic CO_2 **Auto Control**. Begin by setting up a graph so you can see the measurements as the curve develops. Be sure the instrument has a charged CO_2 cartridge and fresh soda lime.

- 1 Set up a graph that shows CO_2_r and CO_2_s .

This might be graph C by default, but if not, select a graph to configure, then:

- Tap **Edit Graphs**
- Tap **+** and add the variables CO_2_r and CO_2_s , if needed
- Set **Y-scaling** to **Fixed**
- Set **Y-axis** to **Y-left**
- Set the range to **Min: 100** and **Max: 600 $\mu\text{mol mol}^{-1}$** .

- 2 Configure the **Flow** controls. Under **Environment > Flow**:

- Turn flow **On**
- Set **Pump** speed to **Auto**
- Set **Flow** to $500 \mu\text{mol s}^{-1}$

- 3 Close the empty leaf chamber.

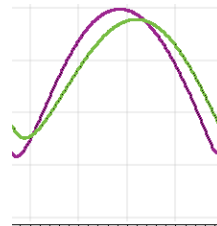
- 4 Configure the **Auto Control**. Under **Environment > Auto Controls > Control: CO₂_r**, set the following:
- **F(0):** 200 $\mu\text{mol mol}^{-1}$
 - **F(x):** 500 $\mu\text{mol mol}^{-1}$
 - **Period:** 1 minute
 - **AutoRepeat:** Check the box
 - **F(t):** Sine/4



- 5 Tap the **Start** button to start the Auto Control.
- 6 Go to the **Measurements** tab and watch the graph develop for a few minutes. After a few minutes go back to the **Auto Control** tab and set **F(t):** linear.

Now set some Auto Controls on fan speed, flow rate, H₂O, light, and temperature to see how they work.

Keep in mind that the instrument is not logging any data. That will come later, in *Open a log file* on page 3-87.



When the instrument is running an Auto Control, the environmental controls will be covered with a box that says **Controlled by Auto** It will not prevent you from making changes, and in fact you can. When you change a setting using the manual controls, the setting will apply until the next auto control takes over. It will be hard to manually override an auto control because the settings are changing every few seconds.

The Constants tab

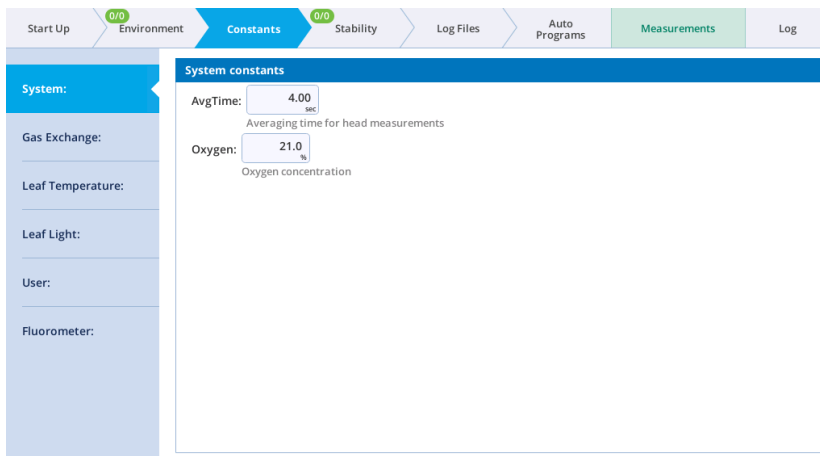
In this section we describe the **Constants** tab. It provides access to settings that describe the system, leaf, environment, and other user-settable constants. In addition, we describe the fluorometer constants, which are available if you have the Multiphase Flash™ Fluorometer connected.

Constants are recorded in the **Header** of the data file (*Table 6-4* on page 6-31) or in columns if the setting was applied in log options.

Note: The **User Constants** can be triggered on a log under **Log Files > Logging Options**, where you select **Prompt on (manual) log**. This will open the prompt when the log button is pressed or when an Auto Log is triggered.

System constants

System constants present two user-settable values: Averaging time for measurements in the head (**AvgTime**) and Oxygen concentration. Typically, you'll use 4 second averaging and 21% oxygen for measurements in ambient air. You should change this value if you are using low oxygen levels.



Gas exchange constants

Here you set the leaf area, stomatal ratio, and geometry. If you are making area-based or mass-based fluxes, this page will be simplified and all you can enter is mass (in grams). If you are making gas exchange measurements, you'll see the following:

- Leaf area (**S**) is the area of the chamber occupied by the leaf. There is an on-board leaf area calculator to assist you. This is a very important parameter to set, because leaf area is used directly when calculating assimilation.
- Stomatal ratio (**K**) is the top-to-bottom ratio of stomata of the leaf. Use 1 for equal stomatal density on top and bottom; 0 for stomata on only one side. If you aren't sure, use 0.5. It doesn't matter if you use the ratio of top to bottom, or bottom to top. Thus, 0.5 is the same as 2; 0.333 is the same as 3, etc. Stomatal ratios for many species are available in published literature.
- **Geometry** and chamber type (**ChType**) are used to determine boundary layer conductance.

The **Broadleaf** setting computes BLC based on leaf area and fan speed, using relationships that have been determined for each chamber. The **Needles** option uses a fixed value, and the **Custom** setting lets you enter whatever you want to use for BLC.

The screenshot displays the 'Gas exchange constants' configuration page. The top navigation bar includes 'Start Up', 'Environment', 'Constants' (active), 'Stability', 'Log Files', 'Auto Programs', 'Measurements', and 'Log'. The left sidebar lists 'System:', 'Gas Exchange:' (selected), 'Leaf Temperature:', 'Leaf Light:', 'User:', and 'Fluorometer:'. The main content area is titled 'Gas exchange constants' and includes the following settings:

- S:** 6.00 cm² (Leaf area)
- K:** 0.5 (Stomatal ratio)
- Geometry:** 0: Broadleaf 1: Needles 2: Custom
- Custom:** 2.00

On the right, the 'Leaf Area Calculator' window is open, showing a diagram of a leaf in a chamber. It displays the following information:

- Leaf Area Calculator
- 6800-01A w/ 6 cm² aperture
- Leaf area: 6.00 cm²
- Leaf Width: 2.76 cm

Leaf temperature constants

The **Leaf temperature constants** tab is where you specify whether the leaf temperature is **Measured** by the thermocouple or computed from the **Energy Balance**. There is also a setting called **deltaTw**, which enables you to set the difference between the chamber wall and chamber air temperatures.

Leaf light constants

The **Leaf Light** tab is where you describe the absorptance of the leaf. These values typically come from published literature or direct measurements. Leaf light constants typically will not need adjustment.

User constants

User constants are user-settable fields. You might use a **User Constant** to record in the file which replicate you are on, which species, who is collecting the data, or other information that you want to collect. User-defined items include user-defined equations (variables) and constants.

When you first open the **Constants > User** tab, it will be empty. *Figure 3-25* below shows the constants window after constants and variables have been added.

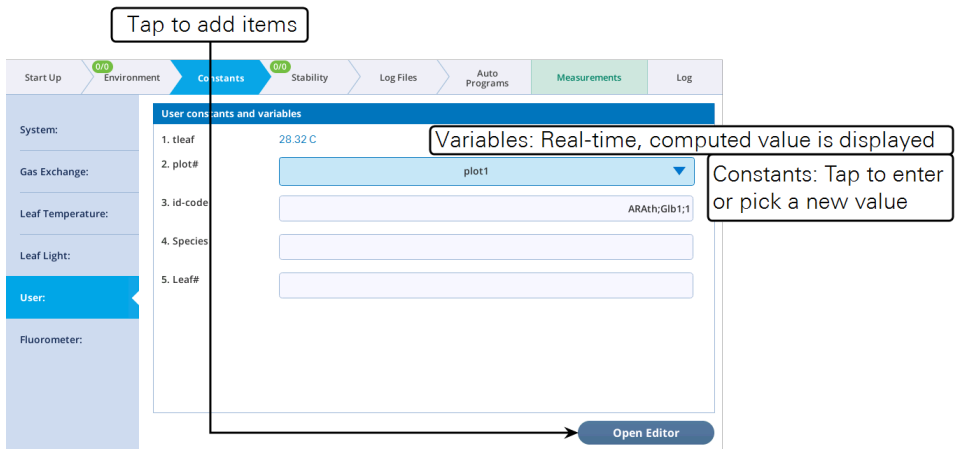
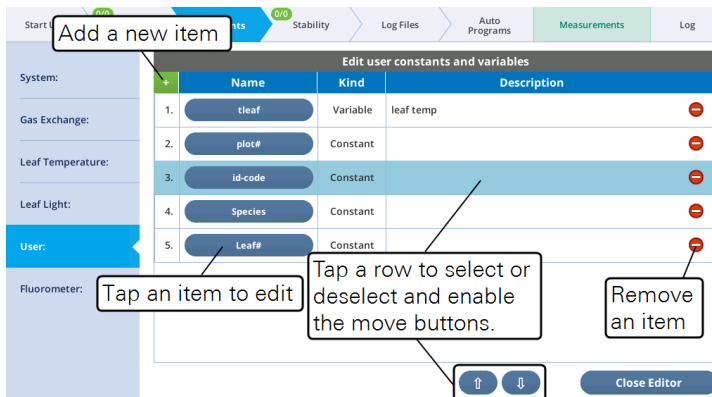


Figure 3-25. User Constants are used to create a customized field for information that will be recorded in the data file.

Editing user constants

Tap the **Edit** button, and then tap the plus sign (+) to add a new constant.



There are two basic types of constants: Those that are entered via the numeric keypad or keyboard, and those that are found in a menu list. For both numeric and list constants, they can be logged as a column with every record, or logged as a row once in the file header, and when its value changes.

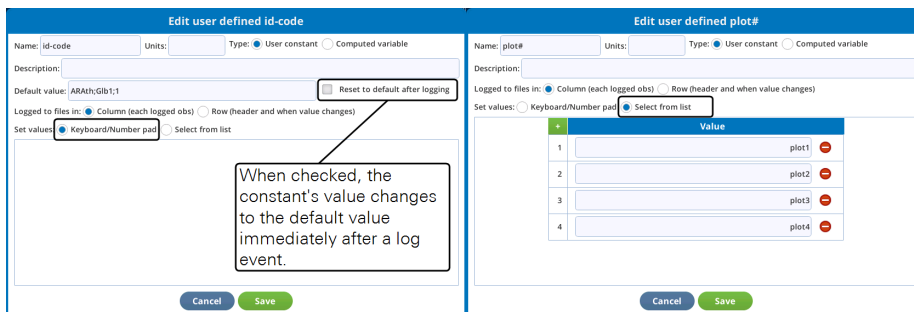


Figure 3-26. Editing a user-defined constant. You can specify a text-entry field (left) or a list (right).

Editing variables

User-defined variables are computed with a simple algebraic expression based on other variables or constants. The rules for entering the expression are:

- For variable names, use v1, v2, etc.
- For constants, either type them in explicitly, or refer to them as a1, a2, etc.
- Use parentheses for grouping. For example, $a1 * (a2 + a3 * v1) * v1$.
- Use +, -, *, and / for add, subtract, multiply, and divide.

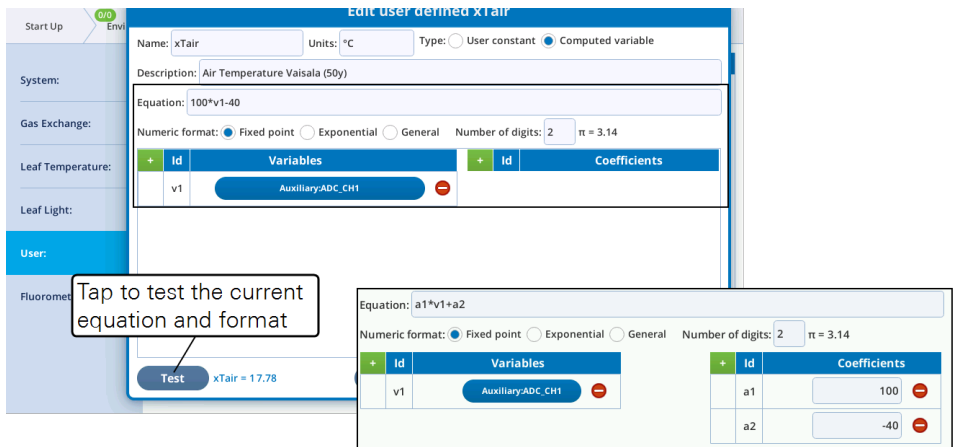


Figure 3-27. Editing a user-defined variable. The **Equation** can have numeric constants directly in the equation, or have named coefficients (inset).

The **Test** button will use the current formula and format and the latest values of variables to compute a result. If there is an error (for example, referencing an undefined variable or coefficient), an error message is displayed instead. User-defined variables will show up in the Excel file with their equation.

You can choose how to log user constants—as a row or column—and enable prompting on logging. See *Log options* on page 3-83 for details.

Fluorometer constants

Fluorescence Constants (Fo, Fm, Adark, and PS2/1) can be loaded from a DarkAdapted file. A DarkAdapted file is created when a flash is triggered by an FoFm log event. DarkAdapted files are stored in the flrevents folder, and begin with "DarkAdapted".

Each DarkAdapted file contains Fo, Fm, Adark, and PS2/1, and the name of the flash event. Loading a DarkAdapted file from the **Constants > Fluorometer** page (Figure 3-28 on the facing page) does two things:

- 1 Sets the constants' values
- 2 Sets the Dark Adapted data on the Results screen.

The figure consists of two screenshots from a software interface. The top screenshot shows the 'Constants' tab with a 'Load' button and a file selection dialog. The dialog shows a list of files, with 'DarkAdapted_RECT-248-20190723-12_19_01.txt' selected. The bottom screenshot shows the 'Results' tab with a graph of 'Dark Adapted' fluorescence and a table of constants. The graph shows a sharp peak at 745.8 nm. The table below the graph lists various constants and their values.

Label	MPF	RECT	IND.	DARK	Notes
DATE	✓	✓	✓	✓	Date of event (see also TIME)
DCmax			✓		Max value of DC/Q fluorescence

Figure 3-28. Set constants and dark adapted results by loading from a previously logged FoFm event.

Fluorometer event file documentation

Table 3-4 below lists the single-value labels in the four types of fluorometer event files (MPF, RECT, INDUCTION, and DARK), and Table 3-5 on page 3-82 the multi-value entries. Items marked (new) are only present in Version 1 files.

Table 3-4. Single-value entries in fluorescence event files, Version 1 and prior.

Label	MPF	RECT	IND.	DARK	Notes
DATE	✓	✓	✓	✓	Date of event (see also TIME)
DCmax			✓		Max value of DC/Q fluorescence

Table 3-4. Single-value entries in fluorescence event files, Version 1 and prior.
(...continued)

Label	MPF	RECT	IND.	DARK	Notes
DCo			✓		Extrapolated pre-flash for DC/Q
DEVICE	✓	✓	✓	✓	Fluorometer serial number
DURATION		✓	✓	✓	Event duration (ms flash, s dark)
D FARRED PERCENT				✓	Farred power as % of max
D RED PERCENT	✓	✓	✓		Red power as % of max
EVENT ID	✓	✓	✓	✓	Event identifier number
F1			✓		F at 1st inflection
F2			✓		F at 2nd inflection
FMAX	✓	✓	✓		Maximal F
FMIN				✓	Minimal F
Fo (present if dark adapted)	✓	✓	✓		Pre-flash F
Fs (present if light adapted)	✓	✓	✓	✓	Steady state fluorescence
InitSlope			✓		Initial slope of DC/Q
MARGIN	✓	✓	✓	✓	Margin (# of points)
MODRATE	✓	✓	✓	✓	Modulation rate (Hz)
OUTRATE	✓	✓		✓	Data points per second
P1 DELTAF (new)	✓				P1 PREDF - P1 MAXF
P1 MAXF	✓				Phase 1 max F
P1 PREDF (new)	✓				Phase 1 predicted F
P2 INT	✓				Phase 2 regression intercept
P2 INT SE	✓				Phase 2 std. error of intercept
P2 R2	✓				Phase 2 regression R squared
P2 SLP	✓				Phase 2 regression slope
P2 SLP SE	✓				Phase 2 std. error of slope
P3 DELTAF	✓				P3 PREDF - P3 MAXF
P3 MAXF	✓				Phase 3 max F
P3 PREDF	✓				Phase 3 predicted F
P3 Q	✓				Max Q during phase 3
PHASE1 DURATION	✓				Phase 1 duration (ms)
PHASE2 DURATION	✓				Phase 2 duration (ms)
PHASE3 DURATION	✓				Phase 3 duration (ms)
Q@P1 MAXF (new)	✓				Q at phase 1 max F

Table 3-4. Single-value entries in fluorescence event files, Version 1 and prior.
(...continued)

Label	MPF	RECT	IND.	DARK	Notes
Q@P3 MAXF (new)	✓				Q at phase 3 max F
QMAX	✓	✓	✓		Maximum Q during flash
QMIN				✓	Minimum Q during dark pulse
Q FARRED SETPOINT				✓	Farred target $\mu\text{mol m}^{-2} \text{s}^{-1}$
Q RED SETPOINT	✓	✓	✓		Red target $\mu\text{mol m}^{-2} \text{s}^{-1}$
RAMP	✓				Percent ramp for phase 2
T@DCmax (new)			✓		Time (s) of DC/Q max value
T@F1			✓		Time (s) of F1
T@F2			✓		Time (s) of F2
T@FMAX (new)		✓	✓		Time (s) of FMAX
T@FMIN (new)				✓	Time (s) of FMIN
T@NIR			✓		Time (s) of half initial rise $(F_1 - F_s)/2$
T@P1 MAXF (new)	✓				Time (s) of phase 1 max F
T@P3 MAXF (new)	✓				Time (s) of phase 3 max F
TIME	✓	✓	✓	✓	Time (local) of the event
TIMESTAMP	✓	✓	✓	✓	GMT time t value of event (s since 1970)
TYPE	✓	✓	✓	✓	MPF, RECT, INDUCTION, or DARK
T OFFSET	✓	✓	✓	✓	Time shift (s) of event start (In version 1, this is already incorporated into the SECS data.)
VERSION (new)	✓	✓	✓	✓	File version
Q FARRED SETPOINT				✓	Farred target $\mu\text{mol m}^{-2} \text{s}^{-1}$
Q RED SETPOINT	✓	✓	✓		Red target $\mu\text{mol m}^{-2} \text{s}^{-1}$
RAMP	✓				Percent ramp for phase 2
T@DCmax (new)			✓		Time (s) of DC/Q max value
T@F1			✓		Time (s) of F1
T@F2			✓		Time (s) of F2
T@FMAX (new)		✓	✓		Time (s) of FMAX
T@FMIN (new)				✓	Time (s) of FMIN
T@NIR			✓		Time (s) of half initial rise $(F_1 - F_s)/2$
T@P1 MAXF (new)	✓				Time (s) of phase 1 max F
T@P3 MAXF (new)	✓				Time (s) of phase 3 max F
TIME	✓	✓	✓	✓	Time (local) of the event
TIMESTAMP	✓	✓	✓	✓	GMT time t value of event (s since 1970)

Table 3-4. Single-value entries in fluorescence event files, Version 1 and prior. (...continued)

Label	MPF	RECT	IND.	DARK	Notes
TYPE	✓	✓	✓	✓	MPF, RECT, INDUCTION, or DARK
T OFFSET	✓	✓	✓	✓	Time shift (s) of event start (In version 1, this is already incorporated into the SECS data.)
VERSION (new)	✓	✓	✓	✓	File version

Table 3-5 below lists the time-series data, which is common to all event files. A difference in version 1 files is that the time (SECS) is adjusted by the T_OFFSET value, so that SECS=0 corresponds to the time the flash began (or when the actinic turned off for a dark pulse). In files with no VERSION, the rst value of SECS is 0.

Table 3-5. Table of multi-value entries in fluorescence event files.

Label	Notes
SECS	Time (s). In version 1 files, the 0 value is the time of the flash on or actinic off.
FLUOR	Modulated fluorescence.
DC	Non-modulated fluorescence.
PFD	Photon Flux Density (includes contribution of REDMODAVG).
REDMODAVG	Average flux contribution from the red modulated LEDs.
CODE	Event phase data identifier:
	2 = pre-flash margin
	3 = rect or induction flash
	4 = MPF phase 1
	5 = MPF phase 2
	6 = MPF phase 3
	7 = post-flash margin
	11 = pre-dark margin
	12 = farred on (actinic still on)
	13 = actinic off (farred still on)
14 = farred off	
	15 = post-dark margin (actinic back on)

The Stability tab

The operating system allows you to set up criteria based on measured and computed variables to determine when a variable is stable. Stability criteria are set under the **Stability** tab. For each variable chosen, stability is based on statistics (a choice of standard deviation, slope, or both) over a time period of your choice.

The screenshot shows the 'Stability' configuration window. It contains three rows of measurement variables:

- F.FlRLS**: Current Value: 7.0, Mean: 3.373, Slope: -3.816 min⁻¹, Stdev: 6.114. Status: Unstable. Settings: Slope limit: 1, Stdev limit: 1, Period: 15 sec.
- ΔH2O.Meas2**: Current Value: -0.007 mmol mol⁻¹, Mean: -0.006062 mmol mol⁻¹, Slope: -0.00548 mmol mol⁻¹ min⁻¹, Stdev: 0.0007721 mmol mol⁻¹. Status: Stable. Settings: Slope limit: 0.1, Stdev limit: 0.1, Period: 20 sec.
- ΔCO2.Meas2**: Current Value: 4.32 μmol mol⁻¹, Mean: 4.385 μmol mol⁻¹, Slope: -0.35 μmol mol⁻¹ min⁻¹, Stdev: 0.04496 μmol mol⁻¹. Status: Inactive. Settings: Slope limit: 0.1, Stdev limit: 0.1, Period: 20 sec.

You can add variables (as many or as few as you want) that you will use to define stability criteria. These can be used for casual observation while logging manually, or to specify criteria for triggering an event in Auto Programs.

The Log Files tab

The Log Files tab is where you create new data files, set up criteria related to logging, and configure the log options.

Logging options

In its default configuration, the LI-6800 provides a continuous stream of measurements and calculations. You can monitor light levels and photosynthesis rates, for example, while changing those light levels. Over the course of 5 minutes, the instrument will have made hundreds of measurements. The question becomes, which of these will you want to retain for future use?

Log options

- **Also Log Data to Excel file:** Select this to log all your data values (those you selected, at least) and the equations that are used for computed values into a Microsoft® Excel® file. When you append an existing file (reopen and log more data to it), the instrument will create a new .xlsx file for the appended data.

- **Prompt on (manual) log:** Select this to open a prompt when logging. Prompts are user-settable questions and system constants that are configured under the Constants tab (see *The Constants tab* on page 3-74). For example, you may wish to open the prompt so you can enter the leaf area for each sample, or to record the species in your data file.
- **Use Additional Averaging Time:** The logged data reflects a potentially longer average time than the 4 second default that is used without extra averaging.
- **Beep when logging:** Turns the beep on or off.
- **Automatically name new log files?** Log files will be named with date and time stamp as the file name. Clear the check box to create new log files without the automatic name.

When opening an existing file, always...

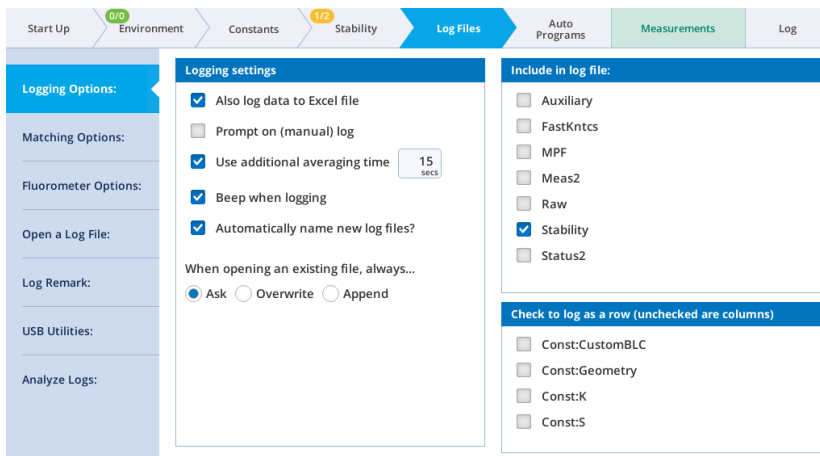
Ask will give you the choice to overwrite or append the file. **Overwrite** will automatically delete all the data in that file and write new data to the file. **Append** will add the new data after any other data in the file.

Include in log file

You have choices about the groups data to include. These include MPF, Match, Meas2, Raw, Stability, and Status2. See *Summary of symbols* on page 6-31 for information about these groups.

Check to log as a row

Variables that are rarely changed do not need to be logged as columns, and can be logged as a row in the top of a file.



Matching options

The Match Options available when logging have changed, allowing independent control of CO₂ and H₂O matching (*Figure 3-29* below). These settings only pertain to whether or not a match is triggered and performed as part of a log event; they do not apply to matching initiated by tapping **Auto** or **Manual** in the match screen.

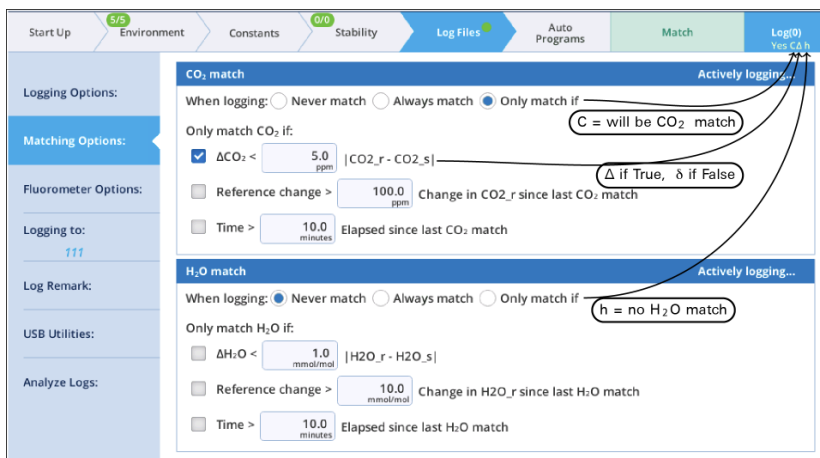


Figure 3-29. The new Matching Options screen.

When a log file is open, the bottom line of the **Log** button indicates if a match will happen at the next log (**Yes** or **No**). **C** or **c** indicates if CO₂ will be matched (**C** is

yes, **c** is no), and **H** (yes) or **h** (no) indicates if an H₂O match will occur. If conditional matching (**Match only if**) is enabled, there will be up to three code letters following the **C/c** and **H/h**, one for each item checked. **Δ** for ΔCO₂ or ΔH₂O if true (**δ** if false), **R** for reference if true (**r** if false), and **T** for time if true (**t** if false).

Examples:

No c h - No match is happening, because both CO₂ and H₂O are set to **Never match**.

Yes C h - A CO₂ match will occur, because it is set to **Always match**.

Yes CΔt h - CO₂ is conditional on ΔCO₂ (true) and time (false). H₂O is conditional on ΔH₂O (false).

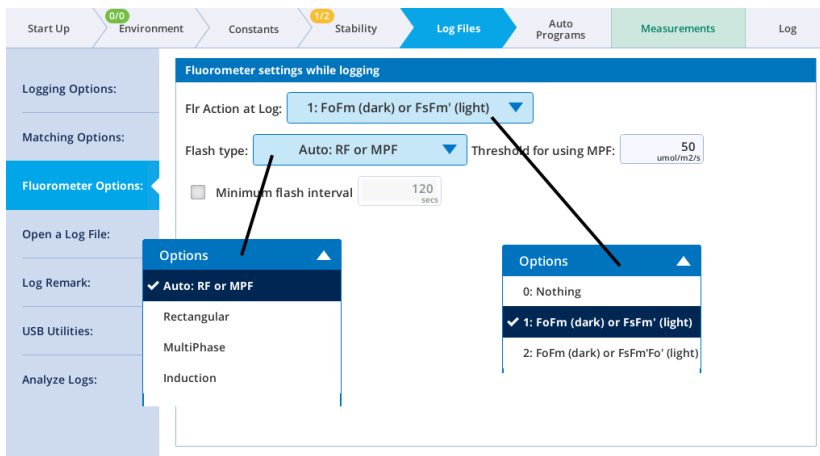
Yes cδrt HδRt - Both CO₂ and H₂O are conditional based on all three options. There will be an H₂O match, triggered by the reference change (**R**).

When a match is triggered because of only one gas (CO₂ or H₂O), only that gas will be matched.

The protocol for matching is described in *Matching the analyzers* on page 4-3.

Fluorometer options

With the Multiphase Flash Fluorometer, two fluorescence options are available: **Flr Action at Log**, where you specify what the fluorometer should do when you press the log button, and the **Flash Type**. From the **Flr Action at Log**: menu, you can select from the following: **0: Nothing**, **1: FoFm (dark) or FsFm' (light)**, and **2: FoFm (dark) or FsFm'Fo' (light)**. The second menu, **Flash Type**, presents four options: **Auto: RF or MPF, Rectangular, MultiPhase, or Induction**.

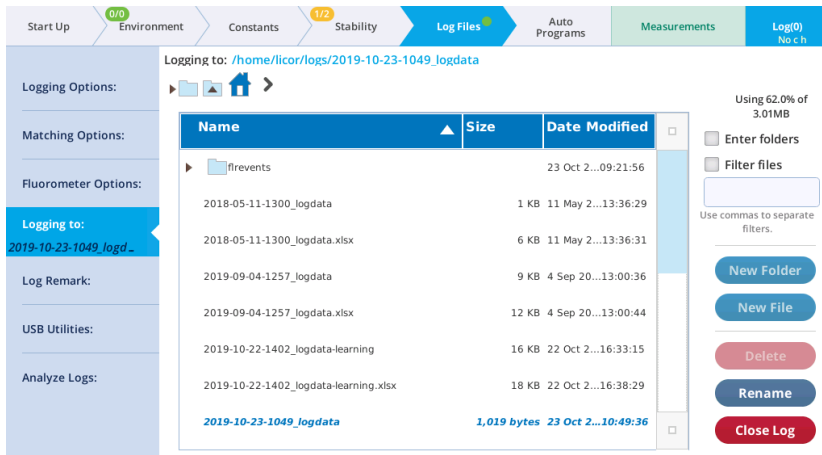


The **Auto: RF or MPF** option lets the system decide between a rectangular flash (RF) and a multiphase (MPF) for the logged flash event. The decision is based on the fluorometer's Q value (Q in group FlrLS) at the time of the log event.

The option for **Minimum flash interval** allows frequent logging with occasional fluorescence events. The LI-6800 keeps track of the time since previous flash (TSPF), and if a log event happens with a TSPF less than the specified value, the Flr Action for that log event will in effect be **0: Nothing** but just for that event. Thus, you can log every 10 seconds, for example, but only have fluorescence events every 2 minutes. This applies to manual and automatic logging. Also, the "previous flash" on which the timing is based is just that. It doesn't matter if that flash was triggered manually or automatically by a log event.

Open a log file

A log file is required to store measurements. You can either create a new file or append an existing file. You can also create a folder structure for your files and assign each log file a name. To create new file, tap **Log Files > Open a Log File**. Tap **New File**. Or, edit an existing file by selecting a file and then tapping **Open**.



Once a log file is opened, the **Log** button at the top right of the display will become a **Log (0)** button.

Observations can be logged by pressing the **Log (0)** button on the screen or the log button on the sensor head. A short tap (<¼ second) is all that is required to log a measurement to the file. A successful log is indicated by the **Log (#)** incrementing with each new log (and a beep, if **Beep when logging** is enabled).

You will not be allowed to open a new log file if the file system is greater than 99.9% full (<4.67 MB available). The instrument will notify you of the issue.

When the measurement is underway, the instrument will show ****Log pending**** in the upper right of the display.

Log remark

This option is active when a log file is open. Remarks are free form text rows saved in the data file to help you understand a particular measurement. For example, "Wow, this plant looks terrible," or "Apparently, deer do eat sotol." For plant # or treatment ID, consider adding a **User Constant** (see *User constants* on page 3-76). When you enter a remark, it will be saved in the log file with the time stamp.

Where are the remarks logged?

- **In text data files:** Remarks are recorded in the text data file, either in the header information or inline with the other measurements. Remarks will always

include the time stamp.

- **In Excel files:** Remarks are in a separate tab from the measurements, one called Remarks. Here again, remarks will always include the time stamp.

USB utilities (Log Files)

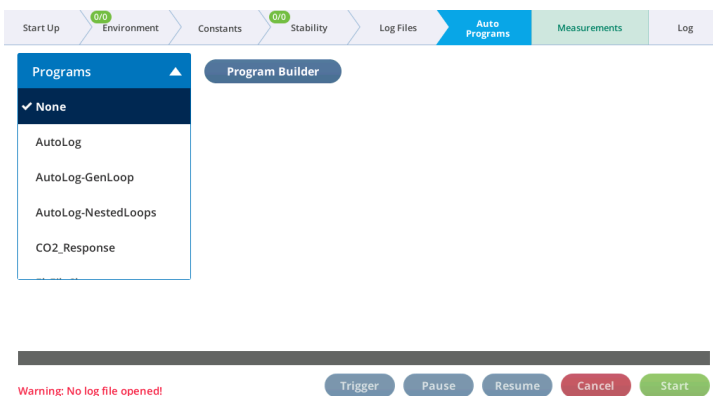
Here you can manage files on an attached USB device. This is described in detail in *USB Utilities (Start Up)* on page 3-9.

Analyze logs

Here are tools that you can use to evaluate logged data on the console. This is explained in greater detail in *Managing logged data on the console* on page 6-3.

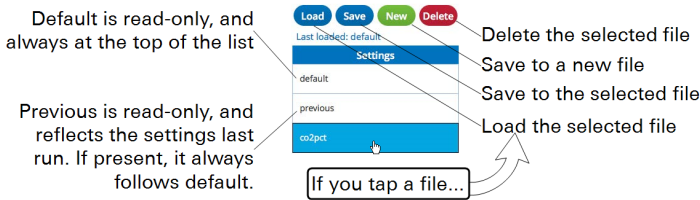
The Auto Programs tab

The LI-6800 can operate autonomously with either an **Auto Program** or a **Background Program (BP)**. Auto Programs log data in some sort of automated fashion, while perhaps maintaining control over one or more conditions in the leaf chamber. A log file doesn't have to be open to run an Auto Program, but no data are logged without one. Background Programs can do the same things as Auto Programs, but they have three significant advantages: 1) you can make your own auto programs, 2) the scope of what can be done with a BP exceeds that of an Auto Program, and 3) any number of BPs can run simultaneously. Auto Programs are loaded under the **Programs** list. Background Programs are started by tapping the **Program Builder** button.



Working with Auto Programs

You can **Load** and **Save** settings from named files, or load settings from the previous time the Auto Program was run (called **Previous**). **Default**, which loads the factory default settings, is always at the top of the list. Previous (if present) is always below default. You also can select any of the named files.



List of auto programs

The LI-6800 includes a number of basic Auto Programs. These are described below.

Program	Description
AutoLog	Automated logging for a fixed time at regular intervals. You can set the interval (Log interval) and the duration (Total time).
AutoLog-GenLoop	Automated logging within a control loop.
AutoLog-NestedLoops	Automated logging within a nested loop.
CO2_Response	Loop over CO ₂ values.
Generic_Response	Loop over any collection of controls
Light_Response	Loop over a range of light values.
LogOnce	Waits for stability and then logs once. The Min. wait and Max. wait settings specify the minimum and maximum wait times, respectively. Early match tells the instrument to initiate the match when the reference gas has stabilized, rather than waiting until both sample and reference have stabilized, which can save time.
Nested_Responses	Two nested control loops
Repeated_Responses	Generic response, repeated N times.
Survey Setup	An easy way to set up survey measurements.

We'll go over response curves later in the tutorials (see *Step-by-step carbon dioxide response curve* on page 4-26 and *Step-by-step light response curve* on page 4-30).

Under any response measurement, you can configure settings to **Wait for Stability** and early matching.

The **Allow early match** option will let matching happen sooner if the reference cells are stable before logging has to happen, and if matching is called for. The window of opportunity for an early match is at least 30 seconds into the minimum wait time, and no later than 60 seconds before the end of the maximum wait time. Thus, if the minimum wait is 60 seconds and the maximum is 120 seconds, then early matching would only have a 30 second window of opportunity—from 30 to 60 seconds into the wait.

WaitForStability: ▼ Wait 60 to 120 secs, early match allowed

Min. wait: sec

Max. wait: sec

Allow early matching

About MultiControl Loops

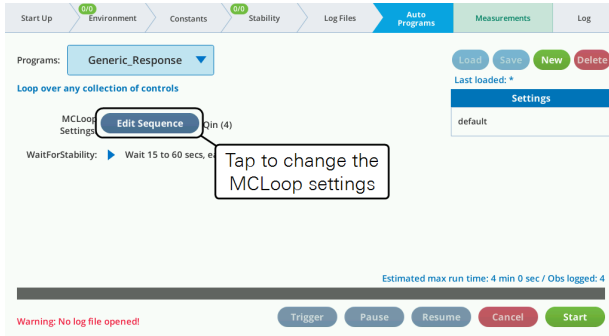
MultiControl loops (**MCLoops**) allow you to control any number of controls (see *Table B-1* on page B-1) and constants and settings (see *Table B-2* on page B-2 in the following Auto Programs:

- AutoLog-GenLoop
- AutoLog-NestedLoops
- Generic_Response
- Nested_Responses
- Repeated_Responses

A simple example is a control loop that sets light intensity and light color all in the same loop. You could just as easily add fluorescence flash parameters to the mix, if you wanted those changed as well.

To illustrate, we start with the **Generic_Response** auto program and configure it to do a four-point light response twice, changing colors midway through. To configure the control loop:

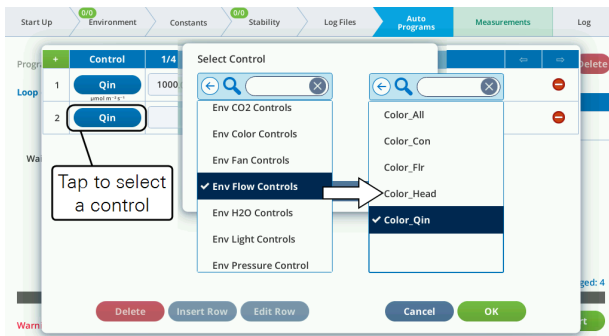
1 Tap Edit Sequence to change the MCLoop settings



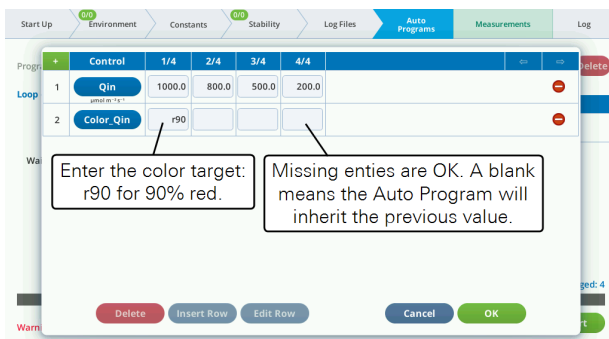
2 Tap the plus (+) to add a new row.



3 Tap an item in the Control column to select a control.

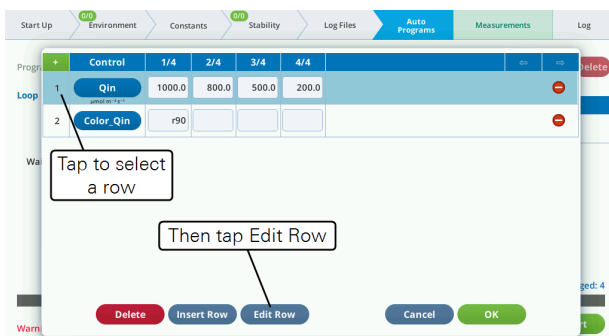


4 Enter the target. In this example the target for the first step is r90 for 90% red.



Now you have a 4-step light curve that will use a color that is 90% red. You can expand this to repeat the curve with a different color. See *Color spec* on page A-1 for details on color.

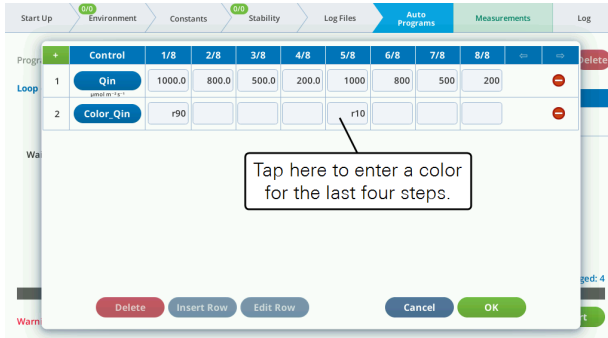
5 Tap a row to select, then tap **Edit Row**.



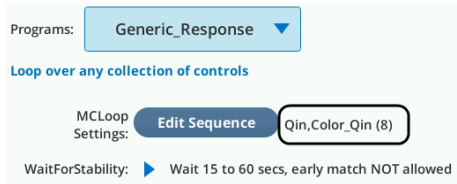
6 Enter new values separated by commas.



7 Enter a new color in step 5/8.



8 When you tap OK, the Auto Programs screen will summarize the settings.

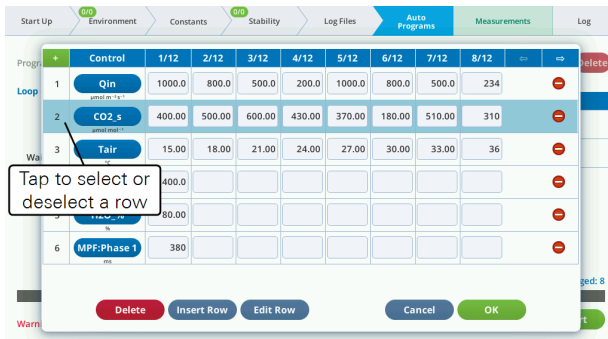


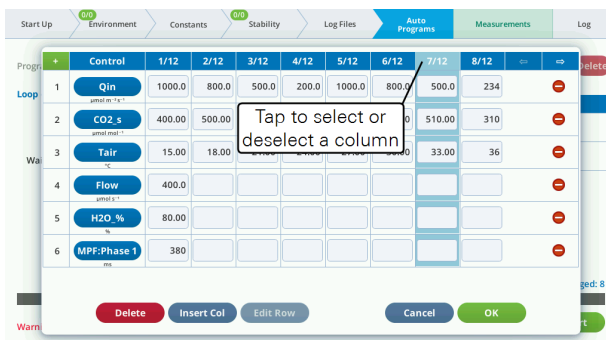
Tips for using MultiControl loops

There are two methods to increase the number of columns in the table:

- Select a row and tap **Edit Row**, and enter the desired number of entries, or
- Select a column and tap **Insert Col**. Insertion goes to the left of the selection.

To decrease the number of columns, select a column and tap the **Delete Col** button.





Launching auto programs

All Auto Programs are launched in the same way:

- 1 Open a Log File.
- 2 Select the Auto Program.
- 3 Tap Start.

When the instrument is done it will stop automatically. You can view the progress under the Auto Program tab and observe the instrument controls under the Environment tab. When the instrument is running an Auto Program, the environment controls will be covered with a box that says **Controlled by AutoProg**. It will not prevent you from making changes, and in fact you can. With something like an Auto Program, which sets the control and doesn't change it again until the next step, you can successfully override the setting until that next step.

Background Program overview

Background Programs (BPs) are a new approach to Auto Programs. While they do not replace the Auto Programs, they do provide an alternative with several advantages. You can, for example, easily modify factory-supplied programs, or generate your own from scratch. Also, multiple BPs can be run simultaneously.

A quick overview of BPs is below. The complete guide to writing and using BPs is found at licor.com/documents/be4i41pnc010o1y5084na8ma2i6lzp5

Accessing

The **Program Builder** button on the Auto Programs page (*Figure 3-30* below) opens the BP's **Open/New** screen, from where one can run or edit existing BPs or create a new BP from scratch.

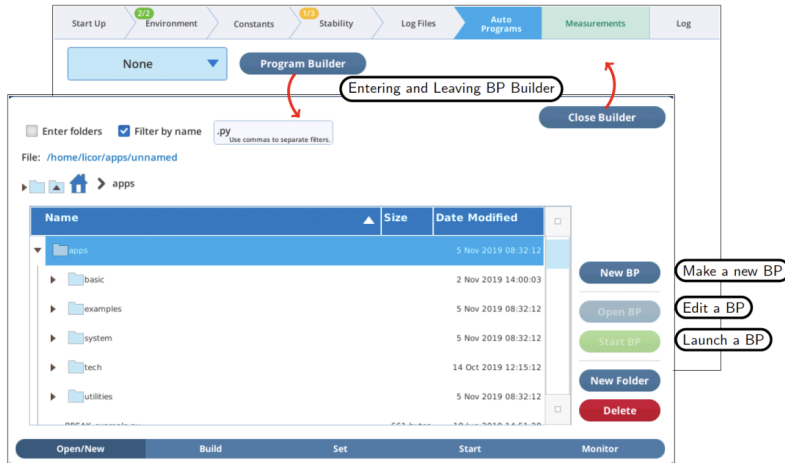


Figure 3-30. BPs buttons and the BP Builder screen.

BPs can be found in the `/home/licor/apps` folder, and are distributed across several subfolders. In the **basic** folder are several simple BP's that duplicate the functionality of some of the basic Auto Programs. The other BP folders include **examples** (example programs used in the BP user guide), **system** (helper programs used by the system), **utilities** (useful tasks, if you need them), and **tech** (specialty tasks that a technician might use or ask you to run).

Several of the new features in version 1.4 are implemented with the help of BPs, such as *Acquiring range match data* on page 4-11, *Balanced Flow* on page 3-21, and *Light source test program* on page 3-48.

Running

BPs can be run by selecting a BP file and tapping **Start BP**. After that, what happens is entirely up to the BP itself. The files in the basics folder will display a dialog (*Figure 3-31* on the facing page) letting the user configure and run, or cancel the program.

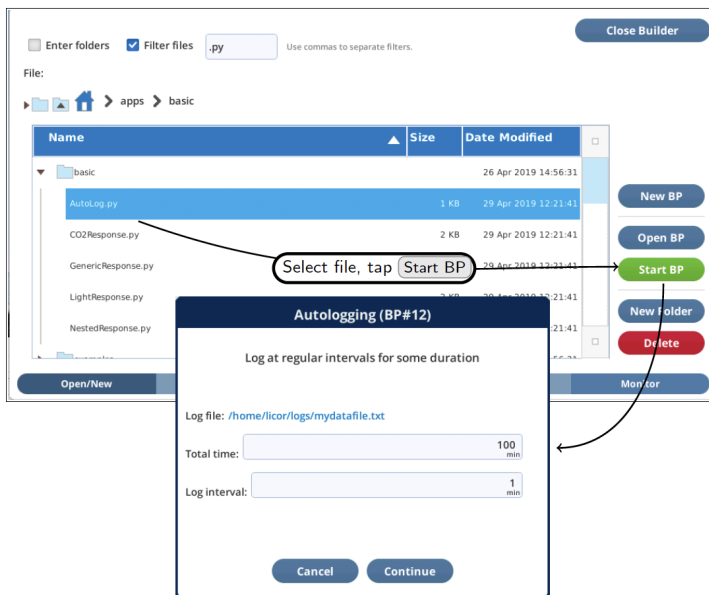


Figure 3-31. BPs in the basics folder have opening dialogs for setting parameters.

Once a BP is running, it can be monitored (and paused, canceled, etc.) from the BP Monitor screen (Figure 3-32 below).

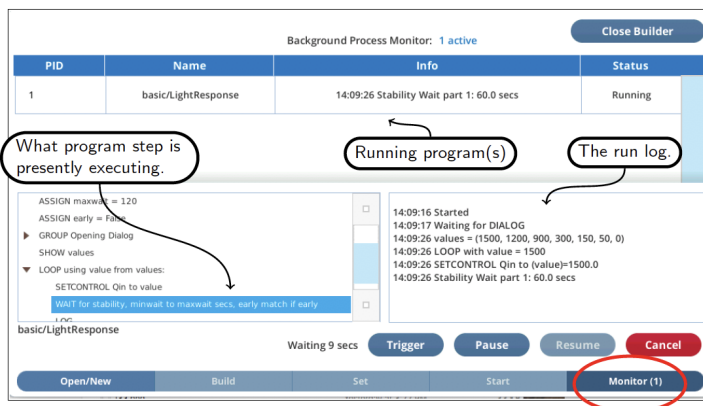


Figure 3-32. Once a BP has been started, you can monitor it in the Monitor screen.

Building

The Builder environment lets you make new BPs from scratch, or modify existing BPs. The steps in *Figure 3-33* below make a trivial BP that waits for 10 seconds then quits, but illustrates the **Build**, **Set**, **Monitor** screens of the Builder environment.

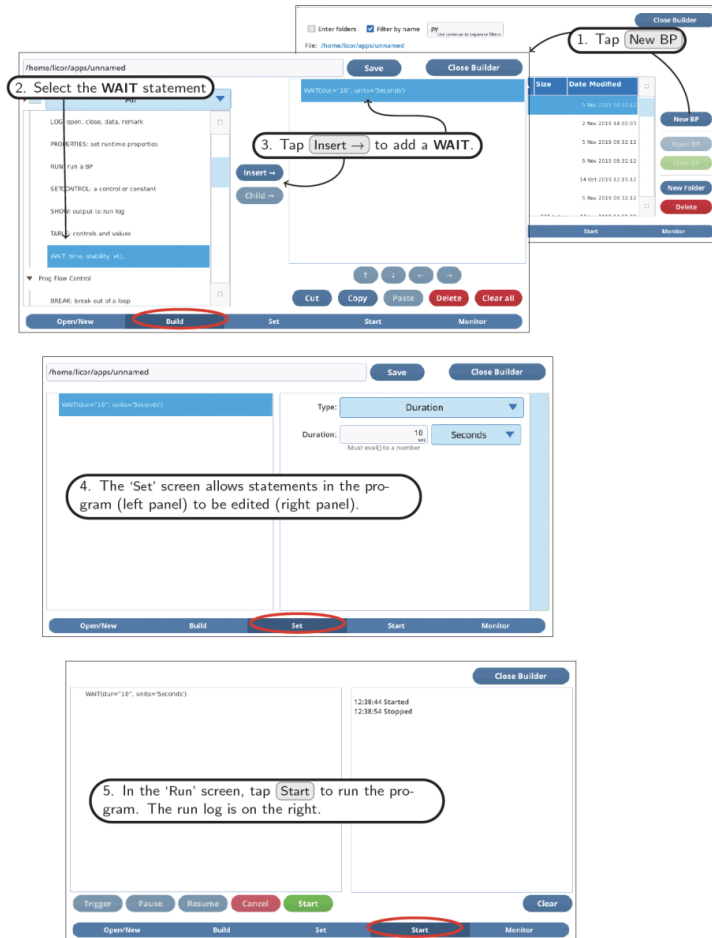


Figure 3-33. **Build** screen: Add and arrange BP steps. **Set** screen: Edit BP steps. **Start** screen: Run the program.

The Measurements tab

Tap **Measurements** to view graphs and values. This is where you view parameters that are measured and computed by the instrument; and where you initiate a match.

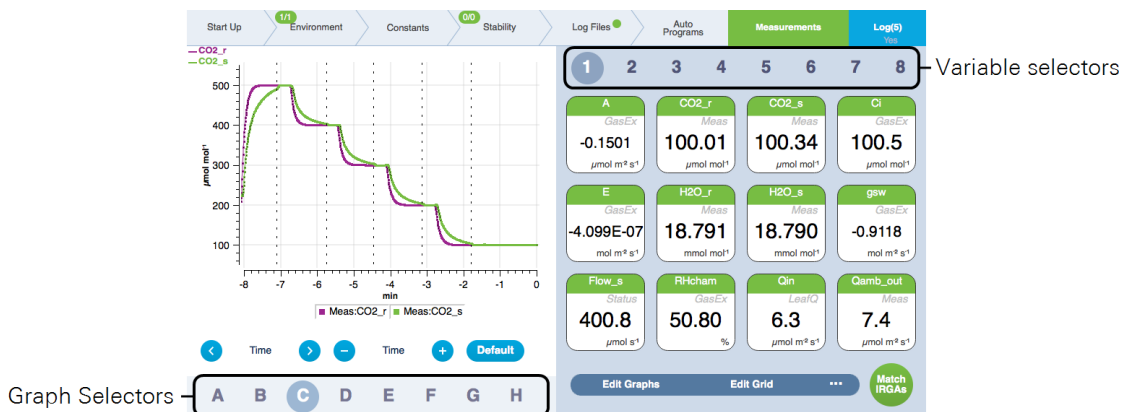


Figure 3-34. The **Measurements** tab, displaying CO_2_r and CO_2_s during a CO_2 response **Auto Program**. Tap a letter to select a different graph and a number to select a different group of variables.

Graphs

Eight **Graph** displays are available and they are designated letters **A** through **H**. Each plot can be a strip chart (a variable plotted against time) or a scatter plot (one variable plotted against another using discrete points usually representing logged observations). The graphs can display multiple variables simultaneously. You can adjust the time by scrolling left for back in time or right for forward in time¹ and zooming in and out. For X-Y plots, you can zoom on each axis.

You can plot multiple variables on one graph and change the graphical display settings in other ways. To configure a graph:

1 Tap **Edit Graphs**

This opens the graph settings window, where you can do the following:

2 Add a variable

¹Not into the future. Not yet, anyway.

Tap the + to add a variable to the graph. Tap a variable to select a different one from a list. The variables menu shows groups based on the source of the variable. You can choose whether to display the units on the left or right axis.

3 Change the graph settings

You can set the color, line type, and marker type.

4 Change the scaling

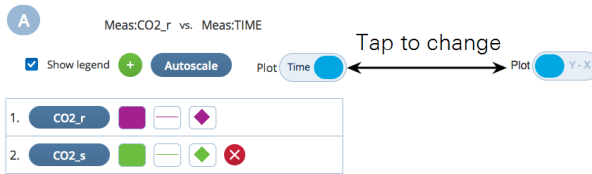
The scaling is **Autoscale** by default, but you can specify a range by setting **Fixed**, **Fixed Min**, **Fixed Max**, or **Fixed Delta**.

5 Turn off the legend

This will enlarge the graph slightly.

6 Plot two variables against each other.

Set **Plot: Time** to show two variables against each other.



Variables

Eight **Variable** pages are available, and each page displays up to twelve variables. You can customize and filter the list of displayed variables, or re-size the variables list. Tap **Edit Grid** to alter the display.

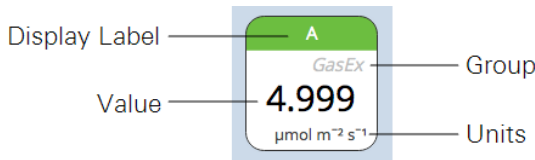


Figure 3-35. The **Display Label** is the variable. The **Group** indicates where this variable is stored (see Summary of symbols on page 6-31). The **Value** is the current measured or computed parameter. **Units** comply with the International System of Units (SI) standards.

Measurements options

You can customize the measurements page. To view the options, tap the ellipsis button (...) to reveal the additional settings.

- **Edit Graphs:** This is where you can alter the settings of whichever graph is displayed. You can plot two variables against each other (tap Plot: X-Y) or over time (tap Plot: Time), adjust scaling, add variables, and alter the graph markers.
- **Edit Grid:** This is where you select the variables that are displayed. To change the variable displayed, tap it on the left, then select another variable on the right. You can change the number of variables displayed, which also changes the size of variables on the display.
- **Clear Graph:** This clears all plotted data from the current graph. It will also reset the autoscaling if autoscale is on. It does not delete any data from your files, though.
- **Clear All Graphs:** Clears all graphs.
- **Mark Graph:** Places a marker on the current graph. The marker is not retained in the log file.
- **Mark All Graphs:** Places a marker on all graphs. The marker is not retained in the log file.



Section 4.

Learning the LI-6800

Fundamentals of good measurements

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Learning the LI-6800

In this section we introduce some basic concepts that will help you be successful with the LI-6800. Typically, you'll proceed through the tabs from left to right, although you may be able to skip some tabs, depending on your objectives.



Start up checklist

When you turn on the instrument for the first time each day, consider the following things.

- 1 Air supply: New CO₂ cartridge and fresh chemicals?
 - Install a new CO₂ cartridge.
 - **H₂O Scrub:** If the desiccant is saturated, replace it with fresh desiccant. Drierite turns from blue to pink when saturated. Sorbead[®] Orange CHAMELEON[®] turns from orange to colorless when saturated. If the desiccant is partially saturated, remove the column and shake it to break up the pellets.
 - **H₂O Add:** If there are no droplets of water condensate on the interior walls of the humidifier column, add 10 to 15 ml of distilled water total. If the Stuttgarter Masse is new, add about 30 ml of water total.
 - **CO₂ Scrub:** If the soda lime has turned from white to violet, replace it with fresh soda lime. If it is partially violet, take off the column and add 10 ml of water. Shake the column so the soda lime absorbs the water.
- 2 Select a configuration.

If you have a configuration file, select **Load Configurations** from the menu, then choose the configuration. If not, just proceed to configure the instrument.
- 3 Run the **Warmup Tests**.

Close the chamber. Under **Start Up > Warmup/System Tests**, select **Warmup Tests** and tap **Start**. The instrument will proceed through a series of tests, taking about 10 to 15 minutes.
- 4 If you have a fluorometer installed, run the **Fluorometer Tests**.
- 5 Review the summary for any failed tests and take action if necessary.

6 Match the IRGAs.

Matching the IRGAs is easily accomplished whether the chamber is empty or not, but it's a good idea to do this once right before starting a measurement. Under **Measurements**, tap **Match IRGAs** on the lower right of the display. Tap **Manual Match**. See *Matching the analyzers* below for more details.

7 Now you are ready to clamp onto a leaf and begin measurements.

See *Clamping onto your first leaf* on page 4-17.

Matching the analyzers

Central to the LI-6800's gas exchange computations is the difference (Δ) between sample and reference gas analyzer readings C_s and C_r . (We are using C for concentration, meaning CO_2 or H_2O). For Δ to be accurate, we need to ensure that $C_r = C_s$ when the IRGAs are receiving identical air streams. We call this condition matching, and it is used to measure correction factor M .

$$M = (C_r - C_s)|_m \quad 4-1$$

where the $|_m$ indicates the readings were made in match mode. M is used to correct subsequent measurements.

$$\Delta = C_s + M - C_r \quad 4-2$$

The value of M is unchanged until another match is performed. How stable is M ? There is some relation between sample and reference that likely depends on concentration, time, temperature, and human activity such as zeroing and/or spanning one or both IRGAs. Let's designate this match relationship as function $m(C)$, to emphasize its potential dependency on concentration. We don't really know what $m(C)$ looks like, but every time we match, we capture its value at a particular concentration, time, temperature, etc. Since matching is done at the reference concentration, we can write

$$M = m(C_r|_m) \quad 4-3$$

Question #1: Does frequent matching assure accuracy?

It may not. It turns out that the shape of $m(C)$ is very important. *Figure 4-1* on the next page illustrates the potential problem.

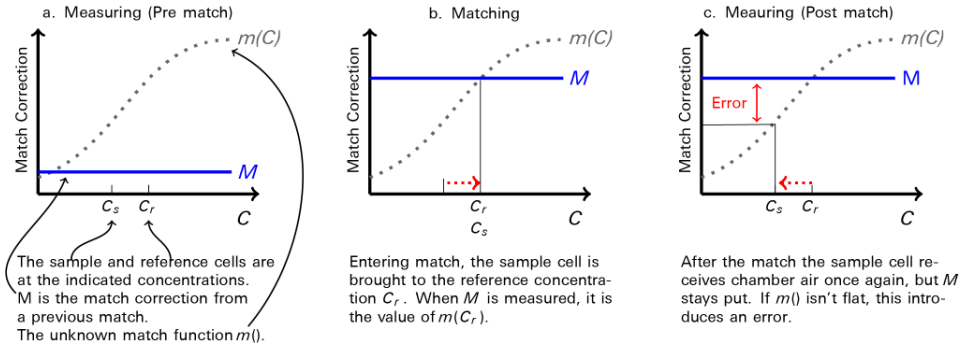


Figure 4-1. Illustration of a hypothetical error function $m(C)$ and its role in a potential post-match error.

The post-match error can be expressed as a fraction by

$$E = \frac{m(C_r) - m(C_s)}{C_r - C_s} \tag{4-4}$$

which is the slope of $m(C)$ over the interval $(C_s; C_r)$. This error (as a fraction or a percentage) is independent of how large or small Δ is. This means that if $m(C)$ has a 1% slope, the post match error in flux will also be 1%.

So, the shape of $m(C)$ is crucial: the more sloped it is, the more you need to re-match if concentration changes, but with each match, there will still be a post-match error. A flat $m(C)$ is good, a sloped $m(C)$ is bad.

Question #2: If $m(C)$ is an unknown function, how can I know the slope?

The match screen in version 1.4 has match graphs for both CO_2 and H_2O . This graph displays the current and previous match values (i.e., values of M). If you do a few matches over a range of concentrations, you will quickly get a sense of the overall slope of $m(C)$ (Figure 4-2 on the facing page).

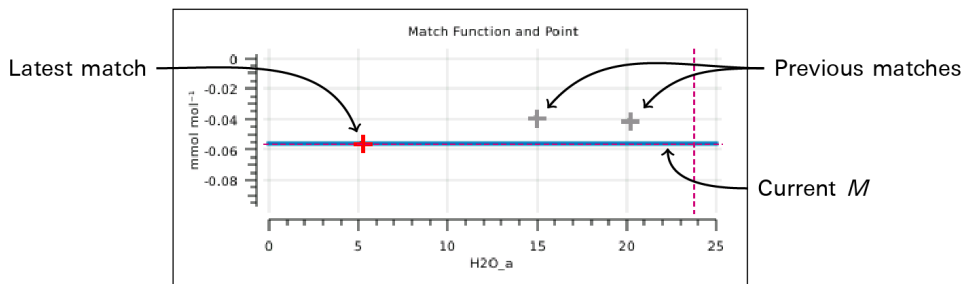


Figure 4-2. The H_2O graph from the match page, shows the history of recent point matches.

In addition, version 1.4 provides a mechanism to acquire "range match" data (Figure 4-3 below). This is a set of values of M taken over the full concentration range in about 5 minutes and shows a very detailed estimate of what $m(C)$ looks like, over that window of time.

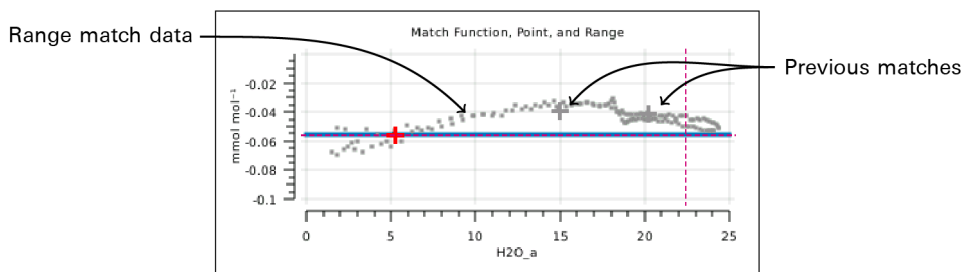


Figure 4-3. The H_2O graph showing recent point matches (+), and range match data (■).

Question #3: Can a sloped $m(C)$ be corrected (i.e., flattened)?

Yes. A sloped $m(C)$ means there is a calibration mismatch that you can correct. Values of M should go to 0 as concentration goes to 0. If they don't, one or both IRGAs need to be properly zeroed. At high concentrations, large M values can be caused by an improperly zeroed IRGA, or when one IRGA (or both) is improperly spanned. Figure 4-4 on the next page illustrates some examples.

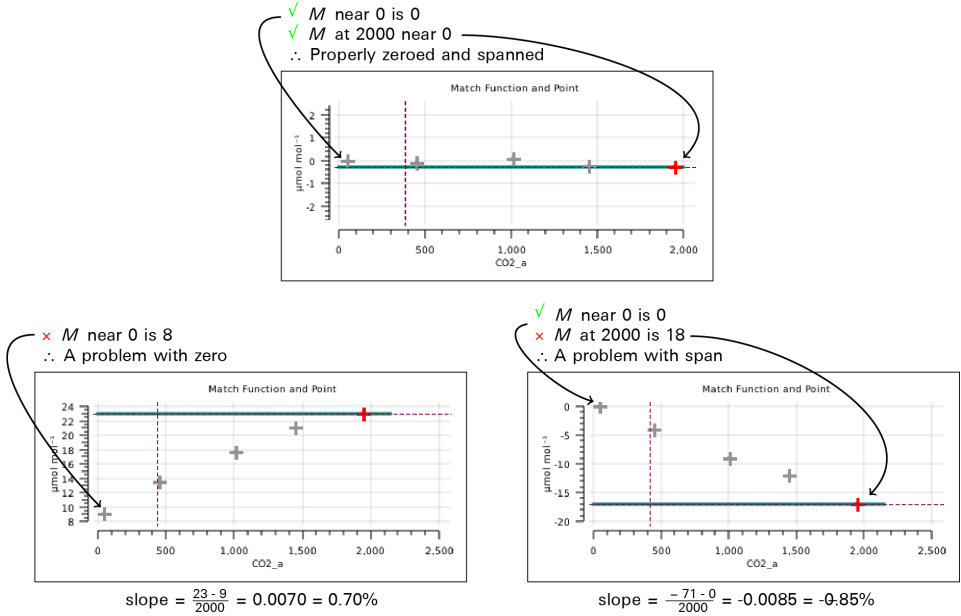


Figure 4-4. Three distributions of CO₂ match points, one with a bad zero, one with a bad span.

Question #4: The post-match error problem stems from the fact that M is a constant. Does it have to be?

Version 1.4 allows the match correction M to be a 3rd order polynomial based on C_s. It can be fit to range match data, yielding an M that continually updates as C_s changes (Figure 4-5 below). This not only removes post-match errors, but reduces the need to match when concentration changes. In other words, instead of a constant M, you can have M(C) ≈ m(C).

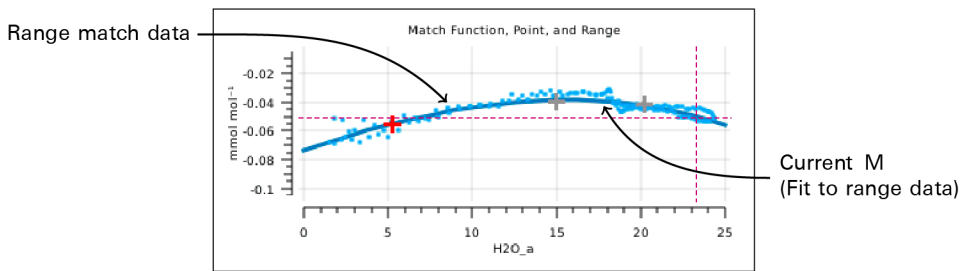


Figure 4-5. The H₂O graph from the match page, with M fit to the range match data.

The match screen

The **Match** screen in version 1.4 has been changed (*Figure 4-6* below). Matching can be triggered by the **Auto** or **Manual** buttons on the left. The status table in the center shows exactly how the current value of Δ is being computed. The graphs to the right show matches done since power on (gray plus marks) and the current match value as a function of concentration (solid green or blue curve). Some new features are available through the **View CO₂** and **View H₂O** buttons at the bottom.



Figure 4-6. Getting in and out of the new Match screen.

The summary table

The summary table (*Figure 4-7* below) makes explicit what the match function is doing. We label it *match()* since the match offset is now potentially a function of uncorrected sample cell concentration.¹

	CO ₂	H ₂ O
Uncorr. Sample:	680.86	15.093
+ match():	0.18	-0.039
Sample:	681.04	15.054
- Reference:	680.59	14.984
Δ:	0.45	0.070

Start with the uncorrected sample cell concentration.
 Add the match correction...
 ...to get the sample cell concentration.
 Subtracting the reference cell concentration...
 ...yields the Δ , on which gas exchange computations are based.

Figure 4-7. Match status table.

¹In general, the match function is a 3rd order polynomial. If the only non-zero coefficient is the offset, then the function is merely a horizontal line: the same value at any concentration.

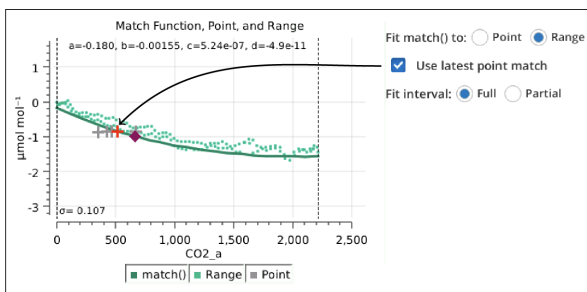
Managing the details

The **View CO₂** and **View H₂O** buttons on the match screen open a screen that allows you to manage matching for that gas. If no range data is present (*Figure 4-10* below), there aren't many options other than acquiring range data (**Acquire**), or removing old match points (**Manage**).

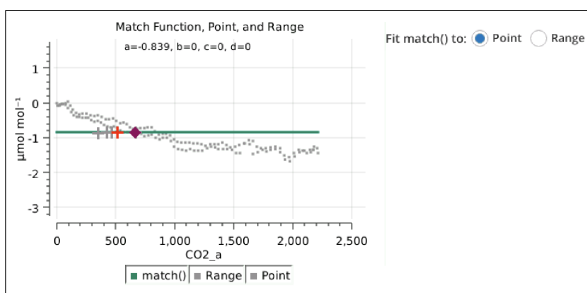


Figure 4-10. The CO₂ Match Data screen shows an expanded version of the CO₂ graph.

When range match data is available (*Figure 4-11* on the next page), you can choose to fit the match function to it, and whether to adjust the fit to the latest point match.



Fit the range match data, but adjust the entire curve vertically so that it goes through the latest point match.



Fit match() through the latest point match.

Figure 4-11. If range match data is available, you can select between using it or not.

The match function can be fit to the full range match data set, or to a subset by setting Fit interval to Partial, which selects a narrow band of range match data that contains the current sample concentration. This band shifts automatically as sample cell concentration changes (Figure 4-12 on the facing page).

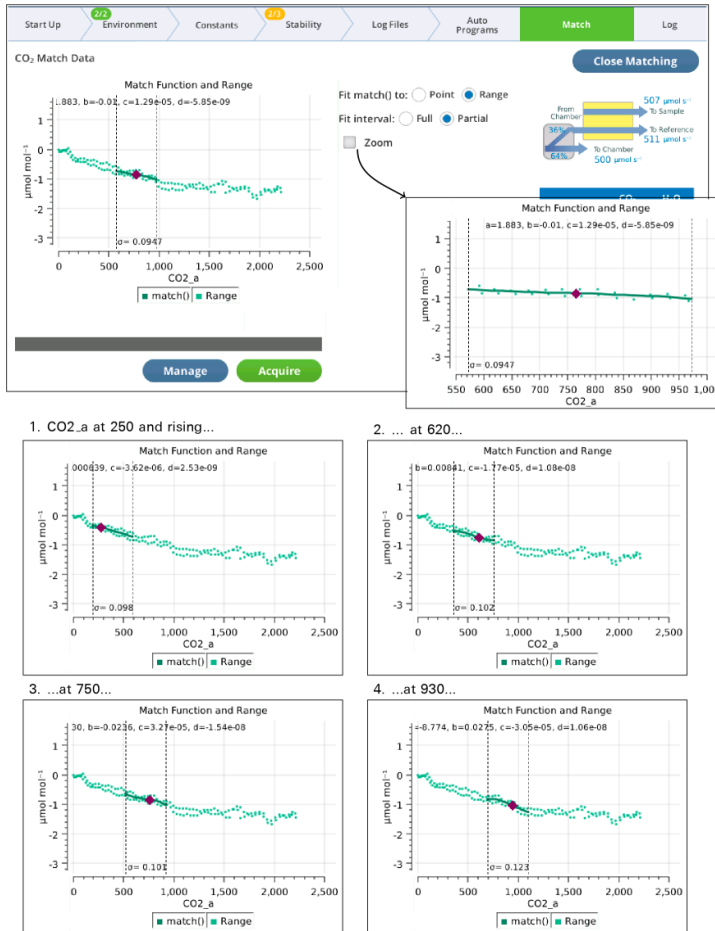


Figure 4-12. When Fit Interval is Partial, a subset of the range match data is fit. This narrower fit interval shifts automatically as needed to keep the sample cell concentration within its domain.

Acquiring range match data

The **Acquire** button will launch a program to collect range match data (Figure 4-13 on the next page). This process takes a few minutes, and should be done without any leaf in the chamber, since the chamber receives no air flow during the process.

The **Acquire** program tries to cover as wide a concentration range as possible. For CO₂ this means having fresh soda lime and an adequate CO₂ cartridge. For H₂O, it means having fresh desiccant and a saturated humidifier tube.

Once running, the **Acquire** program does the following: a) Routes all the flow to the IRGA, splitting it between sample and reference. b) Ramps concentration from low to high and back to low. c) Records the concentrations differences between uncorrected sample and reference during the ramp up and back.

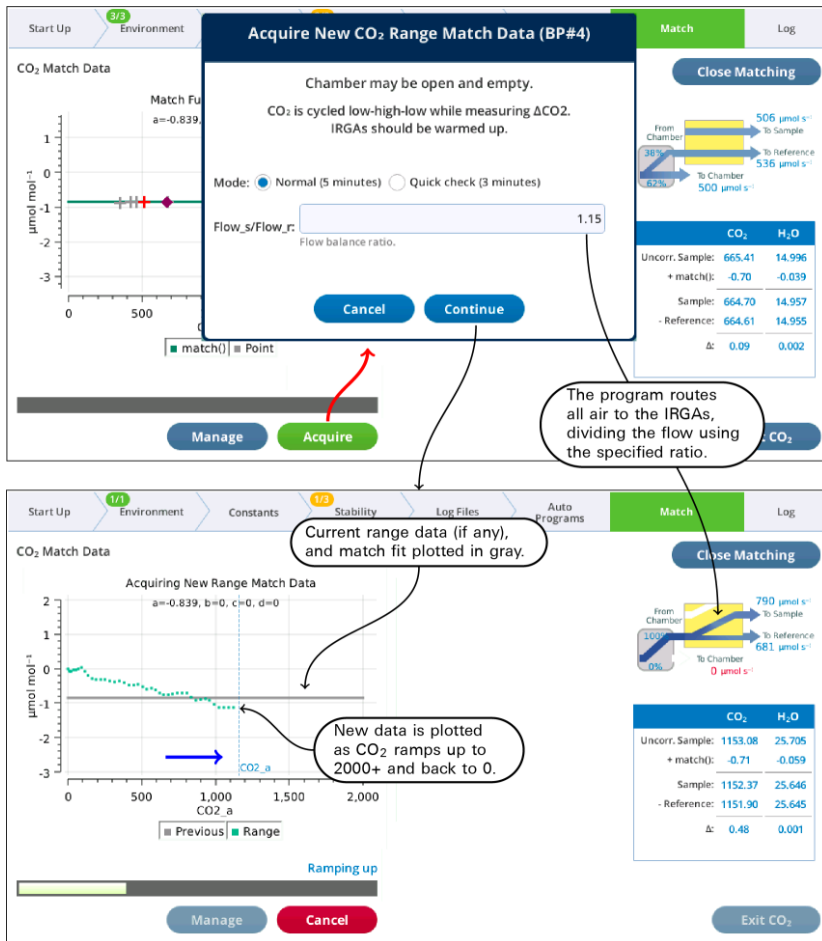


Figure 4-13. Acquiring range match data.

When the program is done, the closing dialog (*Figure 4-14* below) gives you the option of keeping or discarding the new range match data set.

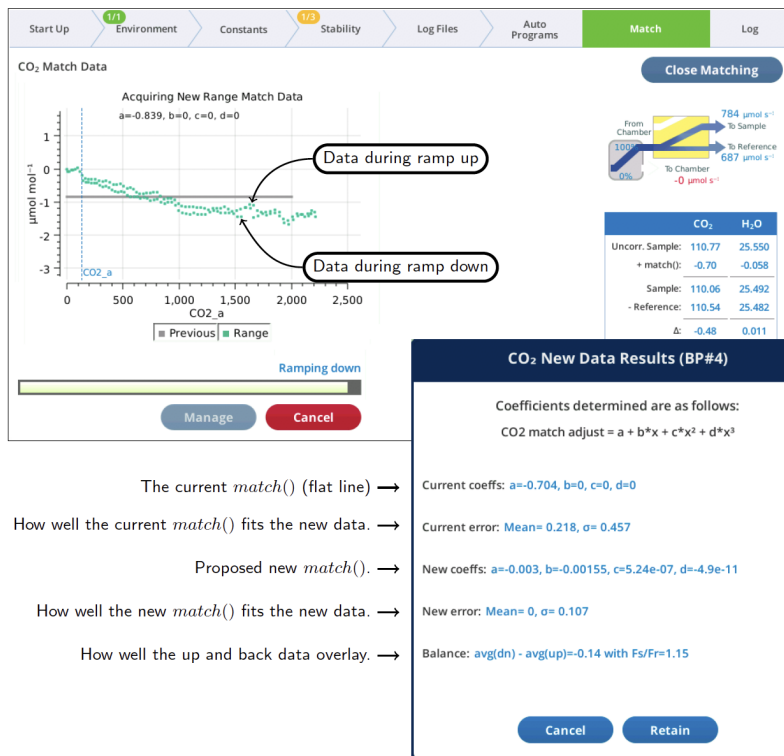


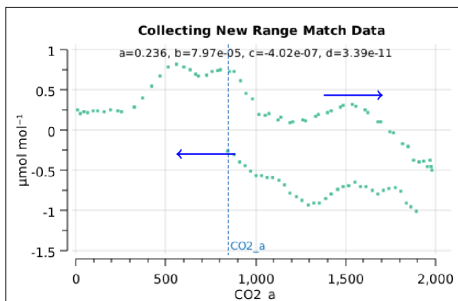
Figure 4-14. The Acquire program closing dialog.

The ramp up values should overlap reasonably well with the ramp down values. If they don't, you can adjust the Flow_s/Flow_r parameter to compensate. Doing the ramp at a faster rate will exaggerate the difference, so you can run Acquire a couple of times at the faster rate to try and get the best Flow_s/Flow_r value. Then, when you slow it down for a normal run, the overlap will be even better.

- If the "coming back" values are higher than the "going up" values, then Flow_s/Flow_r should be *lowered*.
- If the "coming back" values are lower than the "going up" values, then Flow_s/Flow_r should be *raised*.

Values in the 1.10 to 1.20 range seem typical for CO₂. For H₂O, expect flow ratios to be 1.3 to 1.4.

Flow_s/Flow_r = 1.11 (Too low)



Flow_s/Flow_r = 1.20 (Too high)

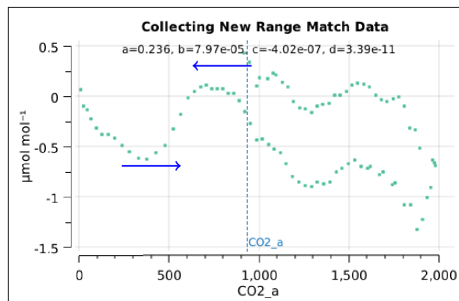


Figure 4-15. The effect of flow ratio on data overlap.

Range match stability

Zero and span

Zeroing and spanning an IRGA will potentially invalidate range match data. In fact, entering those routines will reset the match function to a 0th order polynomial with 0 offset. The range match data is not discarded, but you may need to run **Acquire** after the zero or span adjustments.

Effect of warmup

There is typically a shift in range match data as an instrument warms up. *Figure 4-16* on the facing page compares an original range match data set taken when fully warmed up on a prior day (gray data, same in all plots) with newly acquired data (green points) taken at regular intervals since being awakened from sleep mode, in which it has been for 16 hours prior to the experiment. (The new data is discarded each time, so the plot comparisons are always with the same original data set.)

The largest differences were at 1 minute after warmup, when the curve was shifted down by 0.2 ppm at the low end, and by 1 ppm at the high end. Over the course of 2 hours, the data slowly drifted toward the original data set.

Fortunately, the general shape of the curve was consistent. This suggests you can compensate for disequilibrium by performing a point match (to shift the curve fit), rather than by remeasuring range match data.

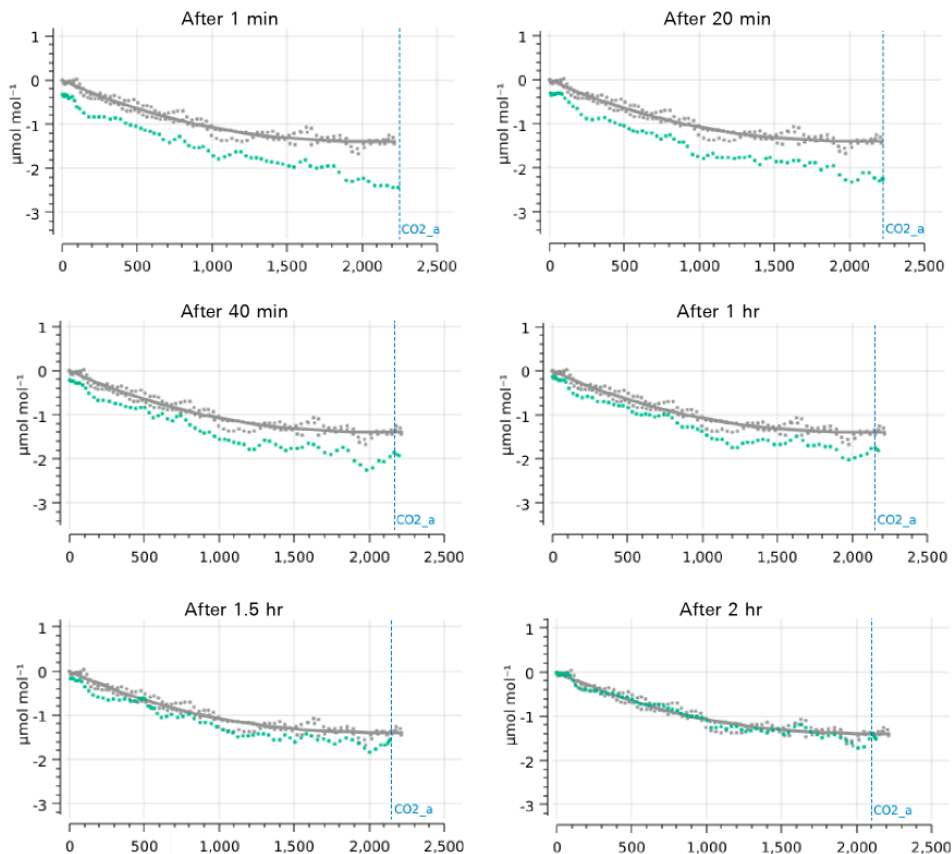


Figure 4-16. An illustration of the effect of warm up on range match data. The time label above each plot is the time since the instrument was waked up after a 16 hour sleep. The gray data points (same in each plot) are the original range match data from a previous measurement.

Match information in data files

Version 1.4 has some changes to what is stored in data files to accommodate the new matching infrastructure:

- The group *Match* has been replaced by *McbEvent* ("match event").
- New group: *McbStatus* ("match status").

- *MchStatus* and *MchEvent* are always in data files, whereas the old *Match* group was optional.
- The variables *CO2_a* and *H2O_a* have moved from group *Meas2* to *Meas*, so are always present in data files.

Additions to *Meas*

as	Meas	Meas	Meas	Meas	Meas	Meas	Meas	Meas	Meas	Meas
1E	CO2_s	CO2_r	H2O_s	H2O_r	CO2_a	H2O_a	Flow	Pa	Pa	ΔPct
	μmol mol ⁻¹	μmol mol ⁻¹	mmol mol ⁻¹	mmol mol ⁻¹	μmol mol ⁻¹	mmol mol ⁻¹	μmol s ⁻¹	kPa	kPa	
56E+09	593.447	593.723	19.02	18.9405	593.735	19.06	610.04	97.7251	0.	
56E+09	593.559	593.744	19.0133	18.9344	593.848	19.053	610.074	97.7213	0.0	
56E+09	438.441	430.666	13.8224	13.4443	438.817	13.8661	610.04	97.7128	0.1	

MchEvent - the most recent Match Event (manual or automatic)

MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent	MchEvent
time	hh:mm:ss	co2_i	h2o_t	count	co2_adj	h2o_adj	co2_match	h2o_match	co2_at	h2o_at	co2_cv	h2o_cv
secs	s	s	s		μmol/mol	mmol/mol	μmol/mol	mmol/mol	μmol/mol	mmol/mol	%	%
1.56E+09	10:45:51	1.56E+09	1.56E+09	1	-0.162	-0.01	-0.288	-0.04	594	19	0.63	0.17
1.56E+09	10:45:51	1.56E+09	1.56E+09	1	-0.162	-0.01	-0.288	-0.04	594	19	0.63	0.17
1.56E+09	10:48:19	1.56E+09	1.56E+09	2	-0.085	-0.01	-0.376	-0.04	431	19	0.28	0.17

MchStatus - The state of the match infrastructure at time of logging

MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	MchStatus	
MatchValv	MatchValv	MatchCO2	MatchH2O	cf_co2_a	cf_co2_b	cf_co2_c	cf_co2_d	cf_h2o_a	cf_h2o_b	cf_h2o_c	cf_h2o_d	co2_range	h2o_range	co2_elapse	h2o_elapse
%	%	μmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	mmol/mol	min	min	min	min
100	100	-0.288	-0.04	-0.10817	-9.4E-05	1.04E-07	6.36E-12	-0.05471	0.003604	-0.00012	0	(0.0,1997.0)	(2.0,24.0)	26039026	26039026
100	100	-0.289	-0.0397	-0.27031	-9.4E-05	1.04E-07	6.36E-12	-0.06449	0.003604	-0.00012	0	(0.0,1997.0)	(2.0,24.0)	0.5	0.5
100	100	-0.376	-0.0377	-0.27031	-9.4E-05	1.04E-07	6.36E-12	-0.06449	0.003604	-0.00012	0	(0.0,1997.0)	(2.0,24.0)	2.2	2.2

Valve pos. | match() | CO2 fit coeffs | H2O fit coeffs | Fit range | Elapsed time

Figure 4-17. Match information as shown in an Excel data file.

Range match information storage

The dataset created during range matching is specific to the head. When it is stored, it is associated with the head serial number that collected the dataset. But the dataset is stored on the console. Therefore, if you attach a different head to a console, you'll need to collect a new range match dataset for that head.

Suggested match protocol

Now that we've covered the important details regarding matching, here is a recommended match protocol.

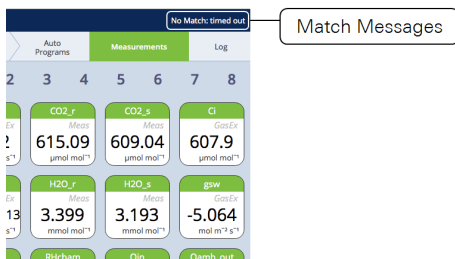
- 1 Acquire range match data for CO₂ and H₂O.

If the values are not small, or if there is a significant slope to the data, zero and span as necessary and check again. Doing this will minimize the need for matching. The range match values should be as small as possible.

- 2 Take care of whatever residual corrections there are by implementing range matching. This should eliminate the need to rematch because of the concentration changes (i.e., any slope in the range match data).
- 3 Consider setting the H₂O matching log option to **Never**. Here's why: During a measurement, it is often the case that the reference air is fairly dry, and the sample air wet. The proper match correction is the one at the sample concentration, not the reference. Range match provides the right correction, point match doesn't. Further, a point match is not only at the "wrong" concentration, but getting there and back can involve some long equilibration times for the sample cell, and not waiting long enough will give you the wrong correction at the wrong concentration. Avoid all that and use range matching.
- 4 For CO₂, implement range matching, and periodically do a point match if temperatures are changing.

Match failure message

When a match fails for some reason besides being cancelled by the user, a message is left on the upper right of the display. The message will persist until another match is started.



If CO₂ was matched but H₂O wasn't, or vice versa, the message will indicate that (e.g., "No CO₂ match: timed out"). The other condition that can generate a message is low flow ("No Match: Flow too low").

Clamping onto your first leaf

If you are new to the LI-6800 and gas exchange measurements, you may wish to work through some of the experiments in this section.

- 1** Select a plant and leaf to measure.

Choose an adequately watered plant that is growing in full or partial sun. Select a leaf that is large enough to fill the whole chamber. Measurements will be more difficult if done on a dry, neglected houseplant that has only seen dim (for the plant) fluorescent lights its whole life. Keep the plant in position near the chamber, but don't clamp onto it just yet.
- 2** Set the **Flow** controls. Under **Environment > Flow**:
 - **Flow**: On
 - **Pump Speed**: Auto
 - **Flow Setpoint**: 500 $\mu\text{mol s}^{-1}$
 - **Press. Valve**: 0.1 kPa
- 3** Set the **H₂O** controls. Under **Environment > H₂O**:
 - Set **H₂O**: On
 - Tap **VDP_leaf**, and enter: 1.0 kPa
- 4** Set the **CO₂** controls. The instrument needs a charged CO₂ cartridge. Under **Environment > CO₂**:
 - **CO₂ injector**: On
 - **Soda Lime**: Scrub Auto
 - Tap **CO₂_s** and enter a setpoint of 400 $\mu\text{mol mol}^{-1}$
- 5** Set the **Fan** speed. Under **Environment > Fan**:
 - Set **Mixing fan**: On
 - Tap **Fan Speed**, then enter 10,000 rpm
- 6** Set the **Temperatures**. Under **Environment > Temperatures**:
 - Check the temperatures to see their current values
 - **Temperature**: On
 - **Tleaf**: On and keep the default setting.
- 7** Set the **Light** level. Under **Environment > Light**:
 - If you aren't using a light source, verify that **Ambient** is selected, then choose the source of ambient light (**Sun+Sky** for outside). Orient the chamber so the leaf is not shaded by the chamber walls.
 - For the LED light sources, be sure that **Head Light Source tab** is selected. Set **Control Mode** to **Tracking** to track ambient. Enter the **Color Spec** of **r90b10** for 90% red and 10% blue.

- For the Fluorometer, go to Environment > Auto Controls. Select a parameter (1 to 6) and set the **Target** to control **Env Light Controls > Qin**. Set the function **F(t)** to tracking. Set **Track this variable** to **Qamb_out**. Check the **AutoRepeat** box, and then tap **Start**. When tracking ambient, the Environment > Light tab will indicate that these parameters are under the control of the **Auto Control**.

8 Under **Constants > System Constants**, check:

- **Chamber type (ChType)**: Automatically selected, but check it to be sure that the chamber is selected
- **Geometry**: Set to the type of leaf you are measuring
- **Leaf Area (S)**: Enter the leaf area enclosed by the chamber. Use the leaf area calculator if you need help
- **K**: Enter the ratio of stomata on one side of the leaf to the other. If you aren't sure, use 0.5.
- **Oxygen**: 21% by default (typical for ambient air). Enter the % oxygen if you are using some other air supply.
- **Leaf temperature**: Choose **Measured** if the leaf temperature thermocouple is able to make contact with the leaf. Otherwise, choose **Energy Balance**.

9 Clamp onto the leaf.

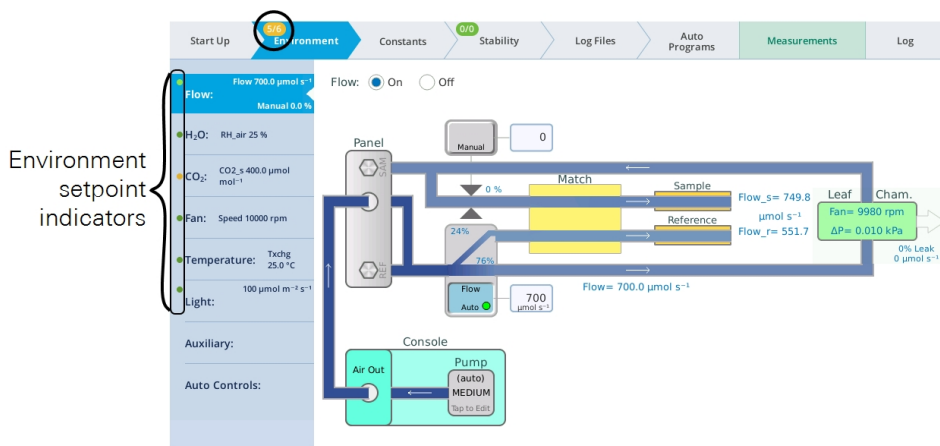
Let the instrument and leaf stabilize for a moment as you go through the remaining steps.

10 Configure logging options under **Log Files**.

- **Match Options**: Select **Always match** for this exercise.
- **Logging Options**: If you are using the fluorometer, set **Flr action at log**: **Nothing**. The other settings don't matter for this tutorial.
- **Open a Log File**: Tap **New File**. Enter a name, then tap **OK**.

11 Check the **Environment** tab.

At the top of the **Environment** tab, you'll see a fraction (5/6 in this case). This shows the number of setpoints achieved over the number of setpoints requested. On the subtabs under Environment, you see a green or yellow dot. Green means the setpoint is achieved, whereas yellow indicates that a requested setpoint is not yet achieved.



- 12 Observe values on the **Measurements** screen.
- 13 After the measurements are stable, press the **Log** button.
The instrument will match and then log the values.

Making measurements

The information in this chapter presumes that you have assembled the LI-6800, learned how to operate the software and control chamber conditions, and that you are sufficiently proficient with the LI-6800 to make measurements on plants.

Survey measurements

The goal of survey measurements is usually to characterize a population, which means sampling a lot of leaves in a short period of time. This means spending a minimal amount of time on any one leaf in order to maximize the sample size.

If your ultimate goal is to be able to say something about a community, or at least a number of plants, it follows that each leaf needs to be measured in similar conditions. The conditions in the chamber should be as close as possible to what the leaf was experiencing prior to the measurement. This saves time as well; you will only be waiting for the chamber to equilibrate (flush out), rather than waiting for the leaf to equilibrate.

Light

Light is the most important variable, so be careful how it changes before and during the measurement. Avoid shading the leaf as much as possible as you move it into the chamber. During the measurement, keep the chamber orientation constant. Be cognizant of the recent light history of the leaf. If you are measuring sunlit leaves, don't select one that happens to be in a small sunfleck, or one that became sunlit when you moved some stems out of the way. When you put a leaf into a clear-top chamber, the light incident on the leaf will be reduced by about 10%. Photosynthesis may respond fairly quickly to that reduction, and should equilibrate in a few seconds. Stomatal responses take longer, but 10% light reduction will usually not cause a measurable change in conductance.

Avoid large changes of light. A common error is to reorient the chamber during a measurement. Whether you do it inadvertently (distracted by your Twitter feed) or intentionally (avoiding shade), it's bad.

For outdoor survey measurements, clear days are a blessing and partly cloudy days are a curse. With only short periods of uninterrupted sun, the leaves will be in perpetual disequilibrium. Measurements taken under these conditions will be nearly impossible to interpret, and therefore meaningless. The use of a light source will guard against the odd cloud shadow interrupting a measurement on a nearly clear day. With more abundant clouds, the most a light source can offer is the chance to let each leaf equilibrate for 10 or 15 minutes in constant light, and that makes for very slow survey work.

CO₂

Since photosynthesis is a function of CO₂, it is important to have the chamber CO₂ as consistent as possible. To accomplish this, set the instrument to control CO₂ on sample and set the target to about 400 $\mu\text{mol mol}^{-1}$, so the leaf draws CO₂ down to ambient levels.

Humidity

Set the humidity between 50% and 80% RH. You want conditions that are familiar to the plant, but be aware that the mixing fan will decrease the thickness of the leaf boundary layer, so you need a setting that is higher than ambient humidity to maintain stable conditions for the leaf and ensure that the plant does not significantly change its stomatal opening after being enclosed in the chamber.

Flow

Use a fixed flow rate, medium or high, with little or no desiccant scrubbing. Here's why: a fixed flow rate minimizes the time for the system equilibration, once a leaf is in the chamber.

Temperature

There are two schools of thought about the temperature control and survey measurements: one is that you shouldn't use the cooler so that your batteries last longer. The other maintains that you should use the cooler to maintain ambient temperature, so that the chamber doesn't get hot from being in the sun. You decide.

Be aware that a large change in temperature will have an effect on photosynthesis, so if the temperature is changing rapidly or these survey measurements are part of a diurnal curve, temperature control is advised.

Matching the IRGAs

Match once on the first leaf (or a "trial leaf"), and perhaps every 30 minutes or so after that, especially if temperature is changing.

Logging considerations

You will need to decide some other logging considerations, as well:

- Leaf Area?
Does the leaf fill the chamber aperture, or is it changing from leaf to leaf? How and when will it be measured? Do you wish to be prompted for leaf area as you go, or enter leaf area into the spreadsheet later?
- Extra Data?
Are there extra data you wish to record, such as plant numbers or remarks, to help identify the data later? If so, consider configuring the user constants and activate the **Prompt on log** option under **Log Files > Logging Options**.
- How Many Log files? Log Options?
Are all the measurements destined for one file, or should there be several? If several, what's the rationale for the grouping? Does it matter in which order the measurements are done?

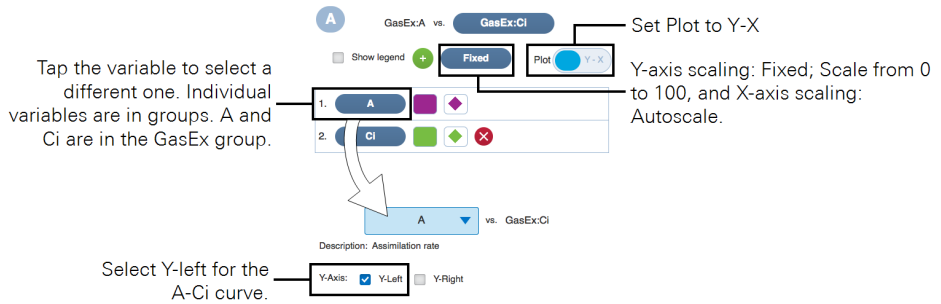
The simplest approach is to log all the data into one file. If, for some reason, you desire multiple files, then make your measurements so that File1 is finished before File2 is started.

- Use Stability criteria?
Do you want to guess at when to log, or use some objective criterion?

A manual response curve

This tutorial is a step-by-step guide to manually generating a CO₂ response curve. The configuration given below will work well for many plants, but you may need to change the environmental control settings if you are measuring a small leaf or a plant with low photosynthesis rates.

- 1 Set the **Environment** controls.
 - **Flow**, set **Flow**: On; **Pump Speed**: Auto; **Flow Setpoint**: 500 $\mu\text{mol s}^{-1}$; and **Press. Valve**: 0.1 kPa. Use a lower flow rate for small leaves or plants with low photosynthesis rates.
 - **H₂O**, set **H₂O**: On; and **RH_{air}**: 50%.
 - **CO₂**, set **CO₂ injector**: On; **Soda Lime**: Scrub Auto; Tap **CO₂_s** and enter a setpoint of 400 $\mu\text{mol mol}^{-1}$.
 - **Fan**: Set **Mixing fan**: On; Tap **Fan Speed**, then enter 10,000 rpm.
 - **Temperatures**: Check the temperatures to see their current values; **Temperature**: On; **Tleaf**: Control at something close to ambient.
 - **HeadLS** or **Fluor**: For field grown plants, use 1,500 $\mu\text{mol m}^{-2} \text{s}^{-1}$; half that much for greenhouse or growth chamber plants. If you don't have a light source, do the experiments in a growth chamber or outdoors. But beware: The experiment is meaningless without steady light.
- 2 Clamp onto the leaf.
Let the instrument and leaf stabilize for a moment as you go through the remaining steps.
- 3 Set up a graph so you can view the data as they are collected.
You'll want to plot Assimilation (A) over intercellular CO₂ (C_i). Under Measurements, select plot A and then tap **Edit Graphs**.



- 4 Configure logging options under **Log Files** and open a log file.
 - **Match Options:** Select **Always match** for this exercise.
 - **Logging Options:** If you are using the fluorometer, set **Flr action at log:** Nothing. The other settings don't matter for this tutorial.
 - **Open a Log File:** Tap **New File**. Enter a name, then tap **OK**. Name the file "Sample CO₂ curve," or something similar.
- 5 Wait for stability, and log the first data point.
- 6 Lower next CO₂ value by 100 μmol mol⁻¹, wait for stability, and then log another point.
- 7 Repeat until done.

Use these targets for the reference concentration: 300, 200, 100, and 30 (for C₃ plants) or 0 (for C₄). The last point is designed to be below the compensation point. Notice that the change in CO₂ setpoint is accompanied by a brief disruption in the system stability. Try to get 4 or 5 points between your starting value and ending points. Go down to 30 μmol mol⁻¹ or so for C₃ plants, or 0 for C₄ plants.
- 8 Finish the curve back at the starting point.

Set the sample CO₂ at 400 μmol mol⁻¹. See how long it takes for photosynthetic rates to return to normal.
- 9 Do some points above ambient, such as 600, 800, 1000, and 1200 μmol mol⁻¹.
- 10 Analyze the data.

Use the graphs to answer these questions: What's the CO₂ compensation point? Did humidity stay constant over the experiment? How much did the stomatal conductance change over the measurement?

Carbon dioxide response curves

Why measure CO₂ response? An $A-C_i$ curve (assimilation rate plotted against inter-cellular CO₂ concentration) can provide a number of insights into the bio-chemistry of a leaf or plant, such as:

- **CO₂ compensation point:** The value of C_i where photosynthesis and respiration are in balance.
- **Carboxylation efficiency:** The initial slope provides an *in vivo* measure of the carboxylation efficiency. If measuring a C₃ leaf, the slope is proportional to the maximum activity of RuBisCO. This is sometimes called the mesophyll conductance.
- **Stomatal limitations:** Stomatal limitation of photosynthesis can be quantified with CO₂ response curves.
- **Carboxylation limitations:** Within the mesophyll, carboxylation limitations can be separated from electron transport limitations.

CO₂ response curve strategies

Below are some things to consider when doing a CO₂ response curve.

Light

Light should be held constant and is typically at a non-limiting light level. The LED light source or fluorometer will work just fine for CO₂ response curves. Set it to the maximum intensity that is familiar to the plant, and give the leaf some time to equilibrate.

CO₂

In what order should the curve be measured? Consider the parameters that you are most interested in modeling. We recommend starting at ambient and tracking downward toward low concentrations, and then returning to ambient and tracking upwards toward high concentrations. Why? Because at low CO₂ concentrations, RuBisCO can be deactivated, while at high concentrations, stomata can close. Avoid spending a lot of time at each extreme.

Temperature

The response curve should be measured under constant temperature conditions. Operate the coolers at a constant leaf temperature.

Humidity control

Set the instrument to control sample RH between 50% and 75%. You can control on VPD leaf. If you're not sure of the proper setpoint, clamp onto the leaf with chamber RH between 50 and 70%, and check the VPD leaf measurement. Remember that the mixing fan will reduce leaf boundary layer, but you want the leaf to experience the same humidity in the chamber as it did before the chamber.

Matching

Since the concentrations of CO₂ are covering a large range, match before each reading.

Step-by-step carbon dioxide response curve

Here's how to make an automatic CO₂ response curve. It uses an Auto Program.

- 1** Set the **Environment** controls.
 - **Flow**, set **Flow**: On; **Pump Speed**: Auto; **Flow Setpoint**: 500 $\mu\text{mol s}^{-1}$; and **Press. Valve**: 0.1 kPa
 - **H₂O**, set **H₂O**: On; and **RH_{air}**: 50 to 75%
 - **CO₂**, set **CO₂ injector**: On; **Soda Lime**: Scrub Auto; Tap **CO₂_s** and enter a setpoint of 400 $\mu\text{mol mol}^{-1}$
 - **Fan**: Set **Mixing fan**: On; **Fan Speed**: 10,000 rpm
 - **Temperatures**: On; **Tleaf**: 27.0 °C or something close to ambient
 - **HeadLS** or **Fluor**: Use 1,500 $\mu\text{mol m}^{-2} \text{s}^{-1}$, or a non-limiting level for your plant.
- 2** Set area and stomatal ratio.

Are the values correct under **Constants > System Constants**?
- 3** Open a log file.

Make sure you have the Match Options and Logging Options configured.
- 4** Clamp onto the leaf.
- 5** Launch the Auto Program.

Under **Auto Programs**, select **CO2 Response**. Configure the settings or just go with the default settings, then tap **Start**.

6 Watch the curve develop.

Under **Measurements**, check **CO2_r** and **CO2_s**. You can also set up an A-Ci graph, as described in *A manual response curve* on page 4-23.

The data are stored in the log file. You can inspect it under **Log Files > Analyze Logs**. Select the file, then tap **View**.

Light response curves

Why measure light response? Starting from total darkness, in which there is no photosynthesis, the first few photons to be absorbed by the leaf will be used with the greatest efficiency. As light increases, the efficiency drops, and eventually subsequent increases in light yield little or no increase in photosynthesis. Thus, a light response curve can provide indications of the:

- **Dark respiration rate:** The photosynthesis rate with no light,
- **Light compensation point:** Absorbed quantum flux for which photosynthesis and respiration are balanced
- **Quantum efficiency:** Initial slope of the assimilation rate, and
- **Light saturated photosynthetic rate** (A_{sat}).

Shade-adapted species tend to have lower dark respiration rates, lower compensation points, higher quantum efficiencies, and lower maximum photosynthetic rates than sun adapted species.

Light curve strategies

Depending upon what you are trying to measure, there are a couple of approaches to light curves.

Rapid

Since the photosynthetic apparatus responds almost immediately to light, especially drops in light intensity, the quickest method is to start with a leaf equilibrated to high light, and decrease the light, spending perhaps 1 or 2 minutes at each light value, and dropping in steps of $200 \mu\text{mol m}^{-2} \text{s}^{-1}$ or less. When you do this, you'll find that the stomata have not had time to adjust, and tend to remain more open at low light values than they normally would. This manifests itself as a steadily rising

C_i throughout the measurement. There's nothing wrong with this, but be careful how you use the conductance measurements from a rapid light curve, because they are not equilibrated values.

Slow

Another approach is to do a slow curve, giving the stomata time to equilibrate at each light level. Going slowly, you can work from dark to light, or light to dark. If you wait 15 or 20 minutes at each level, you will find that C_i will be fairly constant throughout the measurement, indicating that the stomata are fully adjusted. In fact, you could use C_i as an indicator of when to log the next record at all but the darkest levels.

Survey

A third approach is to generate a light curve using multiple leaves that are equilibrated at a range of light values. This has the advantage of being fairly quick, yet has equilibrated values. The potential for difficulty comes from using multiple leaves, thus bringing age differences and other factors into the response curve. The survey approach is better suited for some species than others. In deciduous trees, for example, leaf age is not particularly related to position in the canopy. With this approach you can achieve a range of light levels by selecting leaves that are tilted with respect to the sun, and in varying degrees of shade. The orientation of the sunlit leaves is a problem, however, unless you are using a light source when you clamp onto them. With a clear chamber top, leaves that are tilted with respect to the sun will be shaded by the chamber wall, and this is to be avoided at all costs. If, however, you use a light source, you can set the appropriate value first, or have it automatically track the ambient light as measured by the external PAR sensor.

Sunfleck and shade method

The fourth approach offered here is to separate each new light level with the starting light value, with time to equilibrate. That is, use a sequence such as: 100, 1000, 1800, 500, 1800, 300, 1800 $\mu\text{mol m}^{-2} \text{s}^{-1}$. (The starting point needn't be high; you could work the other direction with shade leaves.) Data collected in this manner might be appropriate for addressing questions of light dynamics in canopies.

Operational considerations

Once you decide on the strategy, you then need to decide how the chamber controls should be set and how data are to be collected.

Light

The best light sources for light response curves are the 3×3 cm light source or the fluorometer. In the absence of a light source, a light curve cannot be automated, but is still possible to make one. Neutral density filters, for example, can provide means to reduce sunlight or other sources by known amounts. The survey technique discussed earlier can be done without a light source.

Temperature

Ideally, the response curve should be measured at a constant temperature, so operate the coolers to maintain constant leaf temperature.

Humidity control

Set the instrument to control sample RH between 50% and 75%. You can control on VPD leaf. If you're not sure of the proper setpoint, clamp onto the leaf with chamber RH between 50 and 70%, and check the VPD leaf measurement. Remember that the mixing fan will reduce leaf boundary layer, but you want the leaf to experience the same humidity in the chamber as it did before the chamber.

Matching

Since the concentrations in the IRGAs aren't going to be changing much during a light curve, there's no real reason to match after every measurement. Match once before starting and set **Only match if $\text{abs}(\text{co2_r-CO2_s}) < 5$** .

Auto programs

Auto Programs enable you to record the light curve using predefined settings.

Step-by-step light response curve

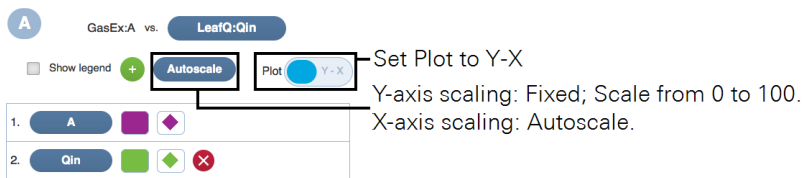
Here's how to make an automatic light response curve. It uses an Auto Program and does a rapid response curve.

- 1 Set the Environment controls.
 - Flow, set Flow: On; Pump Speed: Auto; Flow Setpoint: $500 \mu\text{mol s}^{-1}$; and Press. Valve: 0.1 kPa
 - H₂O, set H₂O: On; and RH_{air}: 50 to 75%
 - CO₂, set CO₂ injector: On; Soda Lime: Scrub Auto; Tap CO_{2_s} and enter a setpoint of $400 \mu\text{mol mol}^{-1}$
 - Fan: Set Mixing fan: On; Fan Speed: 10,000 rpm
 - Temperatures: On; Tleaf: 27.0 °C or something close to ambient
 - HeadLS or Fluor: Use $1,800 \mu\text{mol m}^{-2} \text{s}^{-1}$, or a light-saturating level for your leaf
- 2 Clamp onto the leaf.
- 3 Set area and stomatal ratio.

Are the values correct under Constants > System Constants?
- 4 Open a log file.

Make sure you have the Match Options and Logging Options configured.
- 5 Set up the graphs.

Under the Measurements tab, select Graph A, then tap Edit Graph. Configure a graph of A and Q_{in} .



- 6 Select the Auto Program.

Select Light Response from the Programs: list. Adjust the settings or just go with the default values.
- 7 Tap Start.

Watch the measurement unfold under the Measurements tab.

Fluorescence experiments

These experiments cover some basic fluorescence measurements. They serve as a good introduction into fluorometry and the Multiphase Flash™ Fluorometer.

Determine F_v/F_m

F_v/F_m is an estimate of the maximum quantum efficiency of PSII reaction centers. This ratio is calculated from two parameters: F_o and F_m . F_o is the fluorescence level of a dark-adapted plant with all PSII primary acceptors ‘open’ (Q_A fully oxidized). F_m is the maximal fluorescence level achieved upon application of a saturating flash of light, such that all primary acceptors ‘close’ (Q_A fully reduced). Variable fluorescence, F_v , is the difference between F_o and F_m . The ratio of variable-to-maximal fluorescence is normally between 0.75 and 0.85, depending on leaf health, age, and preconditioning. Since this is a fluorescence-only experiment, don't worry about the gas exchange parameters.

1 Dark-adapt the leaves.

The best technique is to leave the plant in complete darkness overnight, taking measurements just before dawn. For our purposes, however, it will be adequate to dark-adapt the leaf for 20 minutes. You can wrap the entire leaf in aluminum foil or excise it from the plant and put it in a dark drawer (not for too long; you don't want it to wilt).

2 Open a log file and configure the options.

You can do the flash without a log file, but be aware that you won't record gas exchange data if you do it that way. Since you want to initiate the flash when you press the log button, configure the following settings:

- Under **Log Files > Match Options**: Set to **Never match**, since we aren't doing gas exchange measurements right now.
- Under **Log Files > Fluorometer Options**, set **Flr Action at Log**: Select **1: FoFm (dark)** or **FsFm' (light)** and select **Flash type: Rectangular**.
- Under **Log Files > Open a log file**, create a new file for the data.

3 Set up a graph of fluorescence.

Under the **Measurements** tab:

- Select graph **F** to make it easy to remember (F for Fluorescence)
- Tap **Edit Graphs**

- Set Plot: Time
- Select the variable: Under **FirLS**, select **F** (demodulated fluorescence)

4 Configure the Actinic light.

Under **Environment > Light > Fluorometer** set **Control Mode** to **Off** so that the actinic light is off.

5 Configure the Measuring beam.

Under **Environment > Fluor > Measuring**:

- **Measuring**: On
- **Dark mod rate**: 50 Hz

You want the highest rate you can get without inducing photosynthesis in the leaf. Look at the **Q_modavg** value to see how much actinic light is being provided.

6 Configure the Rectangular flash.

- **Red target**: 8000 $\mu\text{mol m}^{-2} \text{s}^{-1}$
- **Duration**: 1000 ms
- **Output rate**: 100 Hz
- **Margin**: 5 points

7 Close the chamber onto the leaf.

Observe the graph of **F**. You'll see the measured fluorescence from the leaf. It should stabilize fairly quickly, unless the measuring beam is too high, in which case you may need to set the **Dark mod rate** lower.

The moment the chamber was closed onto the leaf

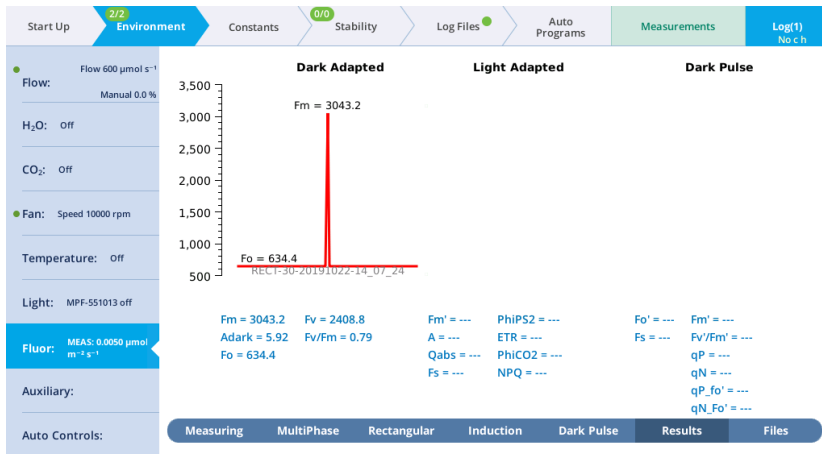


- 8 After the leaf has stabilized (dF/dt will approach 0), tap the **Log** button. The flash will be evident on the graph.

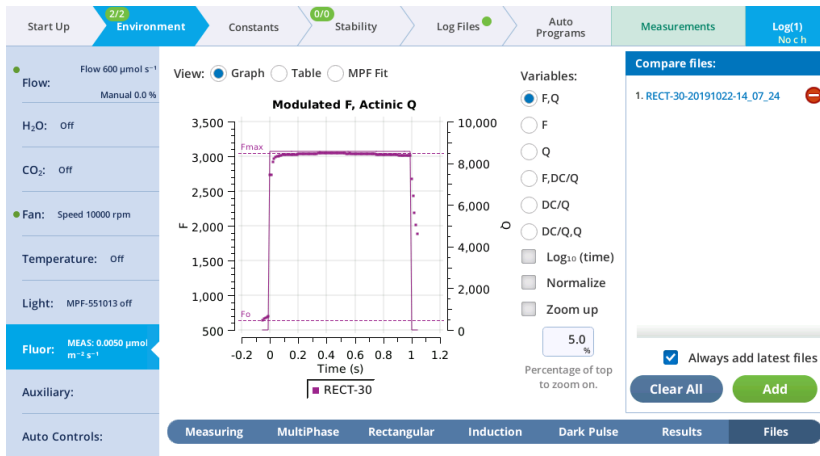


- 9 View the Flash Results.

Go to **Environment > Fluor > Results** to review the flash data.



10 Tap Files to see a graph of the fluorescence emission and flash.



11 Select Table to see the details recorded with the flash.

The screenshot shows the LI-6800 software interface with the 'Log Files' tab selected. The main display is a table titled 'RECT-30' showing recorded flash details. The table has columns for DATE, DEVICE, DURATION, D_RED_PERCENT, EVENT_ID, FMAX, F₀, MARGIN, MODRATE, OUTRATE, QMAX, Q_RED_SETPPOINT, T@FMAX, and T@FMAX. The interface includes various control panels on the left and a 'Compare files' panel on the right.

RECT-30		
DATE	20191022	DATE
DEVICE	MPP-551013	DEVICE
DURATION	1000	DURATION
D_RED_PERCENT	90	D_RED_PERCENT
EVENT_ID	30	EVENT_ID
FMAX	3043.24	FMAX
F ₀	634.4	F ₀
MARGIN	5	MARGIN
MODRATE	125000	MODRATE
OUTRATE	100	OUTRATE
QMAX	8575.06	QMAX
Q_RED_SETPPOINT	8000	Q_RED_SETPPOINT
T@FMAX	0.45	T@FMAX

Determine PSII efficiency

Φ_{PSII} (also known as $\Delta F/F_m'$) is the fraction of absorbed PSII photons that are used in photochemistry, and is measured with a light-adapted leaf. It is calculated from F_s and F_m' , where F_s is steady-state fluorescence and F_m' is the maximum fluorescence from a light-adapted sample upon application of a saturating flash. See also Genty et al. (1989).

1 Select light-adapted leaves.

For this exercise, select a light-adapted plant that has had at least 20 minutes of acclimation at the desired light level. For the first part of this experiment, you'll need several leaves that are about the same age and have similar illumination. At the end, you may want to try some leaves that have been at other light levels (shade vs. sun leaves).

2 Set up a graph of fluorescence.

This is just like the graph in the previous tutorial.

3 Configure the fluorometer.

Under **Environment > Light > Fluorometer**:

- **Control Mode:** Setpoint
- **Setpoint:** set Q target to the ambient level (displayed as **Ambient** to the upper right)
- **Color Spec:** r90B40

Under **Environment > Fluor > Measuring**:

- **Measuring Beam:** On
- **Dark mod rate:** Leave at the default setting
- **Light mod rate:** 50 kHz
- **Flash mod rate:** 250 kHz

Under **Environment > Fluor > Rectangular**:

- **Red target:** 8000 $\mu\text{mol m}^{-2} \text{s}^{-1}$
- **Duration:** 1000 ms
- **Output rate:** 100 Hz
- **Margin:** 5 points

4 Open a log file and configure the options.

Under the **Log Files** tab:

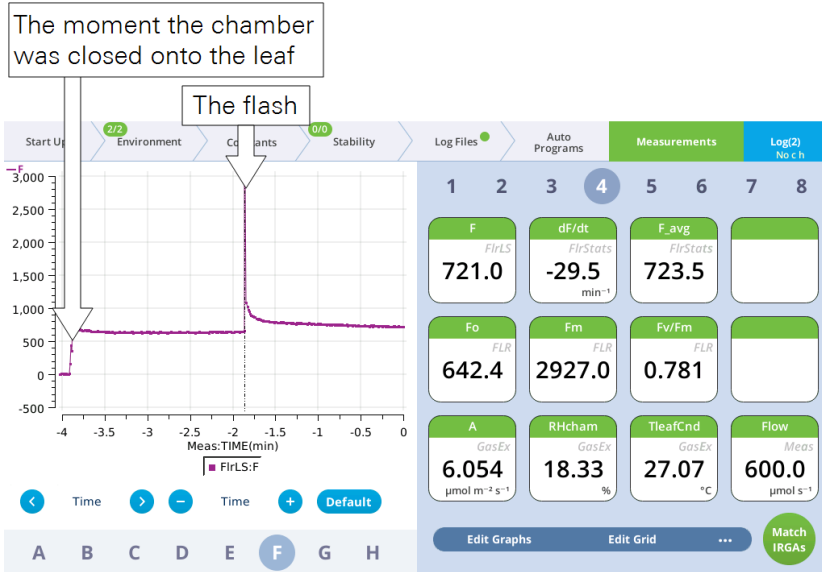
- **Match Options:** Only match if Elapsed time 10 minutes since last match, CO_2_r has changed > 100 ppm since last match, and $\text{abs}(\text{CO}_2_r - \text{CO}_2_s) < 1$ ppm.
- **Flr Action at Log:** Select 1: **FoFm (dark)** or **FsFm' (light)**.
- **Flash type:** Rectangular

Start with a rectangular flash. Rectangular is good for low-light plants, such as those in a greenhouse or growth chamber. A multiphase flash is better for full sun plants.

5 Close the chamber onto the leaf.

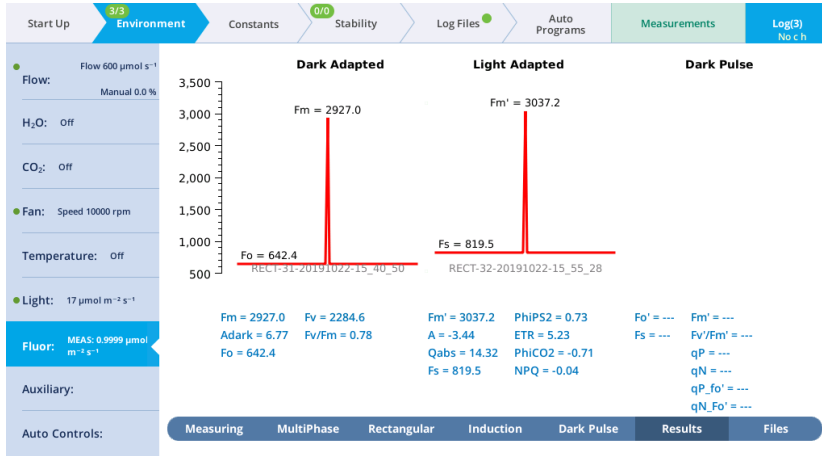
Observe the graph of Fluorescence. You'll see the leaf fluoresce. It should stabilize fairly quickly, unless the measuring beam is too high, in which case you may need to set the Dark mod rate lower.

6 Tap the **Log** button. Check Φ_{PSII} .

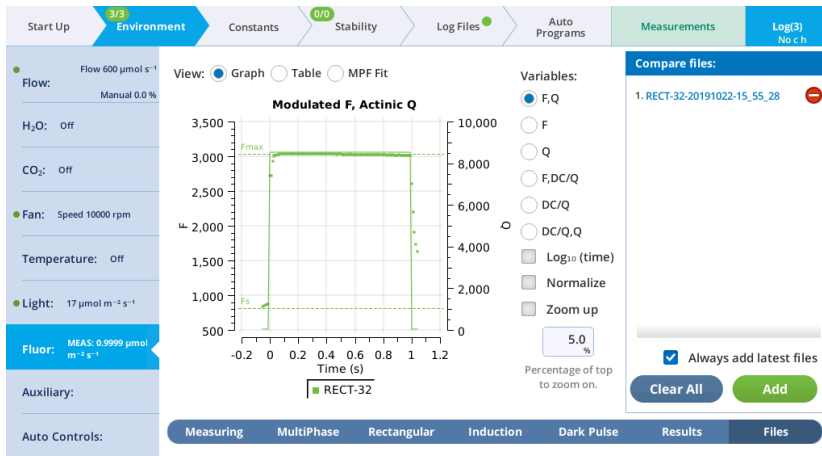


7 View the flash results.

Under **Environment > Fluor > Results**, if you have continued from the previous exercise without turning off the instrument, you'll see **Dark-Adapted** (from the prior measurement) and **Light-Adapted** parameters (from this exercise).



8 Tap View under the Light-Adapted graph.



Repeat steps 7 and 8, measuring leaves of similar age and light history.

For further study, try different light values. Compare PSII quantum yields of leaves adapted to high (2,000 $\mu\text{mol m}^{-2} \text{s}^{-1}$) and low (100 $\mu\text{mol m}^{-2} \text{s}^{-1}$) light levels.

How does $\Delta F/F_m'$ differ between leaves adapted to high vs. low light?

PSII quantum yields are usually high under low light conditions because a large proportion of the absorbed light is used in photochemistry. High light-adapted

plants tend to have low $\Delta F/F_m'$ values because a higher proportion of the absorbed energy is dissipated through non-photochemical processes.

Determine F_v/F_m and quenching coefficients

Three other useful fluorescence parameters will be explored in this experiment. F_v'/F_m' represents the efficiency of energy harvesting by oxidized (open) PSII reaction centers in the leaf. Two competing processes that quench (decrease) the level of chlorophyll fluorescence in the leaf are referred to as photochemical (qP) and non-photochemical (qN) quenching.

Many disciplines use these parameters, but the latter two are particularly useful as quantifiers in stress physiology research. All three of these parameters require F_o' , the minimal fluorescence (in the dark) of a light-adapted leaf. How can this be determined? One method would be to allow the sample to dark-adapt and wait until all PSII centers oxidize (usually 20 minutes or more). A more expedient method would be to use far-red light to preferentially excite PSI and force electrons to drain from PSII. Only a few seconds of far-red time are needed for this to occur. The fluorometer provides a “dark pulse” routine which uses this second method to determine F_o' . See *Figure 3-23* on page 3-68 for an illustration of the dark pulse timing parameters.

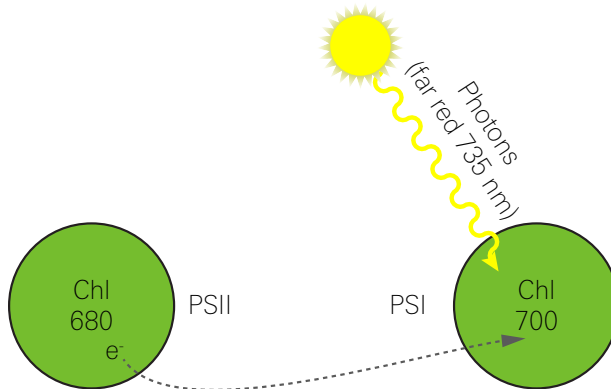


Figure 4-18. PSI is preferentially excited by far-red light, which drives electron transport of PSI, and thus drains electrons from PSII. This is a method of rapid equilibration for determining F_o' .

- 1** Select some plants.
You'll need both dark- and light-adapted leaves for this exercise.
- 2** Configure the fluorometer.
Under **Environment > Light > Fluorometer** set **Control Mode** to Off, so that the actinic light is off.
- 3** Configure the **Measuring** beam.
Under **Environment > Fluor > Measuring**:
 - **Measuring**: On
 - **Dark mod rate**: 50 Hz
 - **Light mod rate**: 1 kHz
 - **Flash mod rate**: 125 kHzYou want the highest rate you can get without inducing photosynthesis in the leaf. Look at the Q_modavg value to see how much actinic light is being provided.
- 4** Configure the **Rectangular** flash.
 - **Red target**: $8000 \mu\text{mol m}^{-2} \text{s}^{-1}$
 - **Duration**: 1000 ms
 - **Output rate**: 100 Hz
 - **Margin**: 5 points
- 5** Configure the **Dark Pulse**:
This experiment requires the calculation of F_o' after the saturating flash, which requires configuring the dark pulse parameters under **Environment > Fluor > Dark Pulse**:
 - **FarRed target**: $25 \mu\text{mol m}^{-2} \text{s}^{-1}$ (target intensity for the dark pulse)
 - **Duration**: 5 (Time that Actinic light is off)
 - **Before**: 1 (Number of seconds that far red is on before actinic is off)
 - **After**: 1 (Number of seconds that far red is off after actinic is off)
 - **Margin**: 5 (Number of data points before and after)
- 6** Set up a graph of fluorescence (F).
This is just like the graph in Experiment 1.
- 7** Open a log file and configure the options.
 - **Flr Action at Log**: Select **2: FoFm (dark) or FsFmFo' (light)**.
 - **Flash type**: Rectangular
- 8** Clamp onto your dark-adapted leaf.

With the actinic light off and the measuring light on, wait until F becomes stable.

- 9 Measure F_v/F_o (Tap the log button).

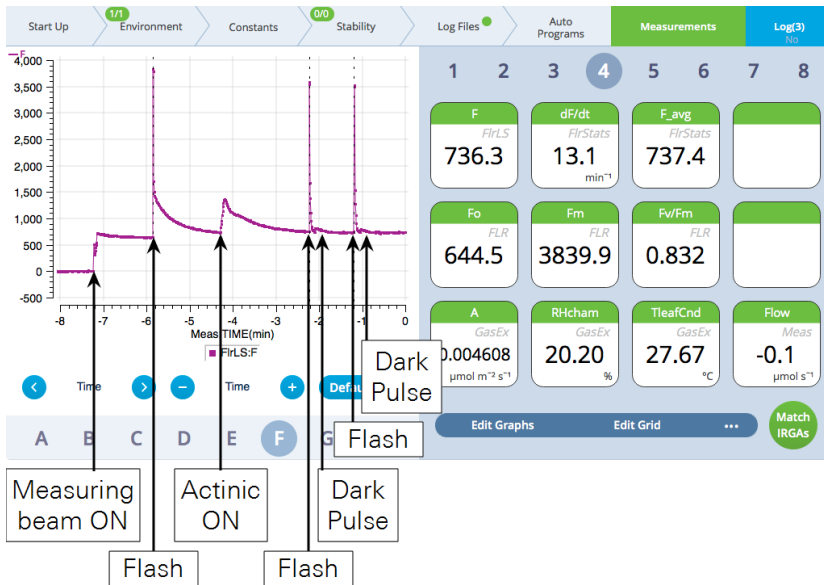
F_o is immediately set to the current value of F . Then a saturating flash is done, and F_m is set to the maximum value during the flash. Following the flash, the data is logged. Check the F_v/F_m value and make sure it is reasonable. Finally, view the flash results.

- 10 Turn on the actinic light and equilibrate.

Under **Environment > Light > Fluorometer**, set the actinic light level to the average mid-day PAR value for your plant. Let the plant adapt to the new light level before going to the next step. To determine when the plant is adapted to the new light level, look for stability in F .

- 11 Tap the **Log** button to do a flash and dark pulse.

The graph should look similar to this (two flashes and dark pulses are represented):

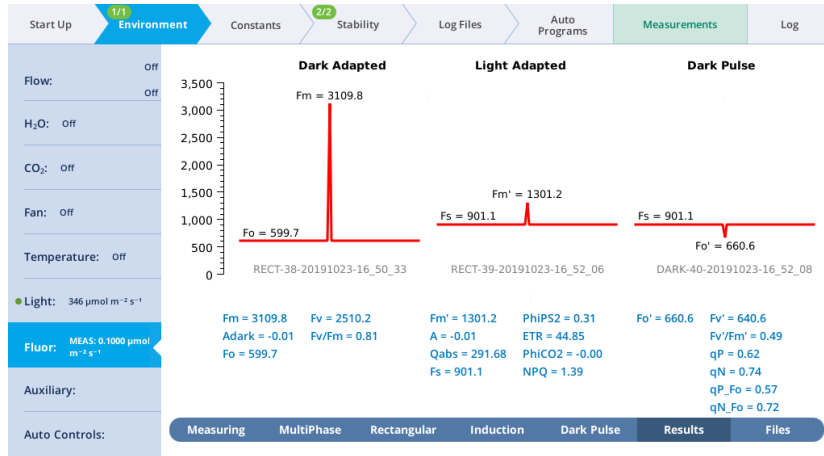


- 12 View the dark pulse details.

Tap **Files** and assess whether the dark pulse settings were appropriate by checking to see if the fluorescence signal levels off before the actinic light is turned on.

- 13 Compare F_v/F_m with and F_v'/F_m' .

Under **Results**, look at the calculated F_v'/F_m' value. F_v'/F_m' should be less than F_v/F_m , since F_v/F_m is the maximum quantum efficiency, and F_v'/F_m' is the effective quantum efficiency at this actinic light level, which is being reduced by the other processes that are competing for light energy.



14 Examine the quenching coefficients.

The quenching coefficients qP and qN are described in *Chlorophyll photophysics* on page C-14. qP and qN range in value between 0 and 1. For a truly dark-adapted plant, $qP=1$ and $qN=0$ at the time F_v'/F_m' is determined. As the actinic light level rises, these coefficients move in opposite directions.

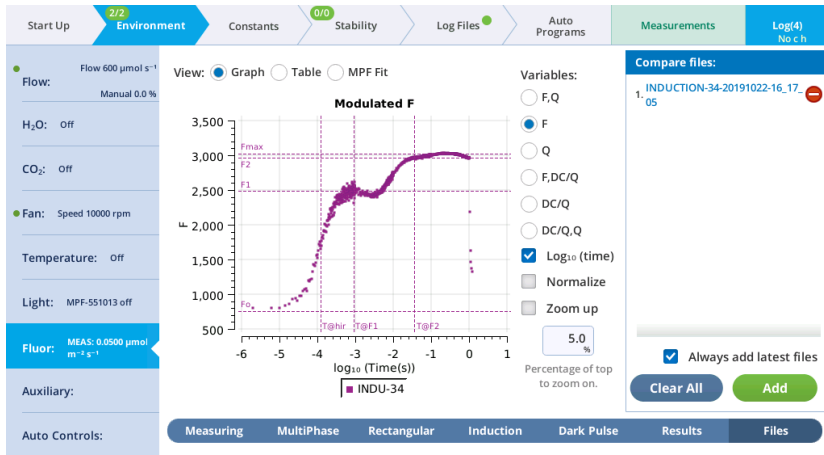
15 Repeat this on additional leaf samples.

Compare the results with leaves adapted to high and low light conditions like in the previous exercise.

An induction kinetics measurement

In this experiment we are interested in collecting the data to plot a chlorophyll fluorescence induction curve (OJIP; origin, inflection, intermediary peak, and peak). You'll need a dark-adapted plant. For purposes of this exercise, you can simply enclose the leaf in the dark chamber for about 20 minutes. For real measurements, it is a good idea to dark-adapt the plants overnight.

- 1 Select a dark-adapted leaf and close the chamber over it.
If your leaf is not dark-adapted, enclose it in the chamber for about 20 minutes. Be sure the chamber light source is off.
- 2 Configure the measuring beam.
Under **Environment > Fluor > Measuring**, set:
 - **Measuring Beam:** On
 - **Dark mod rate:** 500 Hz
 - **Light mod rate:** 1 kHz
 - **Flash mod rate:** 125 kHz
 - **Averaging:** 15 s
- 3 Configure the induction flash.
Under **Environment > Fluor > Induction**, set:
 - **Red target:** 15000 $\mu\text{mol m}^{-2} \text{s}^{-1}$
 - **Duration:** 1000 ms
 - **Margin:** 5 points
- 4 Tap **Flash Now**.
- 5 Upon completion of the flash, tap **Results**.



The results should look like a typical induction curve. It is displayed with a logarithmic time scale.

If you want to initiate the flash from the log button, under **Log Files > Logging Options** set **Flash type** to **Induction** and open a log file. That will cause the flash to

occur when the log button is pressed. The flash data are logged even if you did not open a log file. See *What's in the flevents folder* on page 6-26 for more details.

Note: When you plot these data in a spreadsheet, you'll notice an outlier at the beginning of the dataset (if your margin is 5, the 6th point, which is the first value after the flash begins) and possibly the end of the dataset (the last value during the flash). This is an artifact resulting from the way the modulated AC signal is computed. You can delete the outlier from the dataset. If you view the data on the instrument, we filter the artefactual data point.

Fluorescence and gas exchange experiments

Now we combine gas exchange and fluorescence measurements. Make sure that the IRGAs are zeroed and matched, and that the instrument is ready to measure gas exchange.

Light response curve

In this experiment, we are interested in two parameters: Φ_{PSII} and Φ_{CO_2} (Φ_{PSII} and Φ_{CO_2} henceforth). Φ_{PSII} is the quantum yield of PSII calculated from fluorescence, while Φ_{CO_2} is the quantum yield calculated from CO_2 assimilation. In order to calculate this, we need to know the total assimilation, which comes from measured CO_2 assimilation in the light and an assumption (or prior measurement) of assimilation in the dark. We also need Q_{abs} , which involves knowing incident PAR and leaf absorptivity.

Note: If using a C_3 species for this experiment, it is best to proceed under non-photorespiratory conditions (that is, low oxygen). This can be achieved by attaching a tank of $< 2\%$ oxygen to the LI-6800 inlet. Use an appropriate regulator, T-fitting, and a flow meter to provide adequate flow to the pump and a place for excess flow to be vented. If you do not do this, the measured relationship between Φ_{PSII} and Φ_{CO_2} will likely not be linear. Using a C_4 plant avoids this problem.

In this exercise, we will start with a light-adapted plant and gradually work toward higher quantum efficiencies and yields by decreasing the incident light. We will use the Light Response auto program to accomplish this.

- 1** Select a light-adapted leaf.
- 2** Set up a graph of fluorescence
Under the **Measurements** tab:
 - Select graph **F** to make it easy to remember (F for Fluorescence).
 - Tap **Edit Graphs**.
 - Set Plot: **Time**.
 - Select the variable: Under **FlrLS**, select **F** (demodulated fluorescence).
- 3** Prepare the chamber environment.
Under the **Environment** tab, set:
 - **Flow**: On at 500 $\mu\text{mol s}^{-1}$
 - **CO₂**: Control on sample, around 400 $\mu\text{mol mol}^{-1}$
 - **H₂O**: Set between 50% and 70% RH or VPD_leaf, as described in *Step-by-step light response curve* on page 4-30
 - **Fan**: 10,000 rpm
 - **Temperature**: Setpoint near ambient
 - **Light > Fluorometer**: Control Mode: Setpoint, Setpoint: 2,000 $\mu\text{mol mol}^{-1}$, Color Spec: r90b10.
- 4** Set **Fluorescence** constants.
Under **Constants > Fluorometer Constants**:
 - **Adark**: 0.5 unless you have the value derived from a prior measurement.
 - **Fm, Fo, and PS2/1**: These are populated with data from a prior dark-adapted measurement. This is set automatically if you just did a dark-adapted measurement. If the dark-adapted measurement was not taken immediately prior to the light-adapted measurement, you'll write these values down and enter them manually.
- 5** Open a log file and configure the options:
 - **Matching Options**: Select **Only match if** and select $\Delta\text{CO}_2 < 10.0$ ppm, **Reference change > 100 ppm**, and **Time > 10.0 minutes**.
 - **Fluorometer Options: Flr Action at Log**: Select **1: FoFm (dark) or FsFm' (light)**.
 - **Fluorometer Options: Flash type**: Rectangular.
- 6** Configure the Light Response Auto Program.
 - **Light values**: 2000, 1500, 1000, 500, 200, 100, 50, 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$
 - **Min wait**: 60 seconds

- **Max wait:** 180 seconds
 - Check **Early match**
- 7** Set up graphs.
 - 8** Wait for stability.
 - 9** Start the Auto Program.
 - 10** Watch the curve develop.

A combined induction kinetics and gas exchange measurement

In this experiment we are interested in collecting the data to plot a chlorophyll fluorescence induction curve (OJIP; origin, inflection, intermediary peak, peak). This exercise is similar to , only now you'll measure gas exchange and initiate the measurement with the **Log** button.

- 1** Select a dark-adapted leaf and close the chamber over it.
If your leaf is not dark-adapted, enclose it in the chamber for about 20 minutes. Be sure the chamber light source is off.
- 2** Set up the graph of fluorescence.
- 3** Prepare the chamber environment.
Under the **Environment** tab, set:
 - **Flow:** On at $500 \mu\text{mol s}^{-1}$
 - **H₂O:** Set between 50% and 70% RH or VPD_leaf, as described in *Step-by-step light response curve* on page 4-30
 - **CO₂:** Control on sample, around $400 \mu\text{mol mol}^{-1}$
 - **Fan:** 10,000 rpm
 - **Temperature:** Setpoint near ambient
- 4** Configure the measuring beam.
Under **Environment > Fluor > Measuring**, set:
 - **Measuring Beam:** On
 - **Dark mod rate:** 500 Hz
 - **Light mod rate:** 1 kHz
 - **Flash mod rate:** 125 kHz
 - **Averaging:** 15 s.
- 5** Configure the induction flash parameters.

Under **Environment > Fluor > Induction**, set:

- **Red target:** 15,000 $\mu\text{mol m}^{-2} \text{s}^{-1}$
- **Duration:** 1000 ms
- **Margin:** 5 points

6 Open a log file and configure it.

Under the **Log Files** tab:

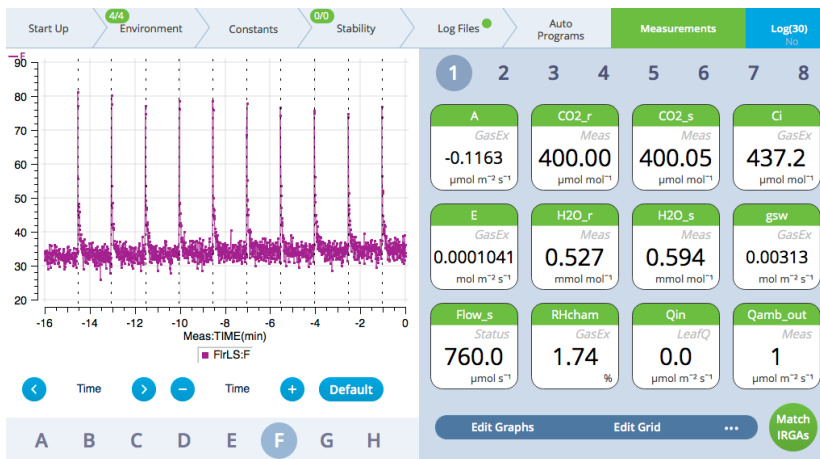
- **Matching Options:** Select **Only match if** and select $\Delta\text{CO}_2 < 10.0$ ppm, **Reference change** >100 ppm, and **Time** > 10.0 minutes.
- **Fluorometer Options: Flr Action at Log:** Select **1: FoFm (dark)** or **FsFm' (light)**.
- **Fluorometer Options: Flash type:** Rectangular.

7 Set up an Auto Program.

The program called **AutoLog** can be configured to log measurements at repeated intervals. Configure it for a 90 second log interval and a total time of 900 seconds (15 minutes).

8 When both you and the plant are ready, **Start** the Auto Program.

The instrument will do the flashes while recording gas exchange data.



Configuring auxiliary channels

The LI-6800 25-pin connector accepts a male 25-pin D-connector, which can be purchased from many electronics stores or directly from LI-COR. Pin assignments are described in *Table 4-1* on the facing page. Raw voltages can be logged directly or

transformed into more meaningful values using basic equations. This is described in *User constants* on page 3-76.

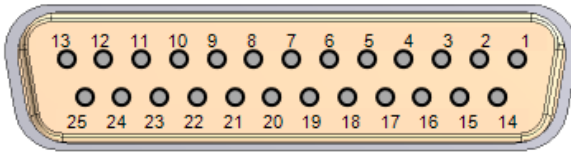


Figure 4-19. The 25-pin connector is labeled **USER I/O**.

Logging the auxiliary channels

Logging of auxiliary channels is controlled under the **Log Files > Logging Options** tab. Under the **Include in log file** option, select **Auxiliary**. When you select auxiliary, all measured data and settings will be saved in the log file. Auxiliary data will be logged in the same row as the other data when you press the log button or when a log is initiated through an **Auto Program**.

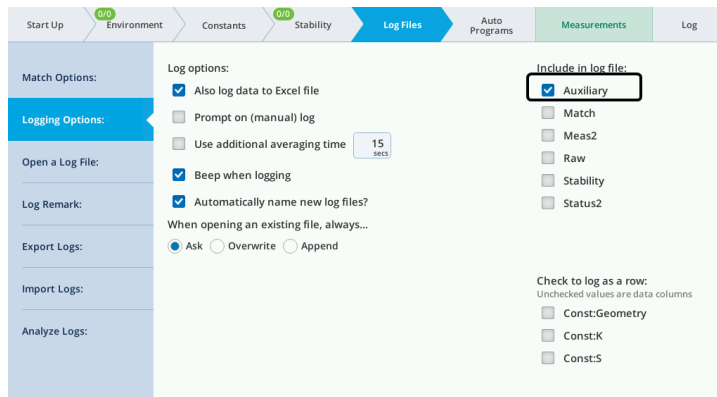


Figure 4-20. Select **Auxiliary** to log values from the 25-pin connector.

Table 4-1. Pin assignments for the 25-pin connector.

Pin #	Label	Description	Range	Resolution	Sampling Rate
1	GND	Ground	n/a	n/a	n/a
2	DAC1	User-programmable	± 5 V	16 bit	n/a
3	DAC2	analog output channels		(152.6	
4	DAC3		$\mu\text{V}/\text{count}$)		
5	DAC4				

Table 4-1. Pin assignments for the 25-pin connector. (...continued)

Pin #	Label	Description	Range	Resolution	Sampling Rate
6	I/O-1	General Purpose Inputs and Outputs (GPIO); <ul style="list-style-type: none"> Input voltage must be ≥ 3.5 V (maximum 5.5 V) for logic 1 and ≤ 1.5 V (minimum -0.5 V) for logic 0. Output (≤ 10 mA) voltage must be ≥ 4.2 V (maximum 5 V) for logic 1 and ≤ 0.5 V for logic 0. 			
7	I/O-2				
8	I/O-3				
9	I/O-4				
10	I/O-5				
11	I/O-6				
12	I/O-7				
13	I/O-8				
14	User5	5 V when on; 0 V when off; Maximum 500 mA; electronic fuse			
15	ADC_CH7	User-programmable	± 5 V	16 bit	10 Hz
16	ADC_CH8	analog-to-digital input		(152.6 μ V	
17	ADC_CH5	channels		per count)	
18	ADC_CH6				
19	ADC_CH3				
20	ADC_CH4				
21	ADC_CH1				
22	ADC_CH2				
23	User12	12V when on; 0V when off; Maximum 1 A, electronic fuse			
24	A-GND	Analog ground			
25	Excite5	20 mA maximum			

In addition to the inputs and outputs, the interface allows you to activate a 10 kilo-ohm 0.1% pullup resistor for 5 V biasing of a thermistor on ADC_CH1 (pin 21). The **AuxPower** feature is not supported yet.

Analog input channels

The eight ADC (analog-to-digital channel) inputs can read from ± 5 volts at 16 bit resolution with a sampling rate of 10 Hz.

To wire an analog input, select a signal pin (pins 15 to 22) and a ground pin (pin 1 or 24). The sampling rate is fixed at 10 Hz, and thus the resolution is 152.6 μ volts per count.

Analog output channels

The analog output channels (DAC1 to DAC4) can be set to a fixed voltage in the **Environment > Auxiliary** tab, or set to a shape (linear, step, sine/4, or sine/2) in **Auto**

Controls and Auto Programs. Output voltages range from -5 to +5 V. To set a fixed voltage, simply enter that value.

If you want to set a controlled parameter to track an auxiliary channel, see *Auto controls: The LI-6800 abides* on page 3-71.

Digital outputs and inputs

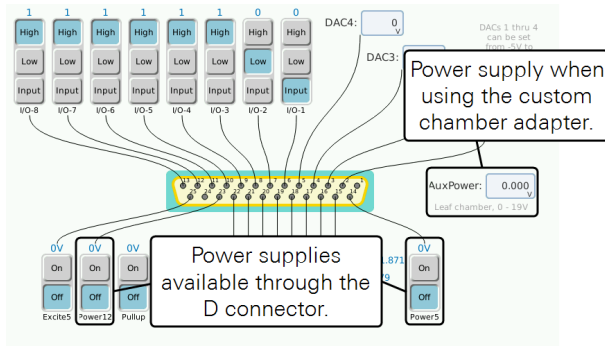
Eight channels are available for use as digital inputs or outputs.

Tap **High** to output 5 V and **Low** to output 0 V through the corresponding pin. When used as a digital input, the voltage must be >3.5 V for **logic 1** and <1.5 V for

logic 0. The logic signals (pins 6 to 13) provide a very low current. Do not use them to power a device, since that could damage the circuitry.

Auxiliary power supplies

One 12 VDC and one 5 VDC power supplies are available through the connector. **AuxPower** is accessible when using the custom chamber adapter. The custom chamber adapter is forthcoming.

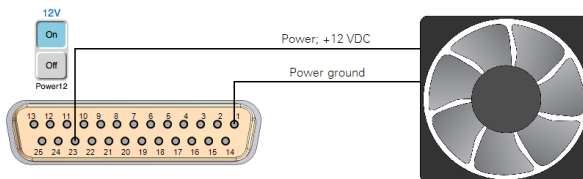


Mixing fan example

Here we'll describe how to connect an external mixing fan. The mixing fan may be used, for example, to ensure well-mixed air in a large custom chamber.

You'll want a small fan, like a fan used to cool a computer processor. It should be rated for operating at 12 VDC if you use pin 23 or 5 VDC if you use pin 14.

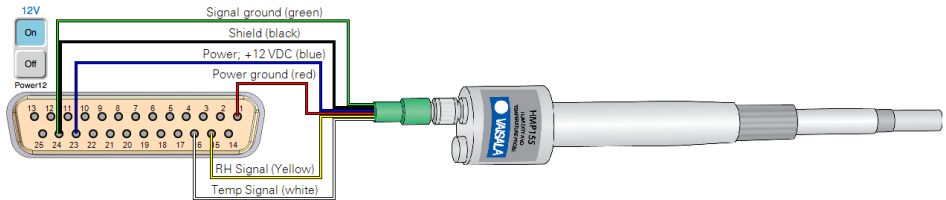
- 1 Connect the red wire to pin 23 using a male 25-pin D connector.
- 2 Connect the ground wire to pin 1.



- 3 Turn on the power. Under **Environment > Auxiliary**, tap **On** for **Power 12**. The fan will turn on. Tap it again to turn the fan off.

Humidity and temperature sensor example

Here we show how to connect a Vaisala HMP155 humidity and temperature probe. Raw voltages can be logged directly or transformed into more meaningful values using basic equations.

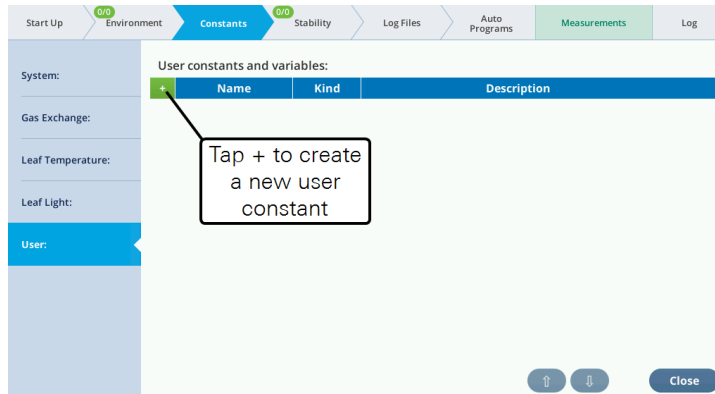


Be sure that the 12V power supply is on; and log the auxiliary channels in the data file (see *Logging the auxiliary channels* on page 4-47).

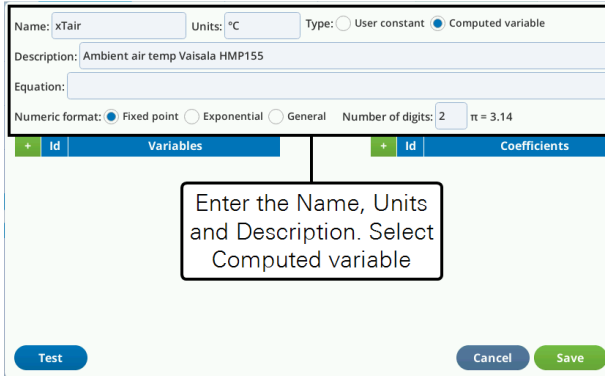
Configuring the auxiliary channels

The HMP155 outputs a voltage signal from 0 to 1 volt over a -80 to 60 °C temperature range and for 0 to 100% RH. You'll configure an equation with a slope and offset for temperature and an equation with a multiplier for RH. Start by creating a new **User Constant** for temperature.

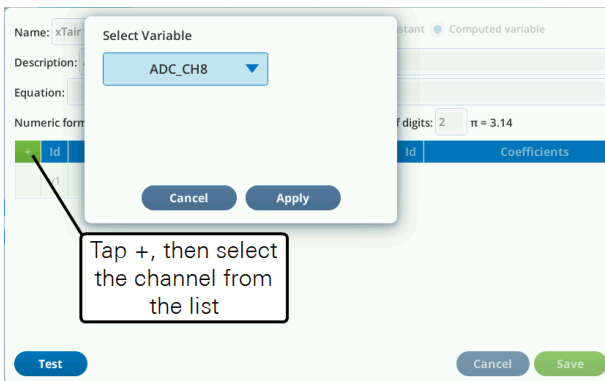
- 1 Under **Constants > User**, tap the **+** button.



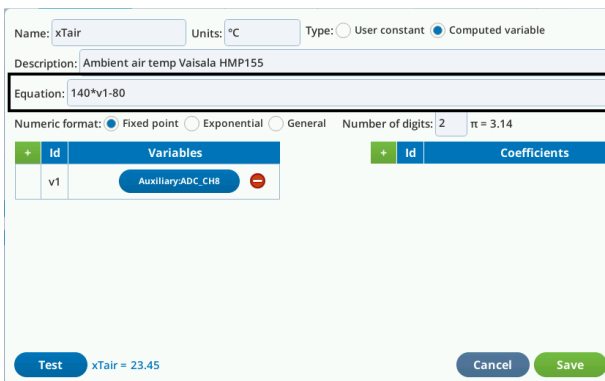
- 2 Enter the **Name**, **Units**, and **Description**. Select **Computed variable**.



3 Before entering the Equation, you should create the Variables and optionally, Coefficients.



4 The equation for temperature in °C is $xTair = 140 * v1 - 80$, so enter the equation.



5 Tap Test to see the results. If you're satisfied, tap Save.

- 6 Create a new user constant for RH.
- 7 Enter the parameters for the new constants

The equation for RH% is $RH\% = v1 * 100$.

Name: RH Units: % Type: User constant Computed variable

Description: RH Vaisala HMP155

Equation: v1*100

Numeric format: Fixed point Exponential General Number of digits: 2 $\pi = 3.14$

Variables		Coefficients	
+	Id	+	Id
	v1		

Auxiliary:ADC_CH7

Buttons: Test, Cancel, Save

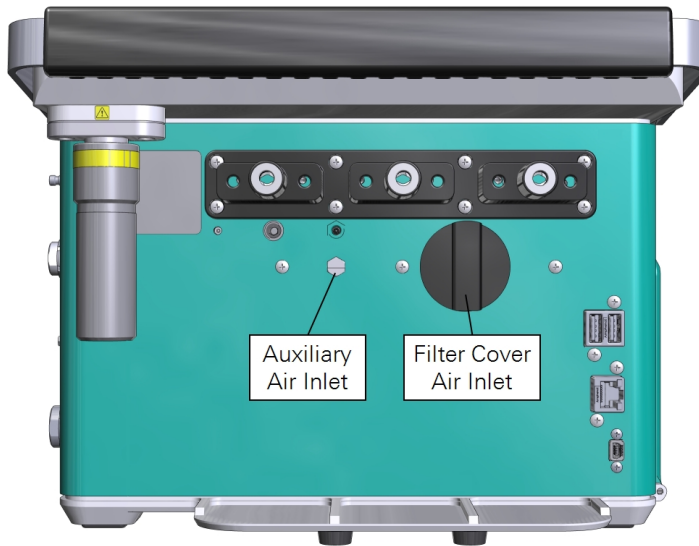
- 8 Tap **Save** if you're satisfied with the configuration.

The auxiliary channels are available for use in **Auto Programs** and graphs. Be sure to log the auxiliary channels with your dataset (see *Logging the auxiliary channels* on page 4-47).

Introducing a custom air supply

Two air inlets are available to introduce your own conditioned air supply if you do not want to start with ambient air. The difference between the inlets is:

- The **Auxiliary Air Inlet** between the **H₂O Scrub** and **H₂O Add** columns on the console, is plumbed downstream of the pump and the CO₂ scrub column (see *Figure 1-2* on page 1-4).
- The **Filter Cover Air Inlet** is a threaded connection on the air filter cover between the **H₂O Add** and **CO₂ Scrub** columns.



Using the auxiliary air inlet

To use the **Auxiliary Air Inlet**, attach an air supply. There is no pump so the connected source will need to provide the correct pressure and flow rate. There is no filter in the air stream because this inlet is downstream of the filter. Use clean air or provide your own filter to keep dust out of the plumbing.

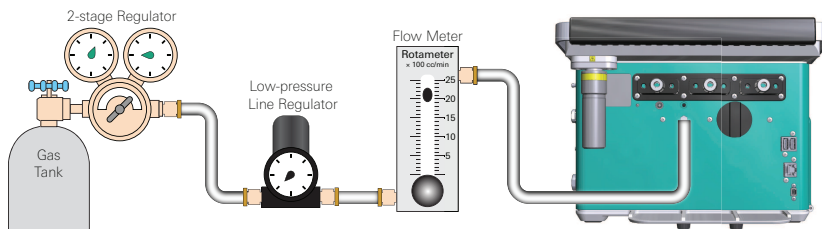
- 1 Remove the plug and set it aside for safekeeping.
- 2 Install a hose barb in the **Auxiliary Air Inlet**.
- 3 Connect the air supply and pressurize the system.

The threads are M5 (metric units) or approximately 10-32 (imperial units). The maximum flow rate is 2.5 lpm and maximum pressure is 28 kPa. The LI-6800 pump will not affect the flow rate because the air inlet is downstream of the pump.



Warning: Do not exceed the ratings on the auxiliary air inlet. Maximum flow rate of 2.5 liters per minute. Maximum pressure of 28 kPa.

Avertissement: Ne dépassez pas les indices sur l'arrivée d'air auxiliaire. Vitesse maximale d'écoulement de 2.5 litres par minute. Pression maximale de 28 kPa.

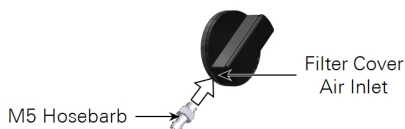


Using the filter cover air inlet

The filter cover air inlet is threaded into the air inlet cap. This inlet is upstream of the pump and CO₂ scrub column, so air that is provided this way can be scrubbed of CO₂ and pumped by the LI-6800 pump.

- 1 Install a hosebarb on the air inlet.

Assemble a t-fitting with one tube coming from the air supply, one tube going to the air inlet, and the other tube venting to the ambient air (see *Figure 4-21* on the next page). Put about 10 to 15 cm of tubing on the vent. This will prevent ambient air from diffusing up the vent, which can happen at low flow rates.



- 2 Install the air inlet on the console, then reinstall the columns.
- 3 Turn on the air supply.
Your air supply should have 2.5 to 3 lpm of flow.
- 4 Use the LI-6800 pump to control the flow rate.

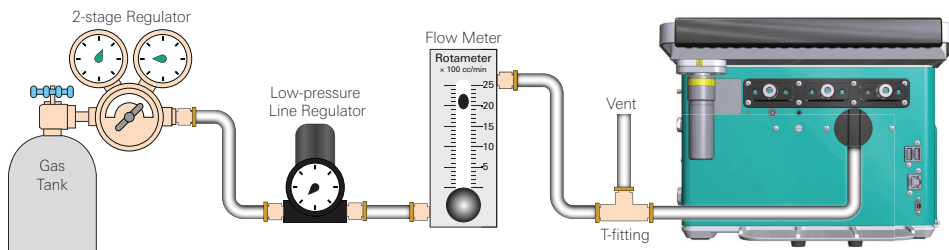


Figure 4-21. The filter cover air inlet can be outfit with an M5 (metric) or 10-32 (imperial) hosebarb. The incoming air should be directed through a t-fitting, with one end of the fitting vented to the air, and the flow should be sufficiently high to prevent the pump from drawing a vacuum.

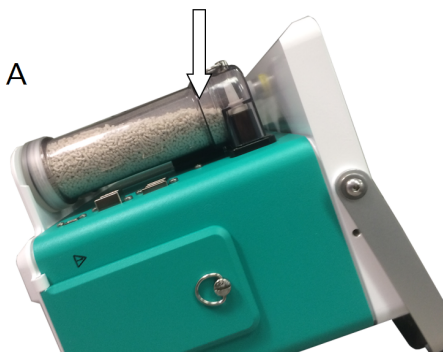
Tips for success

This section includes tips that will help you be successful with the LI-6800.

Keep the console vertical

This will help ensure that the air flows through the gas conditioning chemicals. When the console is tilted, the chemicals tend to "flow" to one side, creating passages through the tubes, limiting the air's exposure to the chemicals. This means that it takes longer to achieve a CO₂ or H₂O setpoint, or even that you can't achieve the setpoint. Additionally, measurements of A , E , g_s , and C_i will be more variable because the incoming CO₂ and H₂O concentrations will be varying.

If the chemicals have gaps like this, the instrument will not achieve setpoints.



Completely fill the columns to minimize gaps, but this has its own drawbacks.

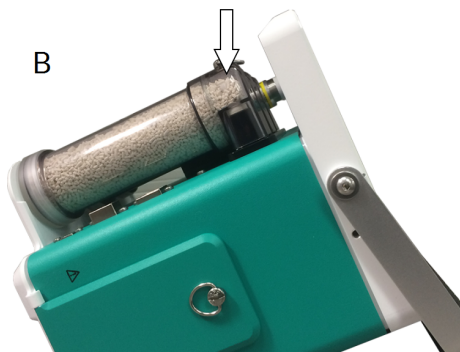


Figure 4-22. Notice that the predominant pathway for air is not through the chemicals (A). This is undesirable because air is not exposed to as much chemical. You can fill the columns completely to prevent gaps (B). The downside of this is that the chemicals may appear saturated more quickly, and it is harder to break up channeling by shaking the column.

If you must tilt the instrument for any reason, fill the columns all the way full of their respective compounds. This makes it harder for the compounds to shift when tilted, but also makes it harder to break up channeling with a gentle shake. So, it is a tradeoff.

Pay careful attention to the warnings and the time it takes to achieve the setpoint. If things are taking longer than expected it might be time to shake up the columns or replenish the chemicals.

Extend your soda lime

You can extend the life of soda lime by adding 5 to 10 ml water. Gently shake the column to distribute the water in the soda lime.

Lighten your load

If you want to make the instrument lighter, install one battery and keep the other one handy. The system will operate normally with one battery, but you'll need to insert the second battery before removing the first.

Controlling the LI-6800 from a computer

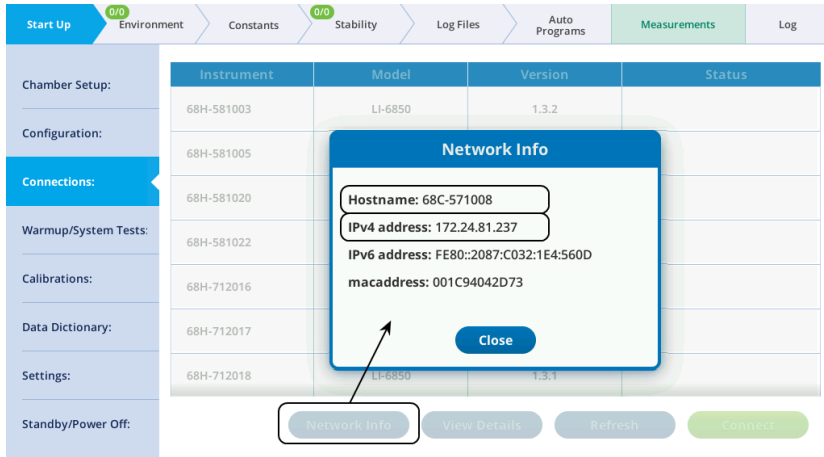
The LI-6800 supports remote access via Virtual Network Computing (VNC) with software version 1.1.5 and newer. This allows you to view and control the console from a computer, tablet, or smart phone. The LI-6800 does not need to be in any special mode of operation—you can connect and disconnect at will. There are two requirements:

- Install a VNC client on your computer, mobile device, or tablet. For example: <http://www.realvnc.com/en/connect/download/viewer>
- Your computer or mobile device needs to be on the *same network* as the LI-6800. You can connect a computer directly to the LI-6800 with a network cable or connect the LI-6800 to an open network port.

Each VNC client may require slightly different steps to make it communicate with the LI-6800. To connect:

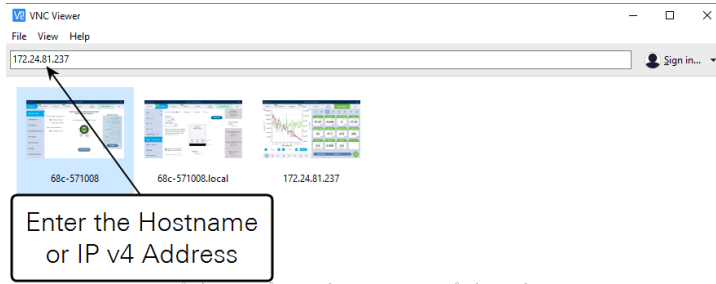
- 1 Run the VNC client.
- 2 Enter the IPv4 address or hostname of the LI-6800 console.

Tap **Start Up > Connections > Network Info** to retrieve this information. Or read the label on the back of the instrument.



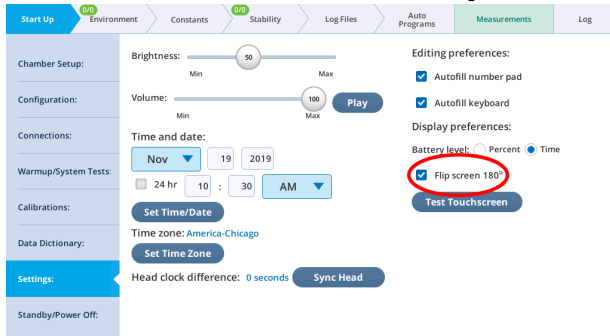
Depending on the network configuration, you may need to enter the hostname or IP address in one of the following formats:

- **172.24.81.237** The IPv4 address for your instrument will probably be different.
- **68C-#####** The serial number (also called the Hostname) will be unique for your instrument.
- **68C-#####.local**. The serial number will be unique to your instrument.



Notes:

- The LI-6800 display will be flipped 180 degrees from how it is normally viewed on the console. Here's how to flip it:



- Only one remote viewer can be connected at a time to an LI-6800. A new connection will end an existing connection.

Section 5.

Leaf chambers and accessories

Big leaf, small leaf, narrow leaf, tall leaf

Removing a chamber	5-2	Installing the chamber	5-47
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Leaf chambers and accessories

The LI-6800 is available with an assortment of chambers and accessories for custom chambers. In this section, we describe many of these chambers and provide guidance for using them. For additional application and technical notes, see the LI-COR technical support web site: www.licor.com/env/support.

Removing a chamber

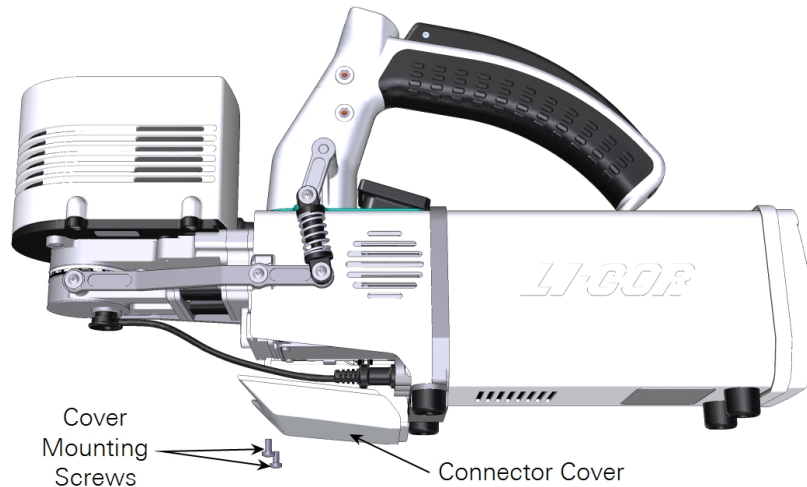
Switching from one chamber to another is straightforward; every chamber connects to the head in the same way. Chambers store information about themselves, so when connected, the head recognizes the chamber and automatically sets the volume and any required calibration constants.

Detach the chamber from the head

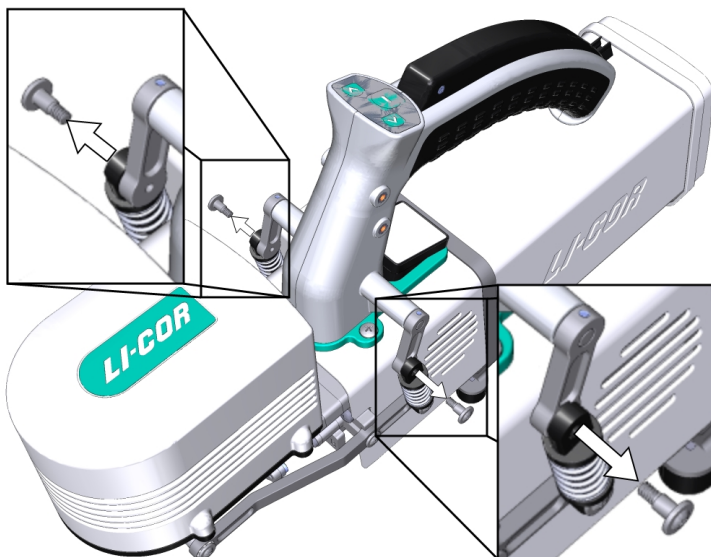
Six screws will be removed to detach a chamber — two on the connector cover, two on the latch mechanism, and two that hold the chamber to the head.

Caution: Power off the instrument before following these procedures.

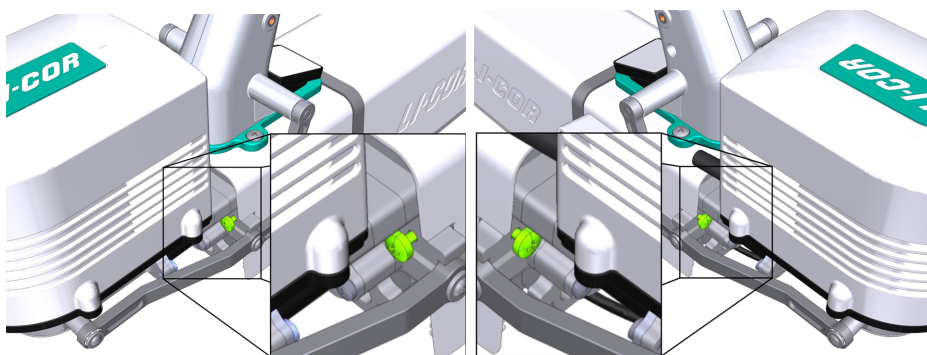
- 1 Remove the two cover screws and the connector cover from the bottom of the head.



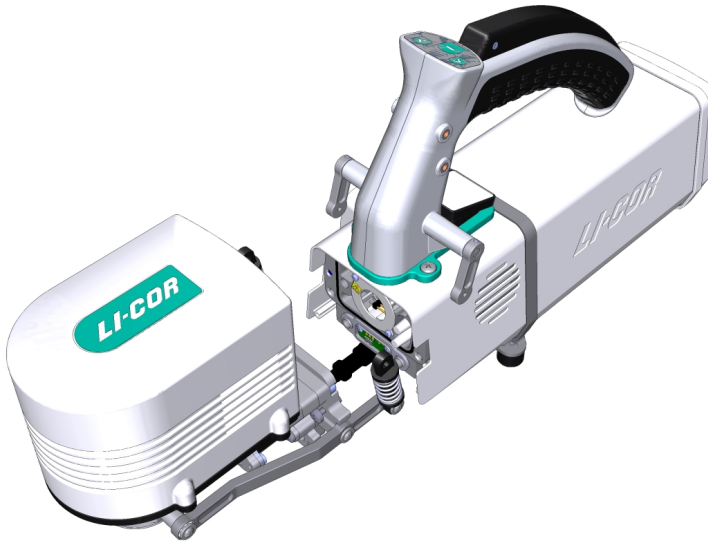
- 2 Unplug the cables from the cable bulkhead (i.e., LI-190R, leaf temperature thermocouple, and light source).
- 3 Position the chamber latch in the parked position and remove the two latch screws.



- 4 Loosen both of the captive chamber screws until the chamber is free of the head.



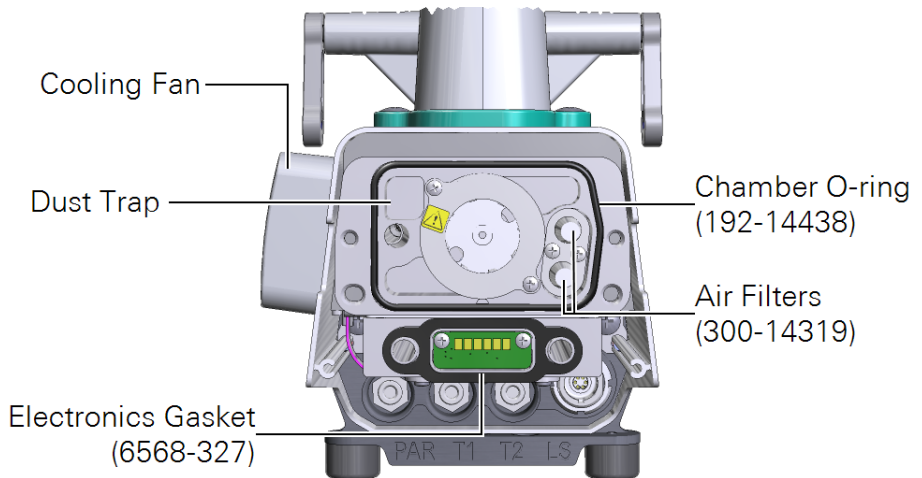
- 5 Separate the chamber from the head.



Warning: Do not attempt to operate the mixing fan without a chamber in place.

Avertissement: N'essayez pas de faire fonctionner le ventilateur mélangeur sans une chambre en place.

While the chamber is removed from the head, inspect the **Air Filters** and **Dust Trap**. Clear any dust or debris before attaching a chamber.



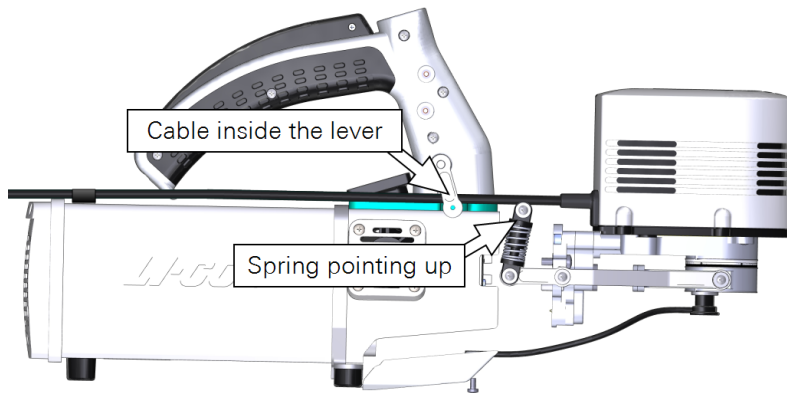
MultiPhase Flash™ Fluorometer

The 6800-01A fluorometer features removable chamber apertures with 6 cm² and 2 cm² areas.

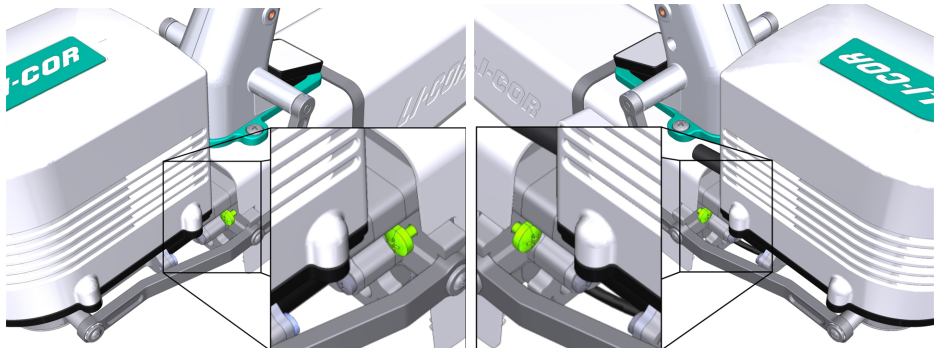
Installing the fluorometer chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

- 1 Hold the fluorometer chamber in position, with the spring that is closest to the cooling fan facing up toward the handle and the cable inside the lever, and tighten the two captive chamber screws.



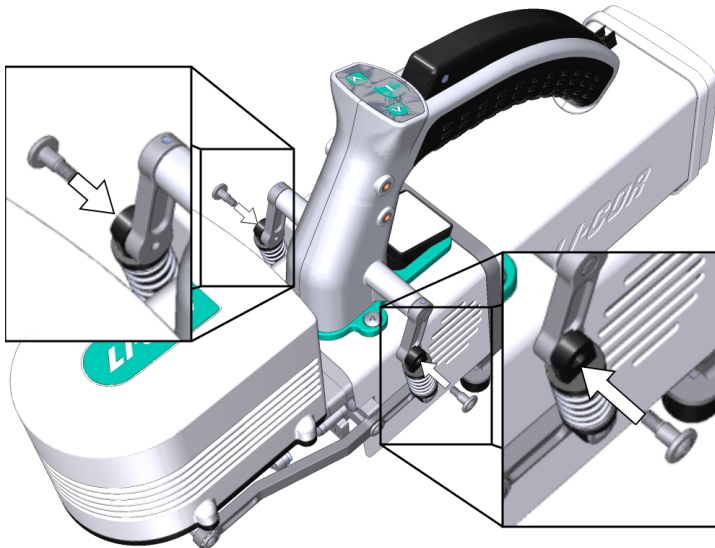
Make them snug—turn each screw until it stops, and then about ¼ turn more. If you have a torque screwdriver, tighten them to 2.5 N·m (22 inch lbs).



Caution: You must use the captive chamber screws (part number 144-15344) to install the chamber. Other screws will cause leaks and problems with the mixing fan, even if they seem to fit.

2 Install the **chamber latch screws**.

With the latch in the parked position, tighten the screws (part number 146-14474) until they are snug. If you have a torque screwdriver, tighten them to 1.1 N·m (9.4 inch lbs). Double check the tightness of each screw.

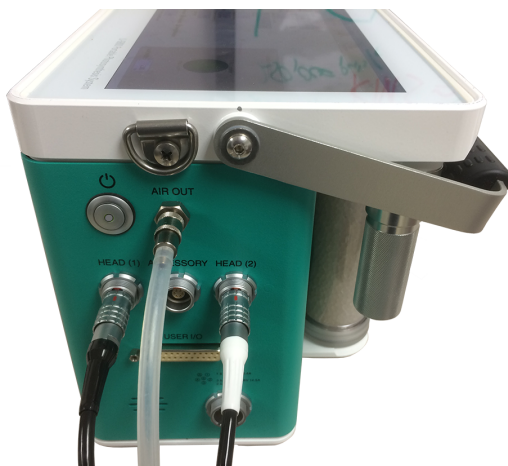


3 If you unplugged the light sensor cable, install the light sensor connector to the connection labeled PAR.

4 Connect the leaf temperature thermocouple cable to the connection labeled T1 or T2. Install the connector cover.

5 Plug the Fluorometer cable connector into the console.

The fluorometer cable (with the white strain relief) and head cable can be plugged into either the **Head (1)** or **Head (2)** connector. The connectors are interchangeable.



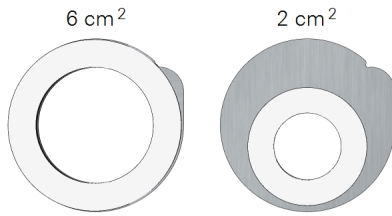
- 6** Slip the cable into the head cable bundle and press the cable into the clasp.



- 7** Power the instrument back on.
- 8** Check to see that the chamber is recognized under **Constants > System Constants > ChType**.
- 9** Run the leak test under **Start up > System Tests > Chamber Leak**.

Using apertures

The 6800-01A fluorometer includes removable chamber apertures that allow you to reduce the leaf area. Two aperture sets are available, providing areas of 6 cm² and 2 cm². The inside surfaces of the aperture are coated with a black light-absorbing surface. Do not damage the coating.



White advanced polymer gaskets are installed on the chamber prior to shipping. Foam gaskets are included in the accessories kit (black and white). The white gaskets are for the upper aperture; the black gaskets are for the lower aperture.

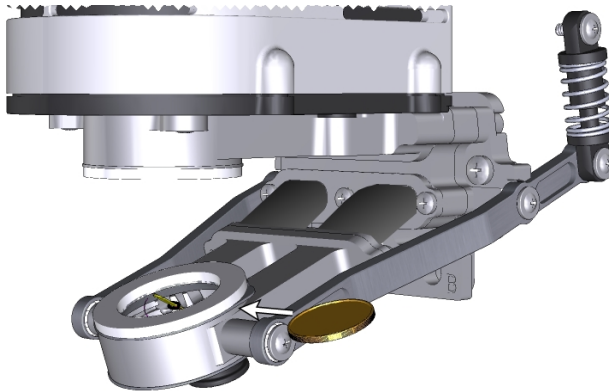
Points to remember

- Top and bottom apertures must be the same size. For example, do not use the 6 cm² upper aperture with the 2 cm² lower aperture.
- When you install apertures, you'll need to select the correct aperture in the software in order to use the correct leaf area and other parameters in computations. You can correct the leaf area and recompute your data later, but it is always a good idea to get the settings right from the outset.

Swapping apertures

Apertures press into the chamber and are held in place by friction from the O-ring.

- 1 Remove both upper and lower apertures.
Use a coin to pop out each aperture.

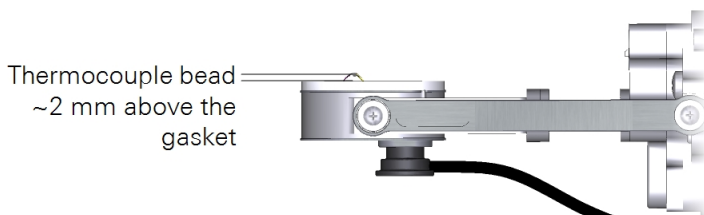


2 Install the apertures.

Align the key in the aperture with the chamber opening and press it into place. If using foam gaskets, the aperture with the white gasket goes into the upper chamber; the aperture with the black gasket goes into the lower chamber.

3 Install the leaf temperature thermocouple.

The tip of the thermocouple should make contact with a leaf in the chamber and should not be in contact with the aperture.



4 Plug the thermocouple connector into the T1 or T2 connector on the head.

5 Power on the LI-6800 and let it warm up.

6 Select the aperture size.

Even though the chamber itself is recognized by the LI-6800, the apertures are not. Under **Start Up > Chambers**, you'll see options for the chamber aperture. Select the aperture size of 6 cm² or 2 cm².

Start Up 2/2 Environment Constants 0/3 Stability Log Files Auto Programs Measurements Log

Chamber Setup:

- Configuration:
- Connections:
- Warmup/System Tests:
- Calibrations:
- Data Dictionary:
- System Settings:
- Standby/Power Off:

Options for the Attached Chamber 6800-01A Fluorometer

Which aperture is installed?

6 cm²
 2 cm²

Fluorometer functions as:

Full Fluorometer
 Light Source Only

What computations?

Complete Gas Ex

Fluorometer MPF-551000 is active

Chamber Info

id: 6800-01A
sn: MPF-551000
rev: 1
ppfd cal: 0
vol: 81
pca: 4.3321e-10
pcb: 1.0215e-05

Lock

Caution: values stored in chamber memory.

Load Factory

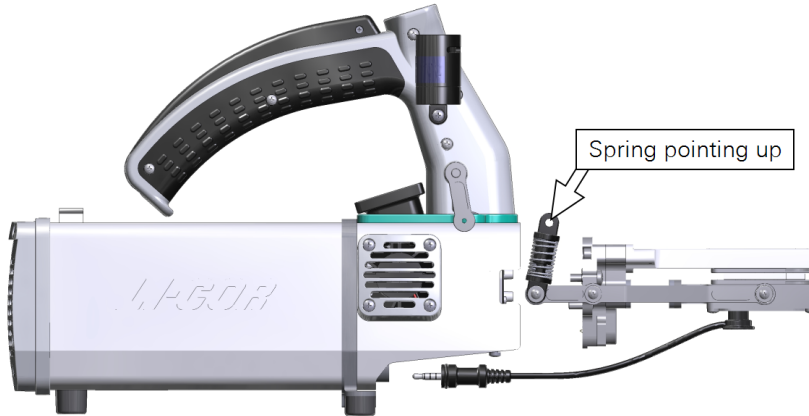
Clear-top small leaf chamber (3×3 cm)

The clear-top chamber is used to measure small leaves. The 6800-12A model features removable aperture inserts that reduce the area of leaf in the chamber. The apertures are 3×3 cm, 2×3 cm, and 1×3 cm. The 2×3 cm and 1×3 cm apertures can be installed with the opening side-to-side or front-to-back. Here we describe how to use the chamber and apertures.

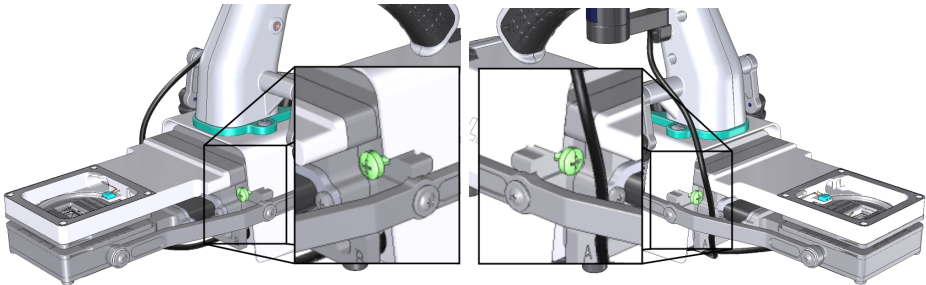
Installing the clear-top leaf chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

- 1 Hold the chamber in position, *with the spring closest to the cooling fan facing up toward the handle*, and tighten the two captive chamber mounting screws (144-15344).



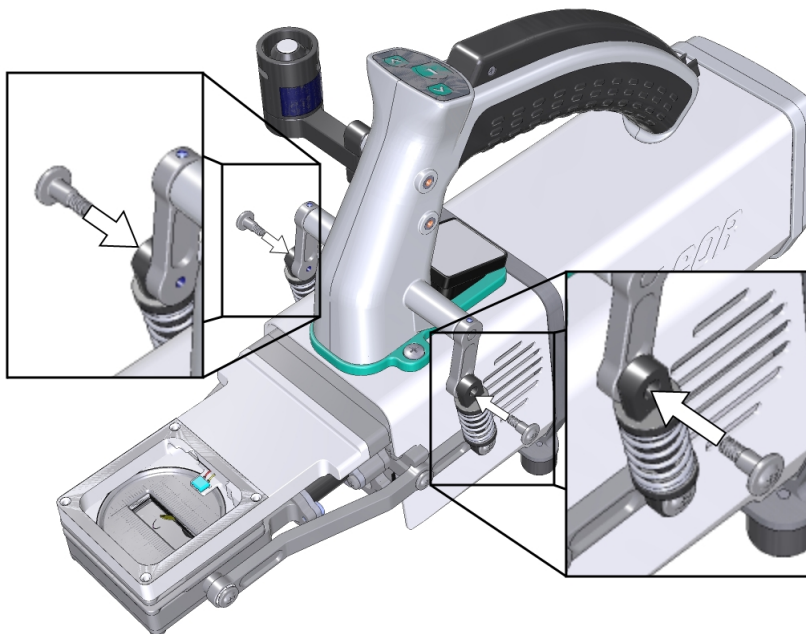
Caution: Use the captive chamber mounting screws to install the chamber. Other screws will cause leaks and problems with the mixing fan.



Make them snug—turn each screw until it stops, and then just a little bit more (up to $\frac{1}{4}$ turn). If you have a torque screwdriver, tighten them to 2.5 N·m (22 inch lbs).

2 Install the latch screws.

With the latch in the parked position, tighten the screws until they are snug. If you have a torque screwdriver, tighten them to 1.1 N·m (9.4 inch lbs). Double check the tightness of each screw.



3 Install the light sensor connector to the connection labeled PAR.

4 Install the leaf chamber apertures and the corresponding chamber bottom plate.

5 Install the leaf temperature thermocouple and attach the connector to the connection labeled T1.

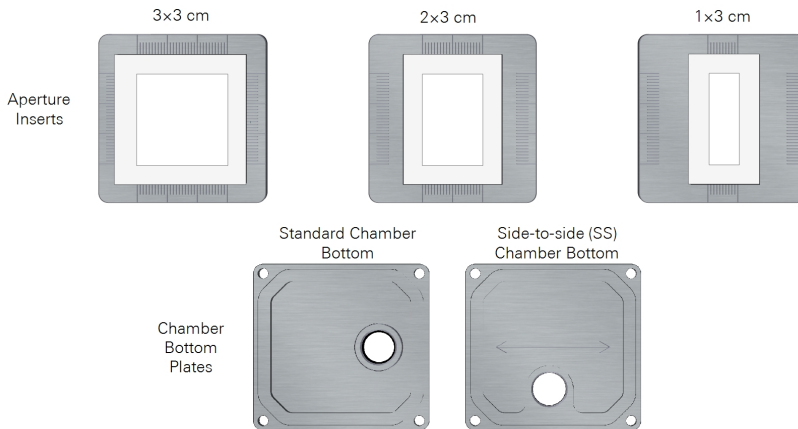
6 Check the thermocouple height and adjust if necessary.

7 Install the connector cover.

After swapping chambers or apertures, it is a good idea to run the leak test (under **Start up > System Tests > Chamber Leak**). Check to see that the chamber is recognized under **Constants > System Constants > ChType**.

Using apertures

The clear-top chamber (6800-12A) includes removable chamber apertures that allow you to reduce the exposed leaf area chamber. Three sizes of apertures are included. There are two chamber bottom plates that allow you to position the leaf temperature thermocouple correctly.



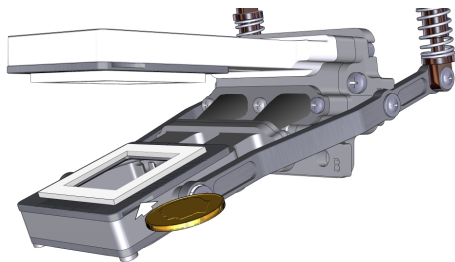
Points to remember

- Top and bottom chambers must be the same size and have the same orientation. For example, do not use the 2×3 cm upper aperture with a 1×3 cm lower aperture or a 2×3 cm upper chamber oriented front-to-back (FB) to the head and a 2×3 cm lower chamber oriented side-to-side (SS) to the head.
- With the 1×3 cm aperture opening oriented side-to-side, use the side-to-side chamber bottom plate. This bottom plate positions the leaf thermocouple so it makes contact with the leaf.
- When you install apertures, select the correct aperture in the software in order to use the correct leaf area and other parameters in computations. You can correct the leaf area and recompute your data later, but it is always a good idea to get the settings right from the outset.

Swapping apertures

Apertures press into the chamber and are held in place by the O-ring.

- 1 Remove both upper and lower apertures.
Insert a coin into the slot and pop the aperture out.



- 2 Align the desired apertures and press each one firmly into the chamber.
If using foam gaskets, the white gasket goes into the upper chamber and the black gasket goes into the lower chamber.
- 3 If you are using the 1×3 cm aperture aligned side-to-side to the head, install the perpendicular chamber bottom plate as shown in *Figure 5-1* belowB.

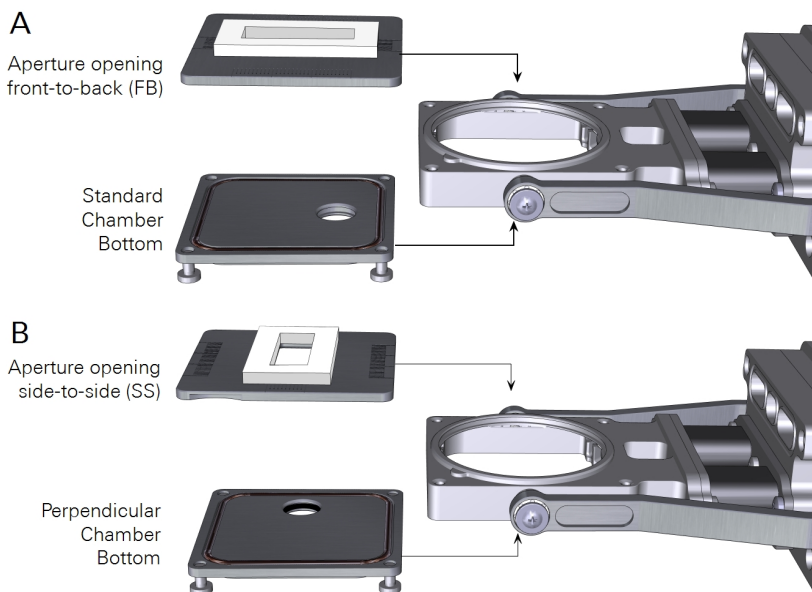


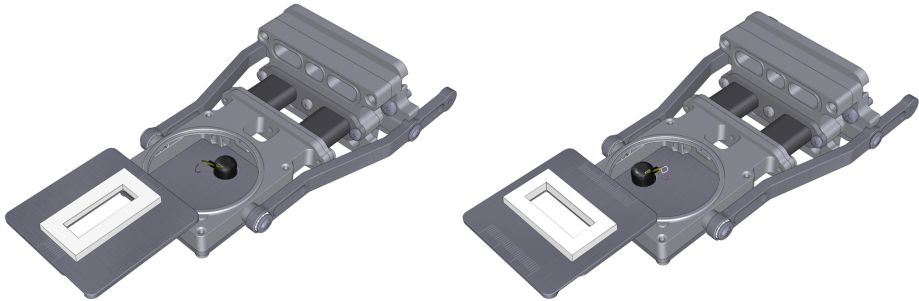
Figure 5-1. The 1×3 cm (shown) or 2×3 cm aperture use the standard chamber bottom (A) if the aperture opening is front-to-back (FB) and the perpendicular chamber bottom (B) if the aperture opening is side-to-side (SS). Upper chamber aperture not shown for clarity.

- 4 Install the leaf temperature thermocouple.

Thermocouple bead
~2 mm above the
gasket

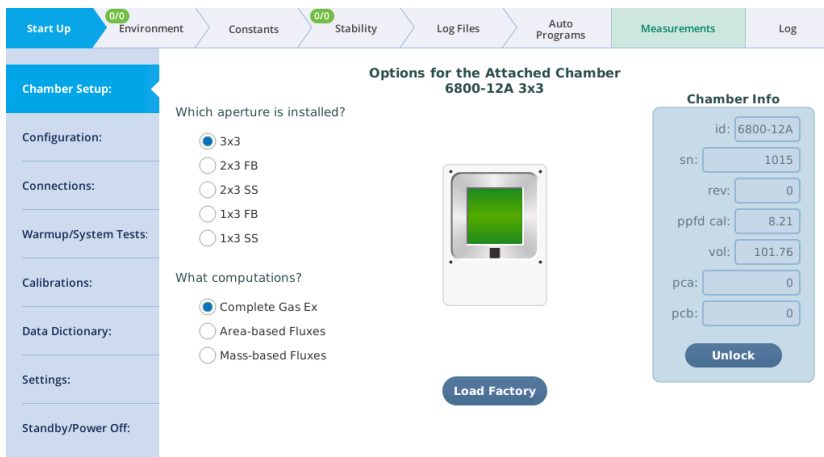


Adjust the height and orientation as needed to maintain good contact with the leaf. 2 mm above the gasket is ideal for most leaves.



- 5 Plug in the temperature connector into the T1 or T2 connector on the head.
- 6 Power on the LI-6800 and let it warm up.
- 7 Select the aperture size and orientation.

Even though the chamber itself is recognized by the LI-6800, you will have to specify which aperture is in use. Under **Start Up > Chambers**, select the aperture and its orientation.



The images on the display will help you understand which setting is which.

- 3×3: Standard aperture
- 2×3 FB: 2×3 cm aperture oriented front-to-back
- 2×3 SS: 2×3 cm aperture oriented side-to-side
- 1×3 FB: 1×3 cm aperture oriented front-to-back
- 1×3 SS: 1×3 cm aperture oriented side-to-side

Connecting the 3×3 light source

The 3×3 cm light source attaches directly on top of the 3×3 cm Clear-top Chamber.



Warning: *The light sources for this product can emit potentially hazardous optical radiation (RG-2 CAUTION POSSIBLY HAZARDOUS OPTICAL RADIATION EMITTED FROM THIS PRODUCT), in excess of the Exempt Risk Group. Potential risk depends upon how one uses and installs this product. Do not operate the light source while detached from the chamber. **Do not look directly into the light source under any circumstances.** Operate the light sources with direct access to ambient air for cooling. Do not use the product in any manner not described in the manual.*

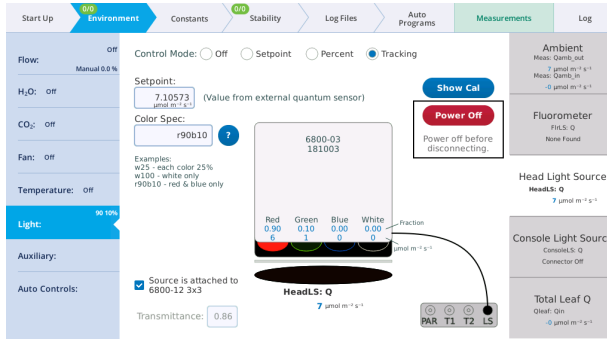


Avertissement: *Les sources de lumière pour ce produit peuvent émettre des radiations optiques potentiellement dangereuses, en excès à l'Exempt Risk Group- Groupe Exempt de Risque. Les risques potentiels dépendent de la façon dont on utilise et installe ce produit. N'opérez pas la source de lumière quand elle est détachée de la chambre. Ne regardez pas directement la source de lumière dans n'importe quelle circonstance. Opérez les souques de lumière avec accès direct à l'air ambiant pour le refroidissement. N'utilisez pas ce produit de n'importe quelle manière non décrite dans ce manuel.*

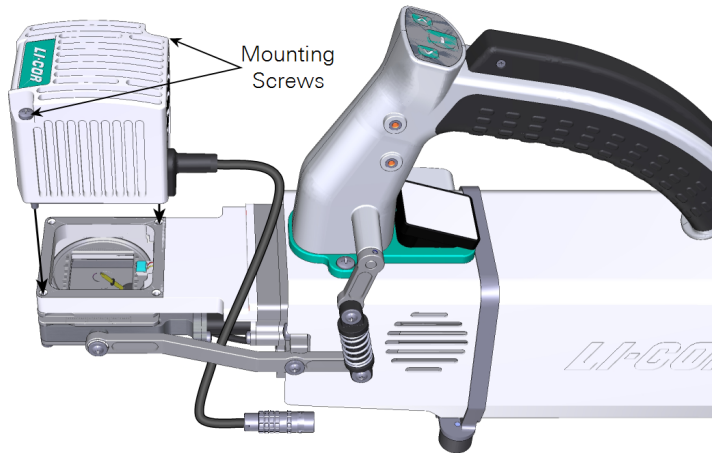
In order to use the light source, the head must have the corresponding chamber installed.

- 1 Prepare the instrument to accept the light source.

Go to **Environment > Light > Head Light Source** and tap **Power Off**. Or, just power off the instrument.

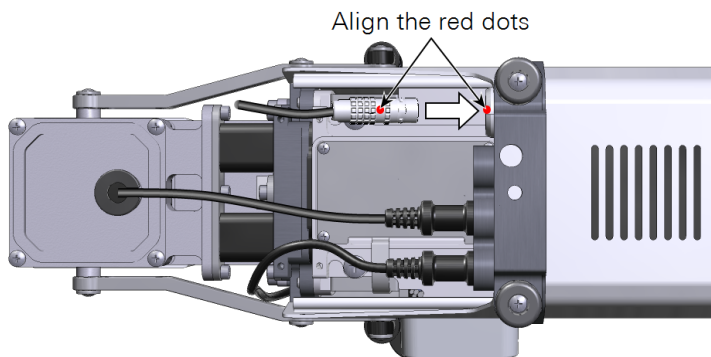


- 2 Check the chamber top Propafilm for dirt or damage. Clean or replace it if necessary.
- 3 Check the light source window for smudges or fingerprints. Wipe it with a soft cloth or alcohol swab if necessary.
- 4 Position the light source over the chamber, then tighten the two screws.



Tighten them snugly—until the screw stops, then just a little bit more (up to ¼ turn).

- 5 Plug in the cable connector. Remove the connector cover, then route the cable between the chamber arm and duct. It connects to the light source connector (labeled LS) under the connector cover. Align the red dot on the connector with the red dot on the connection.



6 Turn the light source software switch ON.

Check the light source under **Environment > Light**.

Start Up	0/0 Environment	Constants	0/0 Stability	Log Files	Auto Programs	Measurements	Log
Flow: off	Manual 0.0 %						
H ₂ O: off							
CO ₂ : off							
Fan: off							
Temperature: off							
Light:							
Auxiliary:							
Auto Controls:							
						Ambient Meas: Qamb_out 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$ Meas: Qamb_in 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$	
						Fluorometer FirLS: 0 None Found	
						Head Light Source HeadLS: 0 Connector Off	
						Console Light Source ConsoleLS: 0 Connector Off	
						Total Leaf Q Qleaf: 0m 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$	

7 Check the response.

Under **Environment > Head LS**, set **Head LS** to **Setpoint**, and set the **Q Setpoint** to $200 \mu\text{mol m}^{-2} \text{s}^{-1}$. Open the chamber and look at the light that shines onto the chamber bottom (never look directly into the light sources).

If this is your first time using the light source, set **Color Ratio** to **Red: 0** and **Blue: 1**. You should see blue light. Then switch to **Red: 1** and **Blue: 0**. You should see red light. Then set **Red: 9** and **Blue: 1** to provide 9 parts red and 1 part blue, which is a typical ratio for gas exchange measurements.

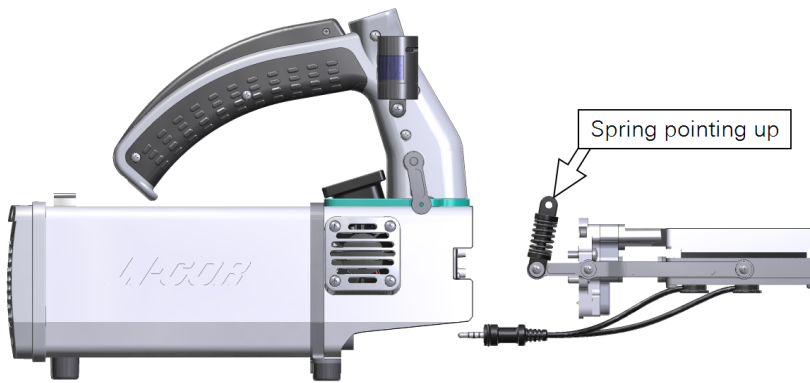
Clear-top large 6×6 cm leaf chamber

The 6×6 cm chamber is used for large leaves. This document describes how to install it on the LI-6800 head.

Installing the 6×6 cm chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

- 1 Hold the chamber in position, *with the latch springs up toward the handle*, and tighten the two captive chamber screws (146-15344).

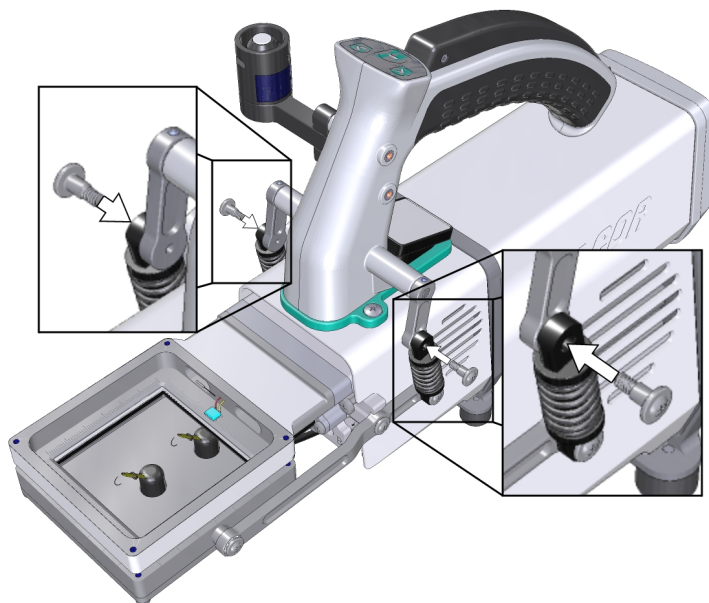


Caution: Use the captive chamber screws (part number 146-15344) to install the chamber. Other screws will cause leaks and problems with the mixing fan.



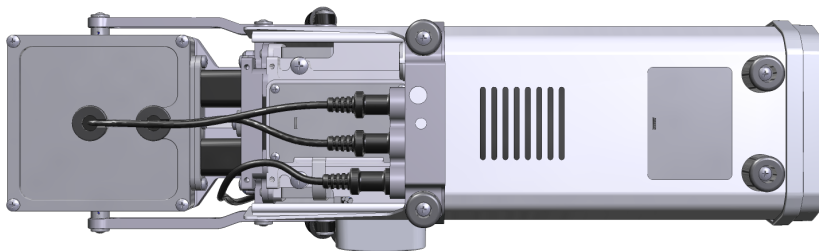
Make them snug—turn each screw until it stops, and then just a little bit more (up to ¼ turn). If you have a torque screwdriver, tighten them to 2.5 N·m (22 inch lbs).

- 2 Install the latch screws.

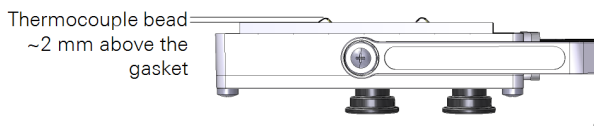


With the latch open, tighten until they are snug. If you have a torque screwdriver, tighten them to 1.1 N·m (9.4 inch lbs).

- 3** If you unplugged the light sensor cable, install the light sensor connector to the connection labeled PAR.
- 4** Install the leaf temperature thermocouple connectors to the connections labeled T1 and T2.



- 5** Adjust the thermocouple height.



- 6** Install the connector cover.

After swapping chambers, it is a good idea to verify that the chamber is sealed and that the software recognizes it. Run the leak test under **Start up > System Tests > Chamber Leak**. Check to see that the chamber is recognized under **Constants > System Constants > ChType**.

Connecting the 6×6 cm light source

The 6×6 cm light source attaches directly on top of the 6×6 cm Clear-top Chamber, Bryophyte Chamber, or Small Plant Chamber.



Warning: *The light sources for this product can emit potentially hazardous optical radiation (RG-2 CAUTION POSSIBLY HAZARDOUS OPTICAL RADIATION EMITTED FROM THIS PRODUCT), in excess of the Exempt Risk Group. Potential risk depends upon how one uses and installs this product. Do not operate the light source while detached from the chamber. **Do not look directly into the light source under any circumstances.** Operate the light sources with direct access to ambient air for cooling. Do not use the product in any manner not described in the manual.*



Avertissement: *Les sources de lumière pour ce produit peuvent émettre des radiations optiques potentiellement dangereuses, en excès à l'Exempt Risk Group-Groupe Exempt de Risque. Les risques potentiels dépendent de la façon dont on utilise et installe ce produit. N'opérez pas la source de lumière quand elle est détachée de la chambre. Ne regardez pas directement la source de lumière dans n'importe quelle circonstance. Opérez les sources de lumière avec accès direct à l'air ambiant pour le refroidissement. N'utilisez pas ce produit de n'importe quelle manière non décrite dans ce manuel.*

To install the light source:

- 1 Install a compatible chamber on the LI-6800; either the small plant chamber or the bryophyte chamber.
- 2 Prepare the instrument to accept the light source.

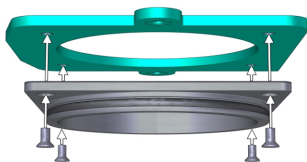
Go to **Environment > Light > Head Light Source** and tap the **Power Off** button. Or, just power off the instrument.

The screenshot displays the software interface for an environmental chamber. The 'Environment' tab is active, showing various control parameters. The 'Light' section is highlighted, indicating a 90-10% light level. The 'Color Spec' is set to 'r90b10'. A 'Color Spec' chart shows the following values:

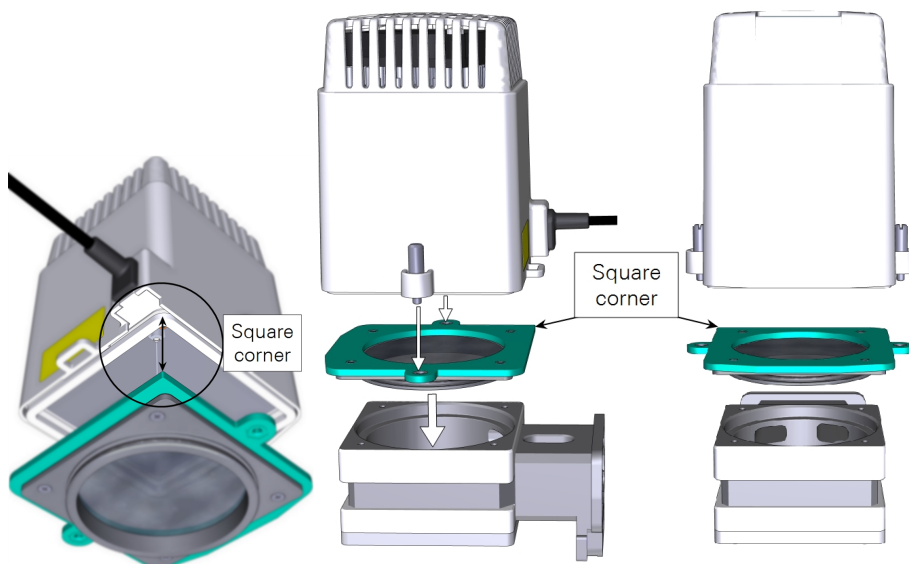
Color	Fraction	Value
Red	0.90	6
Green	0.10	1
Blue	0.00	0
White	0.00	0

The 'HeadLS: Q' is set to 7 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The 'Console Light Source' is also set to 7 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The 'Total Leaf Q' is -0 $\mu\text{mol m}^{-2} \text{s}^{-1}$. A 'Power Off' button is visible with the warning 'Power off before disconnecting.' A physical control panel with buttons for PAR, T1, T2, and LS is shown at the bottom right.

- 3 Check the chamber top Propafilm for dirt or damage. Clean or replace it if necessary. See *Propafilm replacement* on page 8-14.
- 4 Check the light source window for smudges or fingerprints. Wipe it with a soft cloth or alcohol swab if necessary.
- 5 Install the light source adapter plate on the chamber top. Tighten the screws (part number 150-07475) snugly—until the screw stops, then just a little bit more (up to $\frac{1}{4}$ turn).



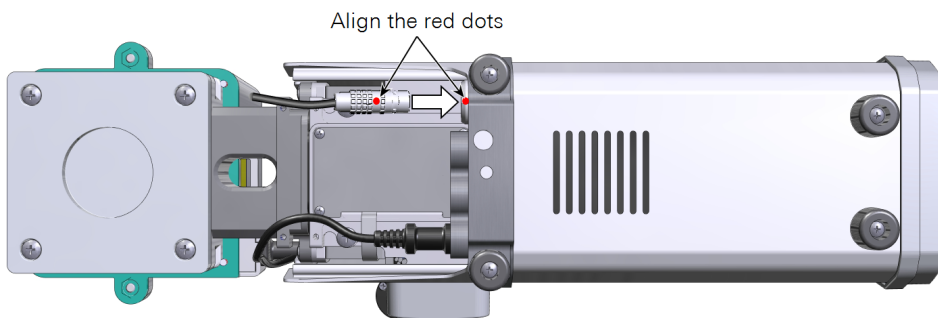
- 6 Install the light source on top of the adapter. The square corner of the adapter plate should be aligned with the cable.



7 Position the light source over the chamber, then tighten the two knurled captive screws until they are tight.

8 Plug in the cable connector.

Remove the connector cover, then route the cable between the chamber arm and duct. It connects to the light source connector (labeled **LS**) under the connector cover. Align the red dots on the connectors. Reinstall the connector cover.



9 Turn the light source software switch **ON**.

Check the light source under **Environment > Light > Head Light Source**.

The screenshot shows the software interface with the following elements:

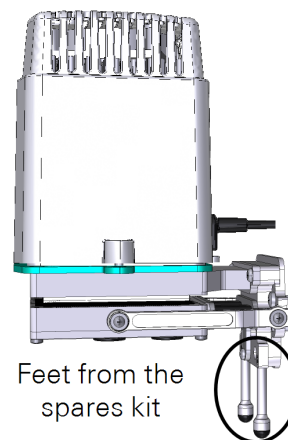
- Navigation Tabs:** Start Up, Environment (0/0), Constants, Stability (0/0), Log Files, Auto Programs, Measurements, Log.
- Environment Settings:**
 - Flow: Off (Manual 0.0 %)
 - H₂O: Off
 - CO₂: Off
 - Fan: Off
 - Temperature: Off
 - Light:** (Selected)
 - Auxillary:
 - Auto Controls:
- Measurements Panel:**
 - Ambient:** Meas: Qamb_out (7 μmol m⁻² s⁻¹), Meas: Qamb_in (-0 μmol m⁻² s⁻¹)
 - Fluorometer:** Flt.S: 0, None Found
 - Head Light Source:** HeadLS: Q, Connector Off
 - Console Light Source:** ConsoleLS: Q, Connector Off
 - Total Leaf Q:** Qleaf: Qin (-0 μmol m⁻² s⁻¹)
- Power On Notification:** It is now safe to connect or disconnect a light source.
- Control Buttons:** PAR, T1, T2, LS.

10 Check the response.

Under **Environment > Head LS**, set **Head LS** to **Setpoint**, and set the **Q Setpoint** to 200 μmol m⁻² s⁻¹. Open the chamber and look at the light that shines onto the chamber bottom (never look directly into the light sources).

If this is your first time using the light source, adjust the Red, Green, Blue, and White light ratios to get a sense of how they work. Then set **Red: 9** and **Blue: 1** to provide 9 parts red and 1 part blue, which is a typical ratio for gas exchange measurements.

For laboratory applications, where the instrument head sits on a bench, install the two chamber feet from the spares kit (part number 9968-253) to prevent the head from tipping over. We do not recommend the feet for field measurements because they can be damaged by inadvertent impacts.



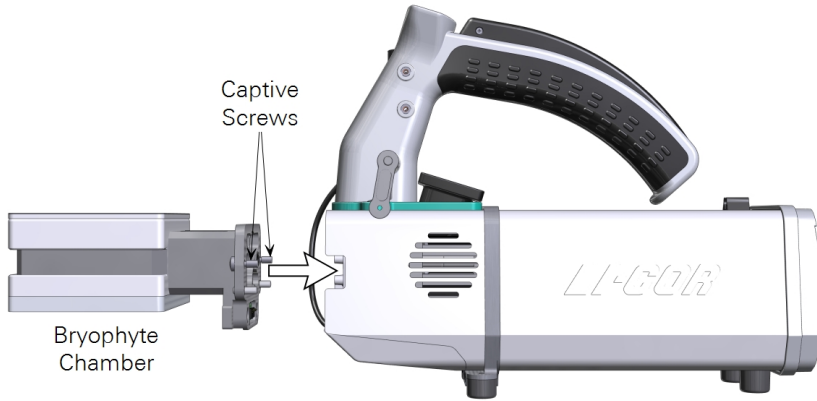
Bryophyte chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

The Bryophyte Chamber (6800-24) is used to measure CO₂ and H₂O gas exchange from mosses, hornworts, liverworts, and lichens. The chamber holds sample material on wire mesh or in a shallow well during the measurement. It features a clear Propafilm™ top for measurements under ambient lighting conditions. It is compatible with the Large Light Source for measurements under controlled lighting with any combination of red, green, blue, and white light.

Installing the bryophyte chamber

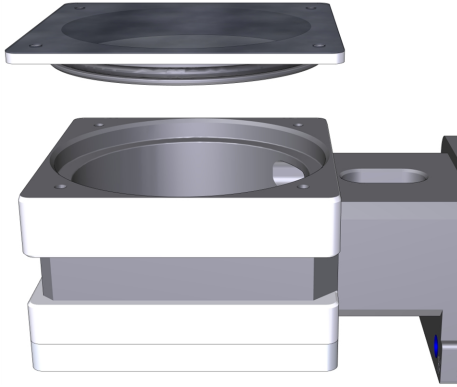
- 1 Install the chamber adapter.



- 2 Tighten the screws snugly—until they are tight and then just a little bit more. Reinstall the connector cover.

Preparing the sample chamber

- 1 Remove the chamber top. It lifts off of the chamber.



- 2 Install the chamber bottom.

There are three options for loading the sample: A) on the screen with short risers, B) on the screen with tall risers, or C) in a well. The well is on one side of the chamber bottom; the screens mount to the other side. Install the chamber bottom in the orientation that you prefer.

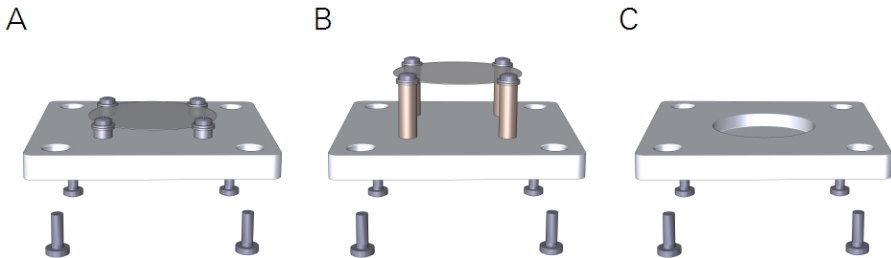
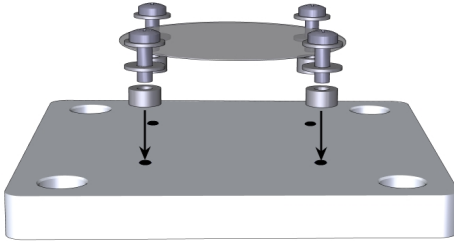


Figure 5-2. Chamber bottom configurations. A shows the short risers; B shows the tall risers; and C shows the well.



- 3 Power on the LI-6800, if it is not already on, and verify that the instrument recognizes the 6800-24 chamber under **Start Up > Configuration > Chamber Setup**.
- 4 Choose whether to compute **Area-based fluxes** or **Mass-based fluxes**.
- 5 Run the leak test under **Start Up > System Tests > Chamber Leak**.

Loading a sample

Remove a sample from the substrate using a knife or cookie cutter. You can place a sample on the screen or in the well. If you're computing fluxes on the basis of mass, you'll need to weigh each sample and make a record of the mass. For area-based fluxes, you'll need to determine the sample area. After loading the sample, press the chamber top onto the chamber.

Making a basic measurement

Below we offer some basic settings that will work for a typical measurement.

- 1 Set the **Environment** controls.
 - **Flow:** Set **Flow:** On; **Pump speed:** Auto; **Flow rate:** 700 $\mu\text{mol s}^{-1}$; **Press Valve:** 0.0 kPa
 - **H₂O:** Set **H₂O:** On; **RH_{air}:** 50%
 - **CO₂:** Set **CO₂ injector:** On; **Soda Lime:** Scrub Auto; Tap **CO₂_s** and enter a set-point of 400 $\mu\text{mol mol}^{-1}$
 - **Fan:** Set **Mixing fan:** On; **Fan Speed:** 10,000 rpm
 - **Temperatures:** Check the temperatures to see their current values
- 2 Set the **Constants**.

Under **Constants > Gas Exchange**, enter the **Area** or **Mass**.
- 3 Configure Logging options.

Configure the **Match Options** and **Logging Options** as desired. Check **Prompt on (manual) log**, which will open a prompt when you press the **Log** button, where you can enter the weight or area.

- 4 Open a log file.
- 5 Tap the **Log** button to record a measurement.

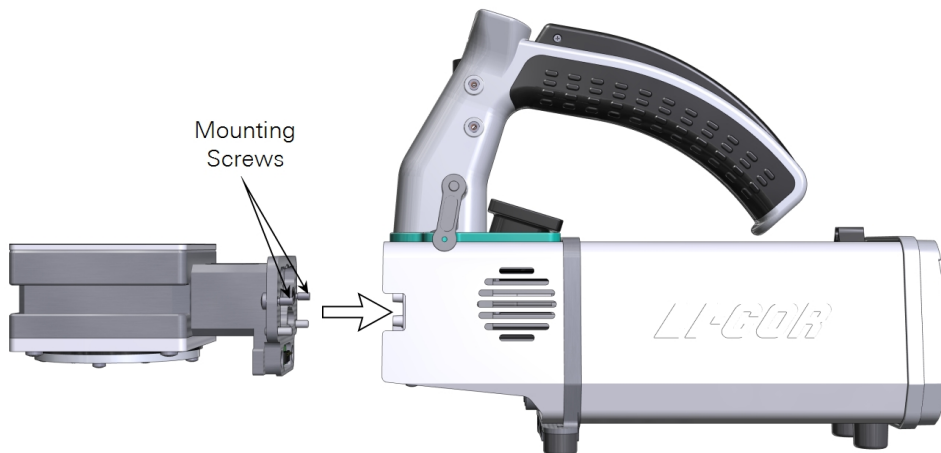
Small plant chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

The Small Plant Chamber (6800-17) is used to measure CO₂ and H₂O gas exchange from whole plants such as *Arabidopsis thaliana*, turf grass, and similar samples. The chamber accommodates 65 mm (2.5 inch) pots or 38 mm (1.5 inch) Cone-tainers™. It has a clear Propafilm™ top for ambient-lighted measurements. It is compatible with the Large Light Source for measurements under controlled lighting. This document describes how to install the chamber on the LI-6800 and how to make basic measurements with it.

Installing the chamber

- 1 Hold the chamber in position and tighten the two chamber mounting screws (146-15344).



Caution: You must use the captive chamber screws (part number 146-15344) to install the chamber. Other screws will cause leaks and problems with the mixing fan, even if they seem to fit.

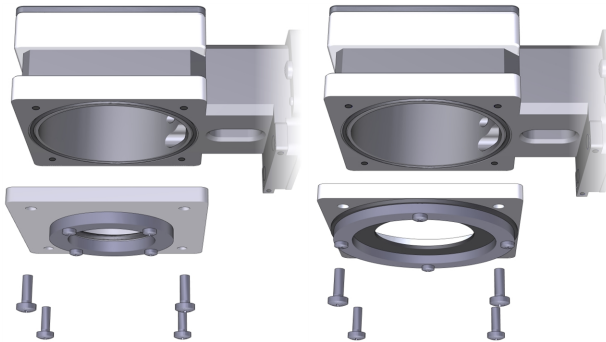
Make them snug—turn each screw until it stops, and then just a little bit more (up to ¼ turn). If you have a torque screwdriver, tighten them to 2.5 N·m (22 inch lbs).

- 2 If you unplugged the light sensor cable, install the light sensor connector to the connection labeled PAR.
- 3 Install the connector cover.

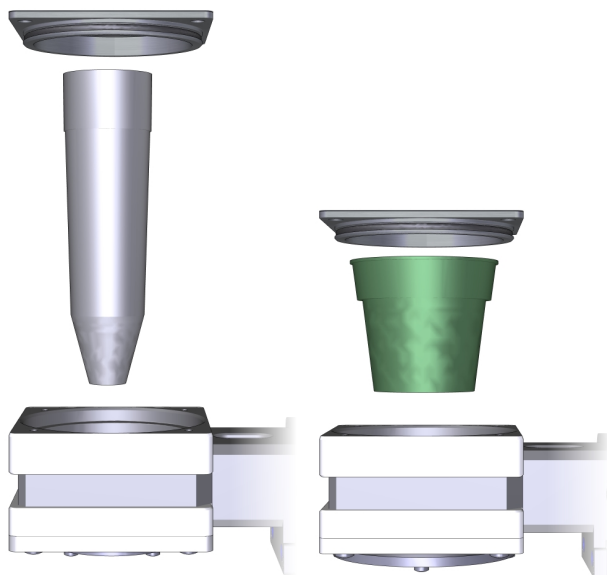
Loading a plant in the chamber

Prior to measuring a plant, install an empty Cone-tainer or flower pot in the chamber and run the leak test to ensure everything is working properly.

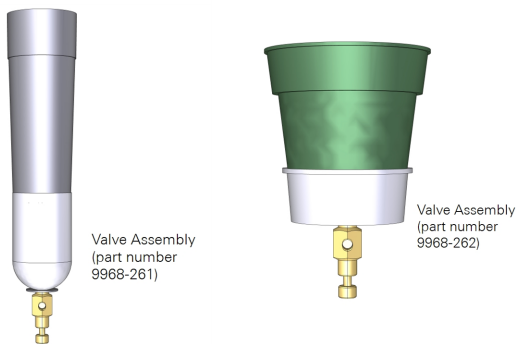
- 1 Install the chamber bottom that is suitable for your pots—either use the bottom with a large opening for the 65 mm pots, or the bottom with a small opening for the Cone-tainers.



- 2 Remove the chamber top cover and insert an empty Cone-tainer or flower pot into the chamber base. Press gently to compress the surrounding O-ring slightly around the Cone-tainer or flower pot.



- 3** Press the top cover onto the chamber.
- 4** Install the valve assembly on the bottom of the Cone-tainer or flower pot. Press it onto the Cone-tainer or pot until it is firmly attached.



- 5** Power on the LI-6800, if it is not already on, and verify that the instrument recognizes the 6800-17 chamber under **Start Up > Configuration > Chamber Setup**.
- 6** Run the tests under **Start Up > System Tests**.

Making a basic measurement

You'll need to grow plants in either the Cone-tainers or the 65 mm pots.

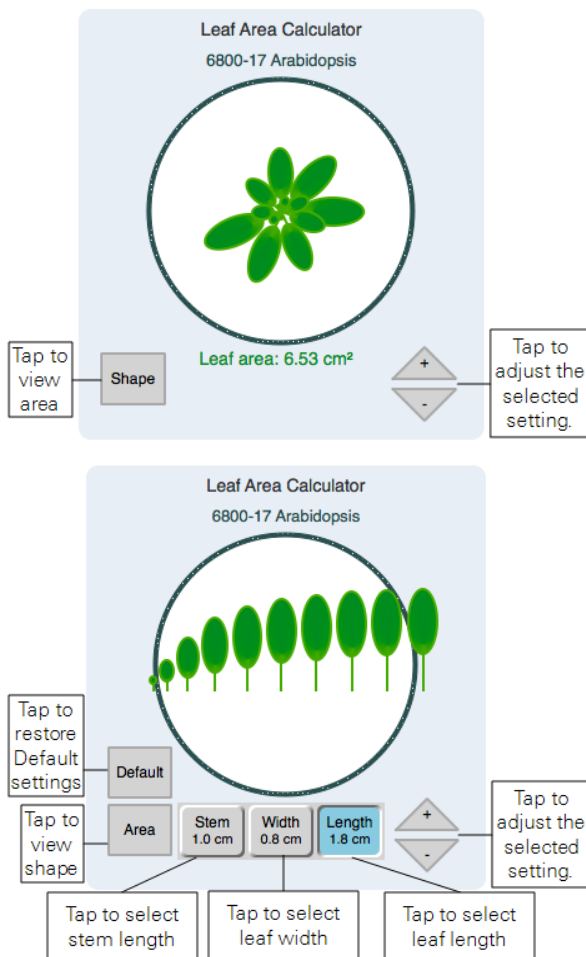
Note: Although you can acquire Cone-tainers and 65 mm pots from greenhouse suppliers, we have found that many commercial flower pots do not seal in this chamber. Therefore we recommend the Cone-tainers and 65 mm pots that we provide because they have been tested in this chamber and are known to work.

You are nearly ready to measure gas exchange. Below we offer some basic settings that will work for a typical measurement.

- 1** Under **Start Up > Configuration**, choose whether to compute **Area-based fluxes** or **Mass-based fluxes**.
- 2** Set the **Environment** controls:
 - **Flow:** Set **Flow:** On; **Pump speed:** Auto; **Flow rate:** 800 $\mu\text{mol s}^{-1}$; **Press Valve:** 0.2 kPa
 - **H₂O:** Set **H₂O:** On; **RH_{air}:** 50%
 - **CO₂:** Set **CO₂ injector:** On; **Soda Lime:** Scrub Auto; Tap **CO₂_r** and enter a set-point of 410 $\mu\text{mol mol}^{-1}$
 - **Fan:** Set **Mixing fan:** On; **Fan Speed:** 10,000 rpm
 - **Temperatures:** Check the temperatures to see their current values
- 3** Load the chamber with a plant, as described previously, only this time use a pot with a plant rather than an empty pot.
- 4** Install the **Valve Assembly** on the bottom of the pot.
- 5** Adjust the small valve on the bottom of the **Valve Assembly** until the leak rate, under **Environment > Flow**, is less than 20%.
- 6** Set the **Constants**.

For area-based fluxes, set the leaf area under **Constants > Gas Exchange**.

- The Leaf Area Calculator uses a simple growth model, using three logistic curves to determine stem length, leaf width, and leaf size as a function of time. Nodes 150 degrees from each other sprout every *n* time units. In **Area** mode, the adjust buttons add or decrement the age by 1 time unit. In **Shape** mode, you can adjust the asymptotic size of the selected attribute, stem length, leaf width, or leaf length, allowing a range of *Arabidopsis* varieties to be simulated.
- Choose between **Shape** or **Area** by tapping the corresponding button.



- Under **Constants > Leaf Temperature**, be sure that the instrument uses **Energy Balance**, unless you are measuring temperature another way.

7 Configure Logging options.

Configure the **Match Options** as desired. If you are not using the **Leaf Area Calculator**, you can determine the leaf area any number of other ways and enter the area manually (e.g., take a photo and use ImageJ to determine area). If you enter the area this way, check **Prompt on (manual) log**, which will open a prompt when

you press the **Log** button, where you can enter the constants, including weight or area.

- 8 Open a log file.
- 9 Tap the **Log** button to record a measurement.

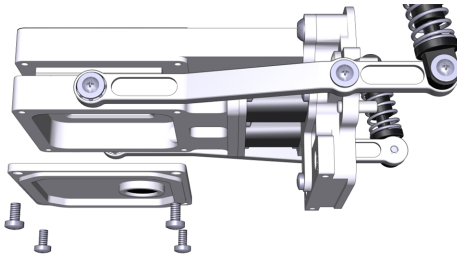
Caution: Do not leave a plant sample enclosed in the chamber with the flow set to off. Doing so can let condensation form in the chamber and gas analyzers, which could damage the LI-6800.

Light source extension cable

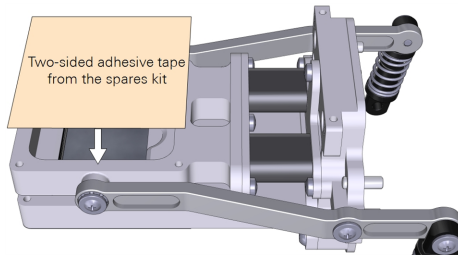
The Light Source Extension Cable (9968-243) is used to connect and operate a light source from the LI-6800 console **ACCESSORY** connector. In this document, we describe how to use the extension cable to add a second light source to illuminate both sides of a leaf in the chamber. The images show a 3×3 cm chamber, but the procedure is essentially the same for the 6×6 cm chamber.

Installing Propafilm on the lower chamber

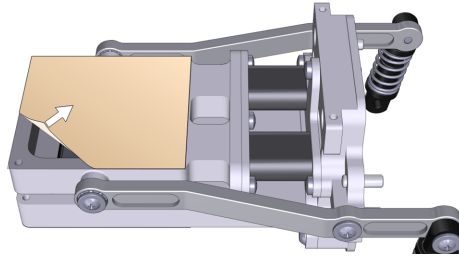
- 1 Remove the lower leaf-temperature thermocouple plate.



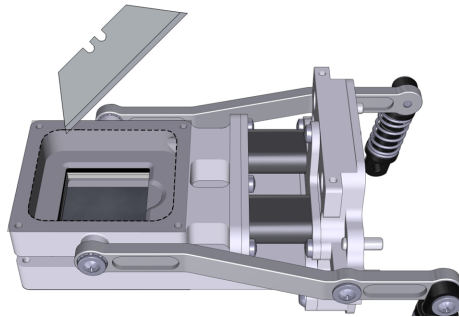
- 2 Cut a piece of double-stick adhesive film.
Make it 6 × 6 cm for the small chamber and about 10 × 10 cm for the large chamber. Separate one side of the film from the paper wrapper, exposing the adhesive surface.
- 3 Press the exposed adhesive onto the lower chamber opening.



- 4** Remove the other side of the paper wrapper, exposing the second side of the adhesive.



- 5** Trim the adhesive tape from the inside of the chamber using a razor blade or a sharp knife.



- 6** Prepare a piece of Propafilm: cut it to 6×6 cm for the small chamber or about 10×10 cm for the large chamber.
- 7** Align one edge of the Propafilm with one edge of the adhesive. While holding the Propafilm tight, press the Propafilm onto the adhesive. Smooth any bubbles or channels in the film.
- 8** Trim the Propafilm from around the outside of the chamber.
- 9** Install the chamber on the LI-6800 head and run the leak test under **Start Up > System Tests > Chamber Leak**.

In this configuration, the chamber is a clear-top and clear-bottom chamber. You can make measurements this way if desired, or install two light sources on the chamber.

Since there is no place for a thermocouple in the chamber, the instrument will use energy balance to compute leaf temperature. You can choose an alternative temperature measurement under **Constants > System Constants > Leaf Temperature Source**.

Installing the lower light source on the chamber

This procedure is slightly different for the small and large light sources. In both cases, the lower light source attaches the same way as the corresponding upper light source.

- 1 Prepare the instrument to accept the light source.

Go to **Environment > Light > Console Light Source** and verify that the light source is powered off. The display will say "It is now safe to connect or disconnect a light source".

The screenshot shows the instrument's control interface. The top navigation bar includes 'Start Up', 'Environment' (highlighted in blue), 'Constants', 'Stability', 'Log Files', 'Auto Programs', 'Measurements', and 'Log'. The 'Environment' section is expanded to show 'Light' (highlighted in blue), 'Auxiliary:', and 'Auto Controls:'. The 'Light' section shows 'SLS-561020 off'. A 'Power On' button is visible, with a message: 'It is now safe to connect or disconnect a light source.' The right side of the screen displays various measurement data including Ambient, Fluorometer, Head Light Source, Console Light Source, and Total Leaf Q.

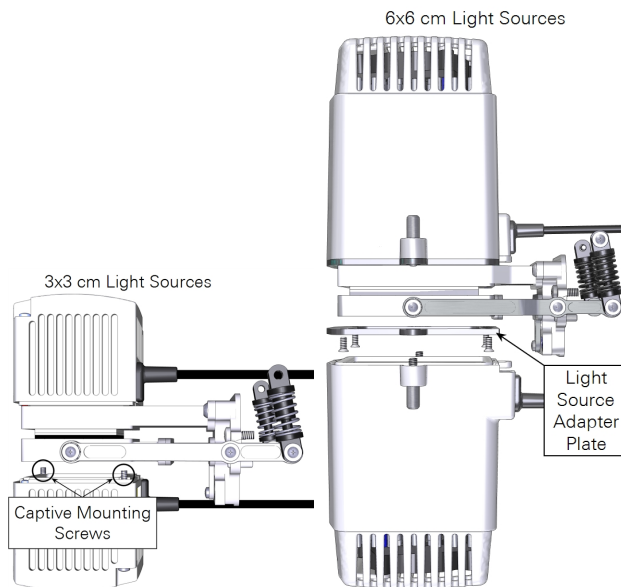
Measurement	Value
Ambient	Meas: Qamb_out: 7 $\mu\text{mol m}^{-2} \text{s}^{-1}$ Meas: Qamb_in: -0 $\mu\text{mol m}^{-2} \text{s}^{-1}$
Fluorometer	FIRLS: Q: None Found
Head Light Source	HeadLS: Q: 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$
Console Light Source	ConsoleLS: Q: Connector Off
Total Leaf Q	Clear: Qin: 0 $\mu\text{mol m}^{-2} \text{s}^{-1}$

- 2 Check the chamber Propafilm for dirt or damage. Clean it if necessary.
- 3 Check the light source window for smudges or fingerprints. Wipe it with a soft cloth or alcohol swab if necessary.

- 4 For the 3×3 chamber, puncture the Propafilm at the screw holes where the light source mounting screws go. Trim the Propafilm around the screw holes.
- 5 Hold the light source in place and tighten each screw.

The small light source attaches to the lower chamber with two screws in the corners.

The large light source has four screws that connect an adapter plate to the chamber and two knurled screws hold the light source to the adapter plate.



- 6 Plug the light source cable into the light source extension cable and connect the extension cable to the console connector labeled **ACCESSORY**.
- 7 Secure the cable in the clasp and sheath.



8 Check the light source under **Environment > Light**.

When the console light source is detected, the instrument will turn **Console LS** on and present you with options to configure the light source.

The screenshot displays the software interface for configuring the light source. The 'Environment' tab is selected, and the 'Light' section is active. The 'Light' parameter is set to 'SLS-561020 off' with a value of '200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ '. The 'Console Light Source' section shows 'ConsoleLS: Q' with a value of '200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ '. A diagram of the light source shows 'Red 0.90' and 'Blue 0.10' fractions, with a '20 $\mu\text{mol m}^{-2} \text{s}^{-1}$ ' value for the blue component. A '6800-02 SLS-561005' light source is shown with a 'ConsoleLS: Q' label and '200 $\mu\text{mol m}^{-2} \text{s}^{-1}$ ' value. A 'HEAD (1) ACCESSORY HEAD (2)' is also shown.

9 Under **Environment > Light > Console Light Source**, set **Control Mode** to **Setpoint**, and set the **Setpoint** to $200 \mu\text{mol m}^{-2} \text{s}^{-1}$. Open the chamber and look at the light that shines out of the chamber bottom (never look directly into the light sources).

You can control both light sources manually or automatically with an **Auto Control**, **Auto Program**, or **Background Program**.



Warning: *The light sources for this product can emit potentially hazardous optical radiation (RG-2 CAUTION POSSIBLY HAZARDOUS OPTICAL RADIATION EMITTED FROM THIS PRODUCT), in excess of the Exempt Risk Group. Potential risk depends upon how one uses and installs this product. Do not operate the light source while detached from the chamber. **Do not look directly into the light source under any circumstances.** Operate the light sources with direct access to ambient air for cooling. Do not use the product in any manner not described in the manual.*



Avvertissement: *Les sources de lumière pour ce produit peuvent émettre des radiations optiques potentiellement dangereuses, en excès à l'Exempt Risk Group-Groupe Exempt de Risque. Les risques potentiels dépendent de la façon dont on utilise et installe ce produit. N'opérez pas la source de lumière quand elle est détachée de la chambre. Ne regardez pas directement la source de lumière dans n'importe quelle circonstance. Opérez les sources de lumière avec accès direct à l'air ambiant pour le refroidissement. N'utilisez pas ce produit de n'importe quelle manière non décrite dans ce manuel.*

Using multiple light sources

Software version 1.4 adds support for using multiple light sources that are contributing to Qleaf:Qin.

Suppose you are using the 6800-12A 3×3 chamber with a 6800-02 3×3 light source attached to the top (and connected to the LS sensor on the sensor head), and another 3×3 light source attached to the chamber bottom (remove lower plate, add Propafilm, attach light source). The bottom light source is attached to the light source connector on the console.

How to set this up? *Figure 5-3* on the next page illustrates the first part of the process—getting the light sensors attached and enabled.

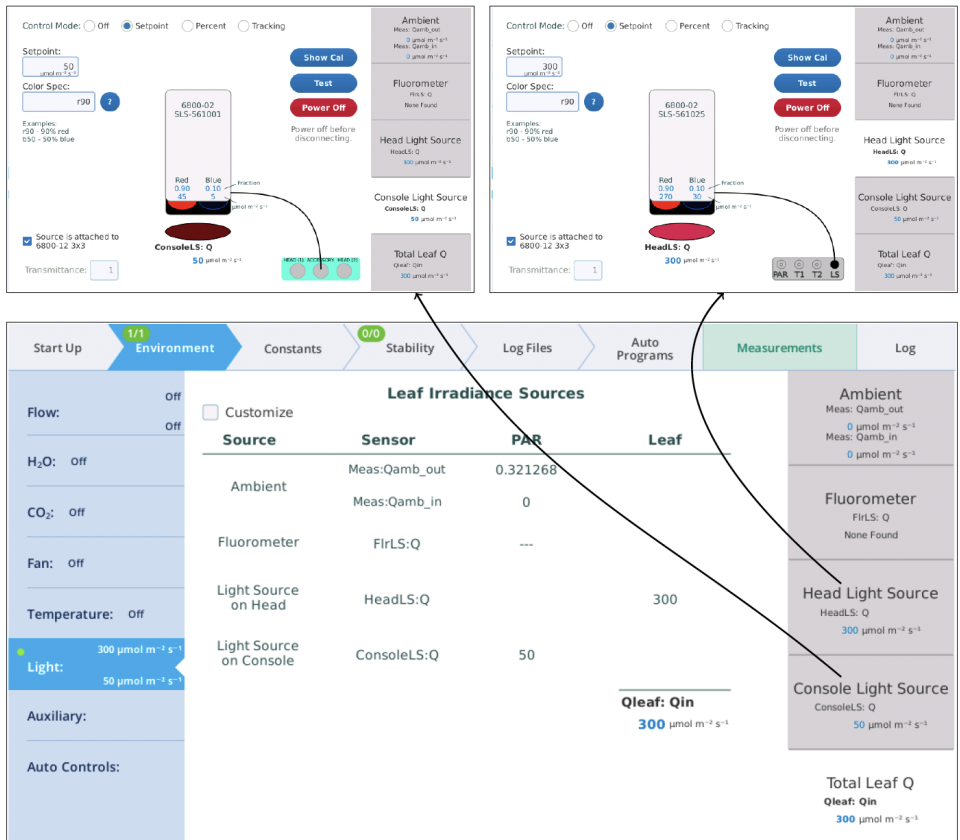


Figure 5-3. When initially configured, only one light source will be contributing to the leaf.

In Figure 5-4 on the facing page, there are two active light sources, but the software is not yet configured to use both of them when computing the total light on the leaf. Figure 5-4 on the facing page shows how to accomplish that.

The screenshot displays the 'Leaf Irradiance Sources' configuration window. At the top, navigation tabs include Start Up, Environment (2/2), Constants, Stability (0/0), Log Files, Auto Programs, Measurements, and Log. The 'Leaf Irradiance Sources' table is as follows:

Source	Sensor	PAR	Multiplier	Leaf
Meas:Qamb_out	Meas:Qamb_out	0.321268	X 0	0
Meas:Qamb_in	Meas:Qamb_in	0	X 0	0
FirLS:Q	FirLS:Q	---	X 0	0
Light Source on Head	HeadLS:Q	300	X 1	300
Light Source on Console	ConsoleLS:Q	50	X 1	50

Below the table, the 'Qleaf: Qin' is shown as 350 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The 'Qin setpoint partitions (Auto & Background programs):' section includes input fields for Head LS (1), Console LS (1), and Qin setpoint (456 $\mu\text{mol m}^{-2} \text{s}^{-1}$), along with a 'Test' button. On the right, a 'Measurements' panel shows 'Ambient' (0 $\mu\text{mol m}^{-2} \text{s}^{-1}$), 'Fluorometer' (None Found), 'Head Light Source' (300 $\mu\text{mol m}^{-2} \text{s}^{-1}$), and 'Console Light Source' (50 $\mu\text{mol m}^{-2} \text{s}^{-1}$), with a 'Total Leaf Q' of 350 $\mu\text{mol m}^{-2} \text{s}^{-1}$.

Figure 5-4. Getting two sources to contribute to the leaf.

At this point, you can set the light sources independently, and their total will be Q_{in} . But, from an Auto Program (or Background Program), if you want to be able to specify Q_{in} , with the light sources "figuring out" what their setpoints should be to achieve that, there is one more step, and that is to specify the proportions each light source should contribute (Figure 5-5 on the next page).

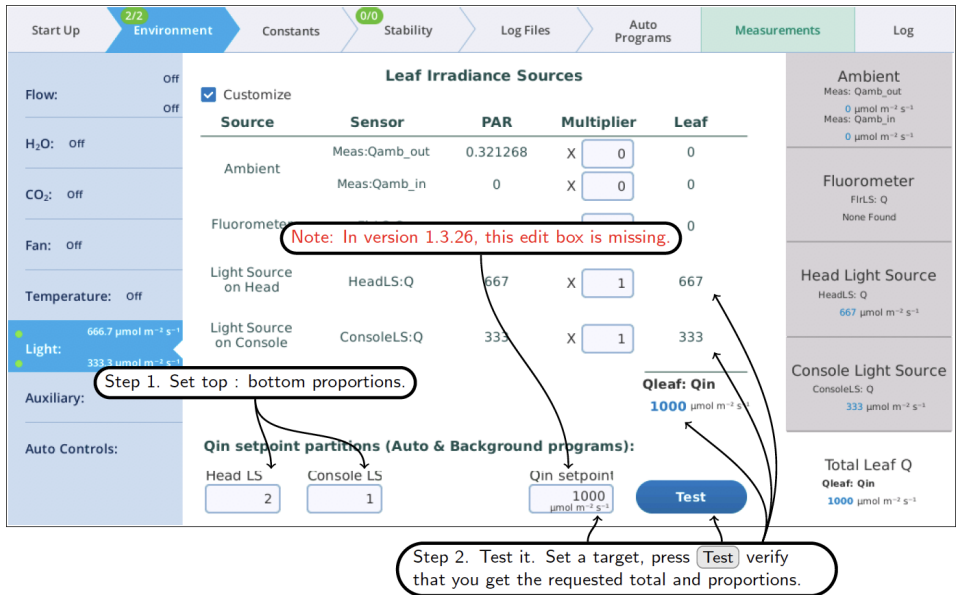


Figure 5-5. Specifying the relative proportions for light sources.

Notes about the values entered in the *Qin* setpoint partition edit boxes (i.e., **Head LS** and **Console LS**) at the bottom of Figure 5-5 above.

- Their values only come into play when an Auto Program (or Background Program) sets a *Qin* target, or when the **Test** button is pressed.
- Their values are relative. To make two light sources balance, you could enter 0.5 and 0.5, or 1 and 1, or 2 and 2, etc. To make one twice the other, you could enter 0.67 and 0.33, or 2 and 1, etc.

Notes about the **Multiplier** edit boxes visible when in **Customize** mode.

- They serve a multiplier that converts the light source or sensor value to the value that contributes to the leaf.
- For light sources, the values would normally be 1 or 0, depending if the source is or is not contributing to *Qin*. If there is a "transmittance" effect (for example, the light source is further away from the leaf than normal, or there is a customized chamber) that effect should be specified using the **transmittance** edit box on the light source's control screen.

- For in-chamber light sensors (or external quantum sensor), the weighting factor is the mechanism for accounting for non-standard configurations. For example, with a clear chamber top and a clear chamber bottom, one method for getting the correct total Q_{in} would be to a) measure incoming from the top by using a multiplier of 1.0 for the internal quantum sensor, b) mount the external quantum sensor so it faces down, and c) use a multiplier for it of 0.9 to account for the bottom chamber's propafilm.
- They serve a multiplier that converts the light source or sensor value to the value that contributes to the leaf.
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Using the Conifer Sprig Kit

The Conifer Sprig Kit (part number 9968-271) installs directly on the large leaf chamber so that the chamber can accommodate sprigs and shoots that have leaves in a spiral arrangement. Be sure to use the Advanced Polymer (AP) gaskets with the Conifer Sprig Kit. The blocks can be used in ambient lighting conditions and with one or two large light sources.

Installing the sprig adapter blocks

The blocks can be installed on the large leaf chamber while it is attached to the head, but you may find that it is easier to install the blocks while the chamber is removed from the head.

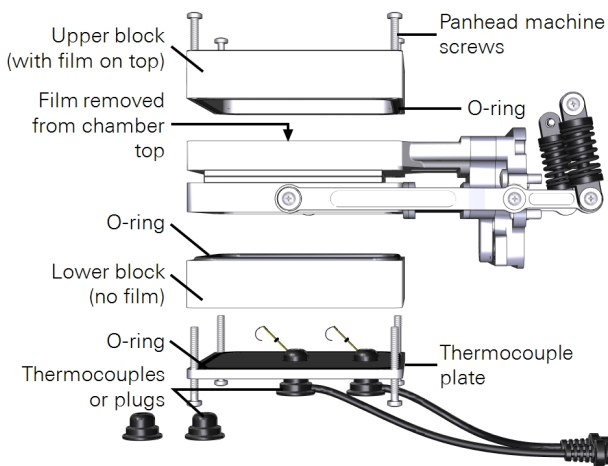
Preparing the chamber

- 1** Remove the Propafilm from the upper part of the 6x6 cm leaf chamber (not the sprig block). Puncture the film and pull it free of the top chamber. Pull carefully so that you remove as much adhesive as possible. Use rubbing alcohol to soften any remaining adhesive residue on the chamber, and remove it so that the upper surface is clean. There is extra propafilm in the chamber spares kit so you can replace the cover when you aren't using the adapter blocks.
- 2** Remove the thermocouple plate from the bottom of the lower chamber. Keep the screws, plate, and thermocouples close by for re-installation.

Measurements with ambient light on one side

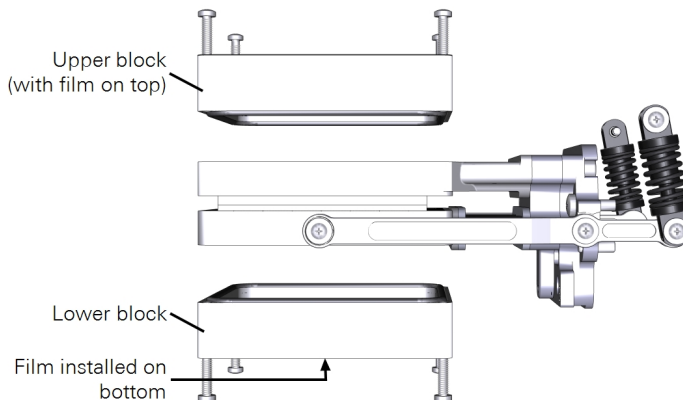
The blocks can be used to make measurements under ambient lighting from above. In this scenario, you'll have the thermocouples available to measure leaf temperature, but it is unlikely that they will make contact with the leaf. Therefore, we recommend that you use energy balance to compute leaf temperature. To install the blocks:

- 1** Attach the block with film on top to the upper chamber using four panhead screws (part number 150-16889).
- 2** Install the thermocouples or thermocouple plugs in the thermocouple plate.



Measurements with ambient light on both sides

For measurements with ambient light on two sides, you must remove the thermocouple mounting plate from the lower chamber and install propafilm on the lower block as described on *Using the Conifer Sprig Kit* on page 5-41. In this arrangement, there is no leaf thermocouple, so the instrument will automatically switch to energy balance to compute leaf temperature.

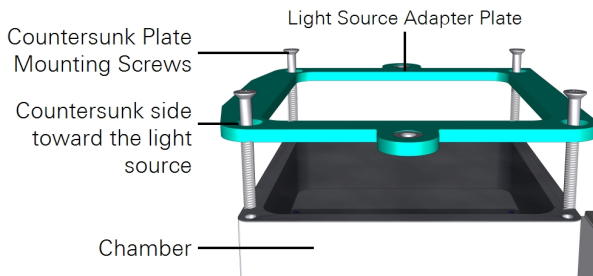


Measurements with a light source on one side

The Light Source Adapter Plate is required to connect the light source. To install the light source:

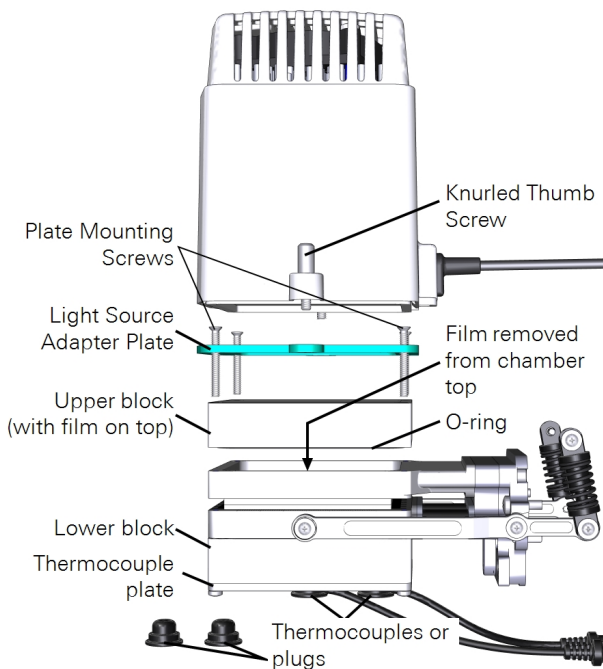
- 1 Attach the plate to the chamber using four countersunk plate mounting screws (part number 150-06115).

Pay careful attention to the orientation of the plate. The countersunk side will be in contact with the light source.



- 2 Mount the light source to the plate with both knurled thumb screws.

- 3** Install the thermocouples or thermocouple plugs in the thermocouple plate.
- 4** Connect the light source cable to the **LS** connector on the head.



Measurements with light sources on two sides

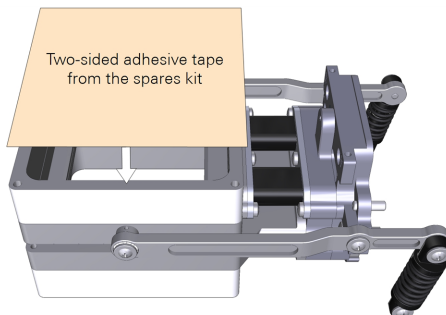
When using two light sources to illuminate both sides of the chamber, you'll use the Light Source Extension Cable (part number 9968-243) to connect one of the light sources to the LI-6800 console accessory connector. The other light source cable will connect to the **LS** connector on the head.

- 1** Install Propafilm on the bottom of the lower block.
 - A** Carefully clean the surface that is accepting the film.

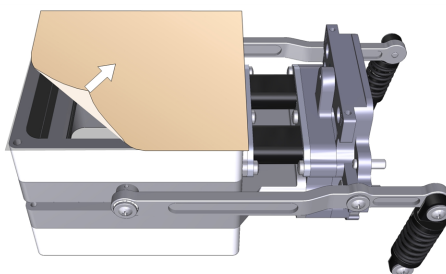
Be sure that there is no film or adhesive residue remaining on the chamber. Use rubbing alcohol to soften the adhesive, if needed.
 - B** Cut a piece of double-stick adhesive film.

Make it about 9×9 cm for the large chamber. Separate one side of the film from the wrapper, exposing the adhesive surface.

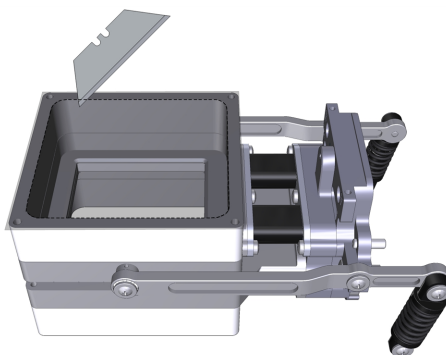
- C** Press the exposed adhesive onto the chamber opening.



- D** Remove the other side of the wrapper, exposing the second side of the adhesive.



- E** Trim the adhesive tape from the inside of the chamber using a razor blade or a sharp knife.

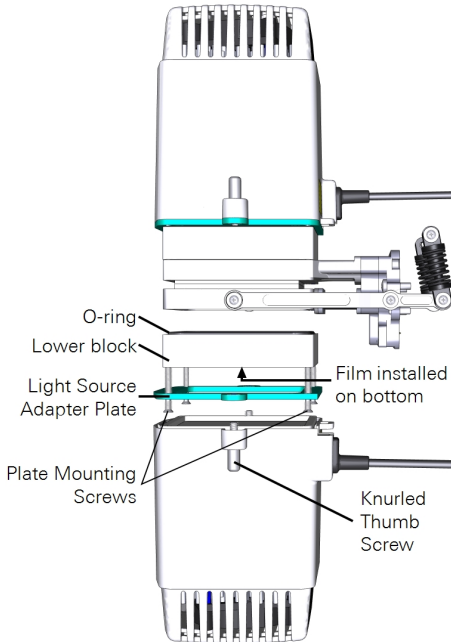


- F** Prepare a piece of Propafilm.
Cut it to about 9×9 cm for the large chamber.

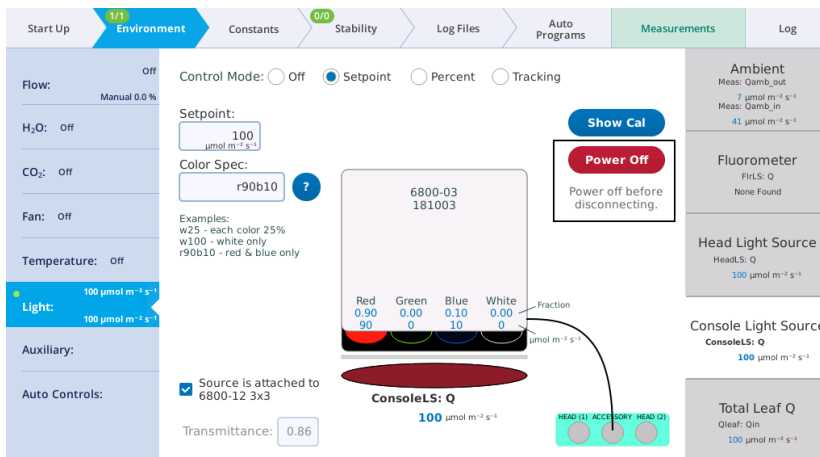
- G** Attach the Propafilm

Align one edge of the Propafilm with one edge of the adhesive. While holding the Propafilm tight, press the Propafilm onto the adhesive. Smooth any bubbles or channels between the film and adhesive.

- H** Trim the Propafilm from around the outside of the chamber and cut the film out of the screw holes.
 - I** Install the chamber on the LI-6800 head and run the leak test under **Start Up > System Tests > Chamber Leak**.
- 2** Attach the plate to the lower chamber using four plate mounting screws (part number 150-06115).
 - 3** Mount the light source to the plate by turning both knurled thumb screws.
 - 4** Connect one light source cable to the **LS** connector on the head.
 - 5** Connect the second light source cable to the console **Accessory** connector using the Light Source Extension Cable.



The second light source has the same options as the first light source. It is configured under the **Environment > Light > Console Light Source**.



Insect respiration chamber

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

The Insect Respiration Chamber (6800-89) is a column and adapter that allows you to measure gas exchange from small, discrete samples using the LI-6800. In this section we describe how to install the chamber and take measurements. The chamber uses the instrument pump to circulate air. The mixing fan is deactivated because laminar flow through the chamber provides adequate mixing.

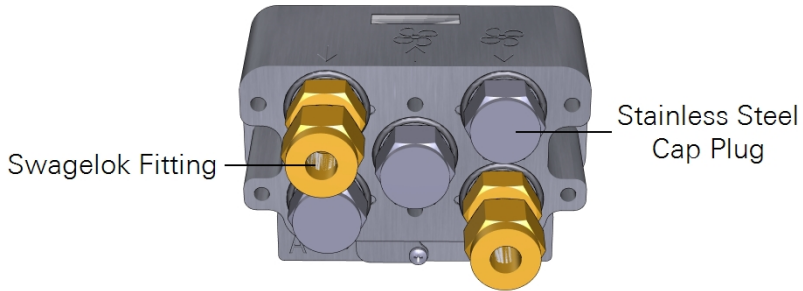
Installing the chamber

- 1 Install the chamber adapter.



- 2 Tighten the screws snugly—until they are tight and then just a little bit more.

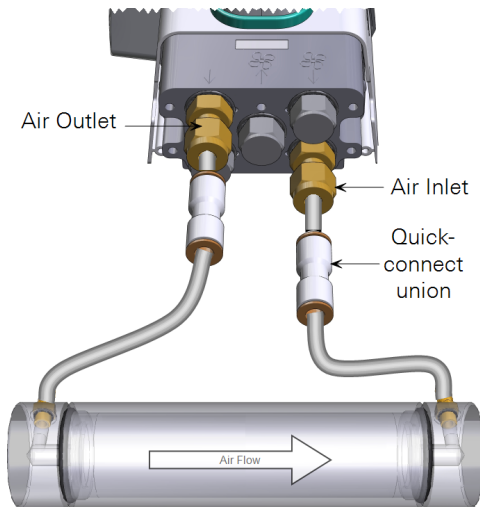
- 3** Install the three stainless steel caps and two Swagelok fittings onto the chamber adapter.



- 4** Cut two pieces of hose, each about 5 to 10 cm long. Remove the brass nuts on the air inlet and air outlet. Remove the brass ferrules and set them aside. Install a set of vinyl ferrules on each hose, as shown.



- 5** Install the hoses between the chamber and the head, as shown.

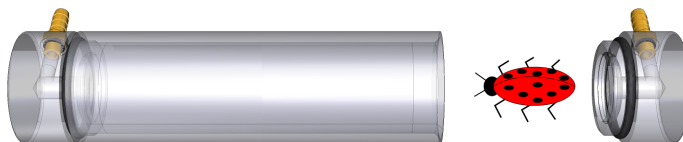


- 6 Verify that the chamber is recognized under **Constants > System Constants**.
- 7 Run the **Chamber Leak Test** under **Start Up > Warmup/System Tests**. Correct any issues indicated by the test.
- 8 Reinstall the connector cover.

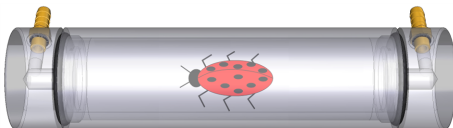
Making a basic measurement

- 1 Set the **Environment** controls:
 - **Flow:** Set **Flow:** On; **Pump speed:** Auto; **Flow rate:** 400 $\mu\text{mol s}^{-1}$ or your desired flow rate; **Press Valve:** 0.0 kPa
 - **H₂O:** Set **H₂O:** On; **RH_{air}:** 50%
 - **CO₂:** Set **CO₂ injector:** On; **Soda Lime:** Scrub Auto; Tap **CO₂_r** and enter a setpoint of 400 $\mu\text{mol mol}^{-1}$. If you are expecting large fluctuations in gas exchange, e.g., human breath cycles, it is best to control on **CO₂_r**. One of **CO₂_r** or **CO₂_s** should always be stable when you make measurements.
 - **Fan:** Set **Mixing fan:** Off
 - **Temperatures:** Check the temperatures to see their current values
- 2 Load the chamber.

Disconnect one of the tubes (either **Air Inlet** or **Air Outlet**) and unscrew the corresponding cap from the chamber.



- 3 Load the sample, replace the cap, and then reinstall the tube.



- 4 Under **Log Files > Open a Log File**, create a new file or open an existing file. If desired, configure the software to **Prompt on Log**, which will allow you to enter the sample weight with each measurement.
- 5 Take a measurement.
Under **Measurements**, tap the **Log** button.

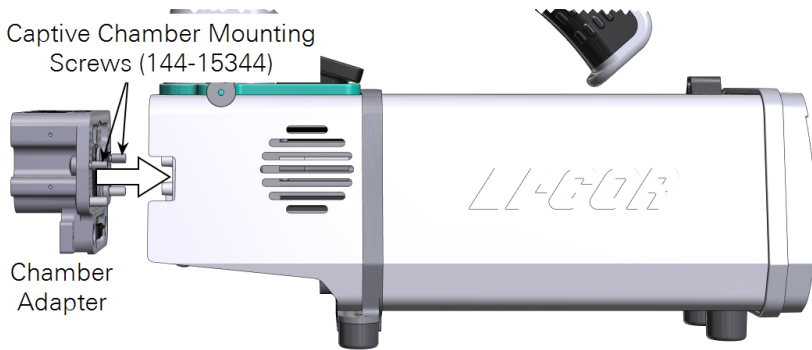
Custom chamber adapter

The Custom Chamber Interface (6800-19) is used to attach a custom-made chamber to the LI-6800 gas exchange system. The interface has a power supply connector for one powered peripheral, such as a mixing fan.

Installing the chamber adapter

See *Removing a chamber* on page 5-2 to detach a currently-installed chamber.

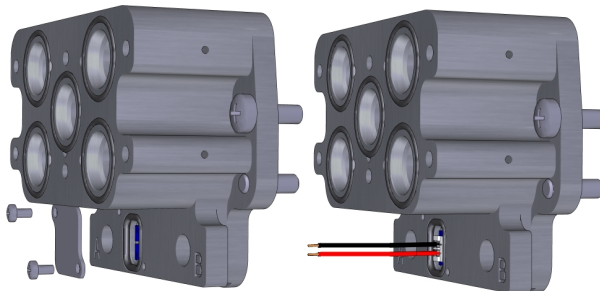
- 1 Install the chamber adapter.



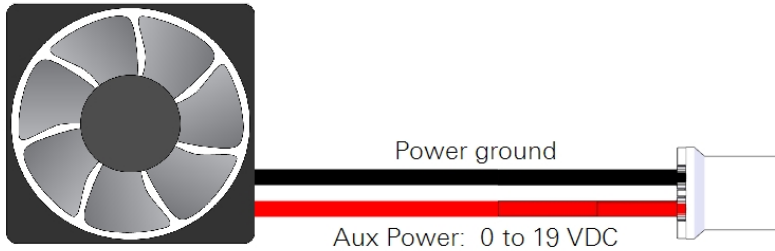
- 2 Tighten the screws snugly—until they are tight and then just a little bit more. Reinstall the connector cover.

Connecting the auxiliary power cable

If you are using the auxiliary power cable, remove the connector cover from the custom chamber interface.



Set the screws and cover aside, where they won't get lost. The power supply cable simply plugs in. Connect the leads to your mixing fan (or whatever it is powering). The red lead is positive (+) and the black is ground (-).



The auxiliary power is turned on or off in the LI-6800 interface under **Environment > Auxiliary > AuxPower**. Power can be 0 to 19 VDC.

Connecting to a custom chamber

A custom chamber can connect directly to the custom chamber interface by one of two methods: direct mount or tubing.

Flush mount

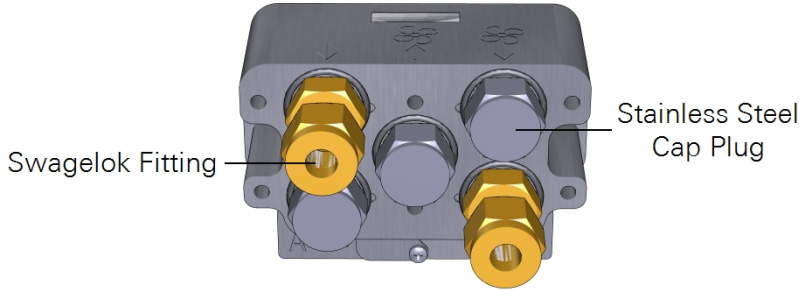
The LI-6800 sensor head can be in any orientation when directly mounted to a chamber. The chamber interface mounts directly to a flat surface. After machining the ports and mounting holes in the custom chamber, connect it to the LI-6800 head using 6 panhead screws (part number 150-14476). Be sure that each port on the chamber adapter has an O-ring. Use the *Custom chamber template* on the next page as a guide to create the interface on your chamber.

Tubing mount

Tubing can be used to connect to the chamber when the direct mount is not suitable. With tubing, you need two ports on the custom chamber—air in and air out—to be fit with Swagelok fittings. The remaining three ports should be sealed with stainless steel caps.

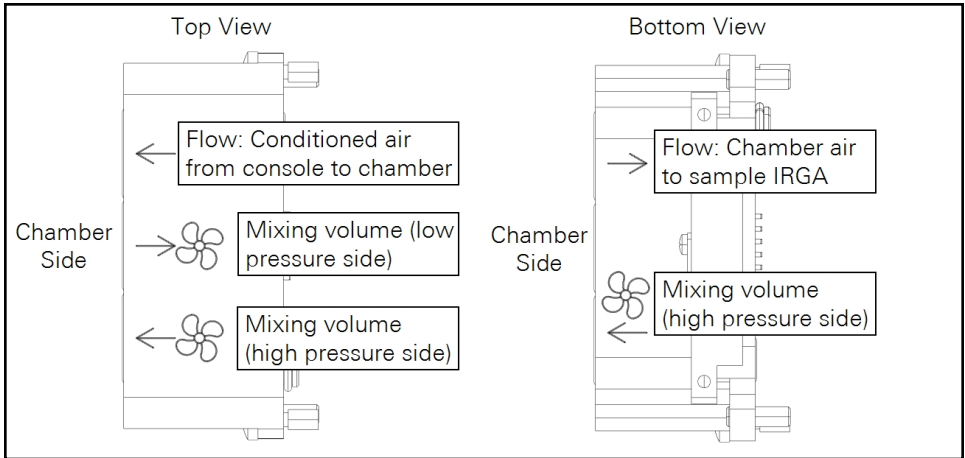
You can use the ¼" Swagelok fittings with the included Bev-a-line tubing, or the 3/8" Swagelok fittings with user-supplied tubing. In either case, you'll need to connect the tubing to the chamber. We recommend hose barbs or Swagelok fittings.

The Elbow connections from the spares kit are compatible with the 1/4" tubing, however, we recommend using vinyl ferrule sets on the Bev-a-line tubing, rather than steel or brass.



Flow and mixing indicators

The adapter is marked to indicate the function of each port. The meaning of each marking is indicated below.



Custom chamber template

The following diagrams can be used as templates to determine drilling locations on a custom chamber.

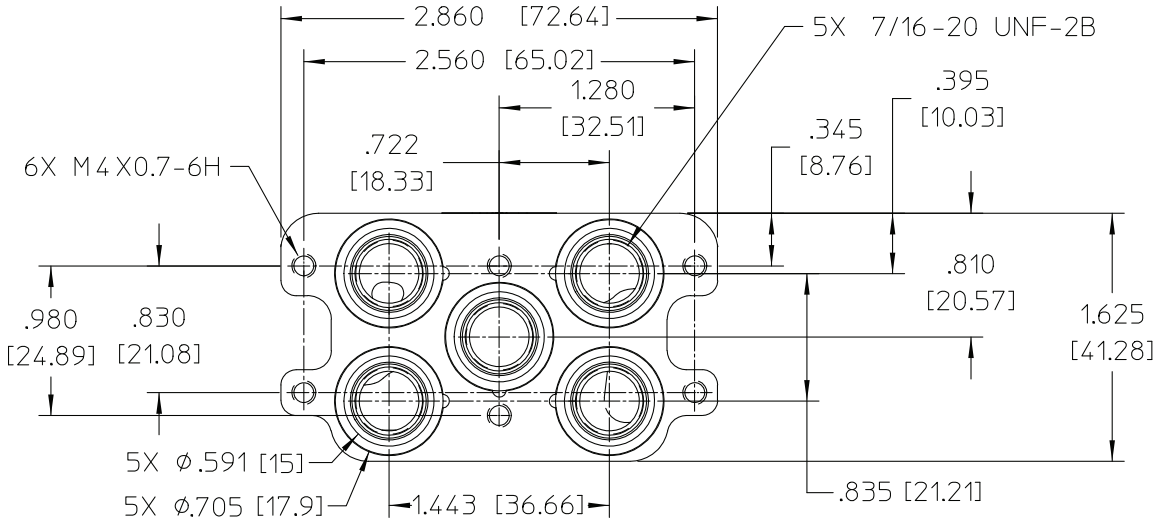


Figure 5-6. Custom chamber interface dimensions. Inches are given first, followed by millimeters in brackets.

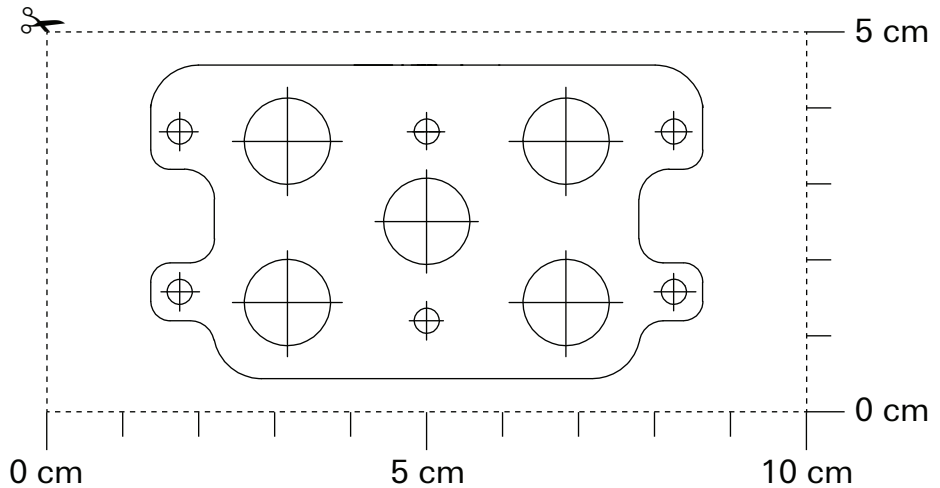


Figure 5-7. 1:1 template showing drilling locations for the custom chamber.

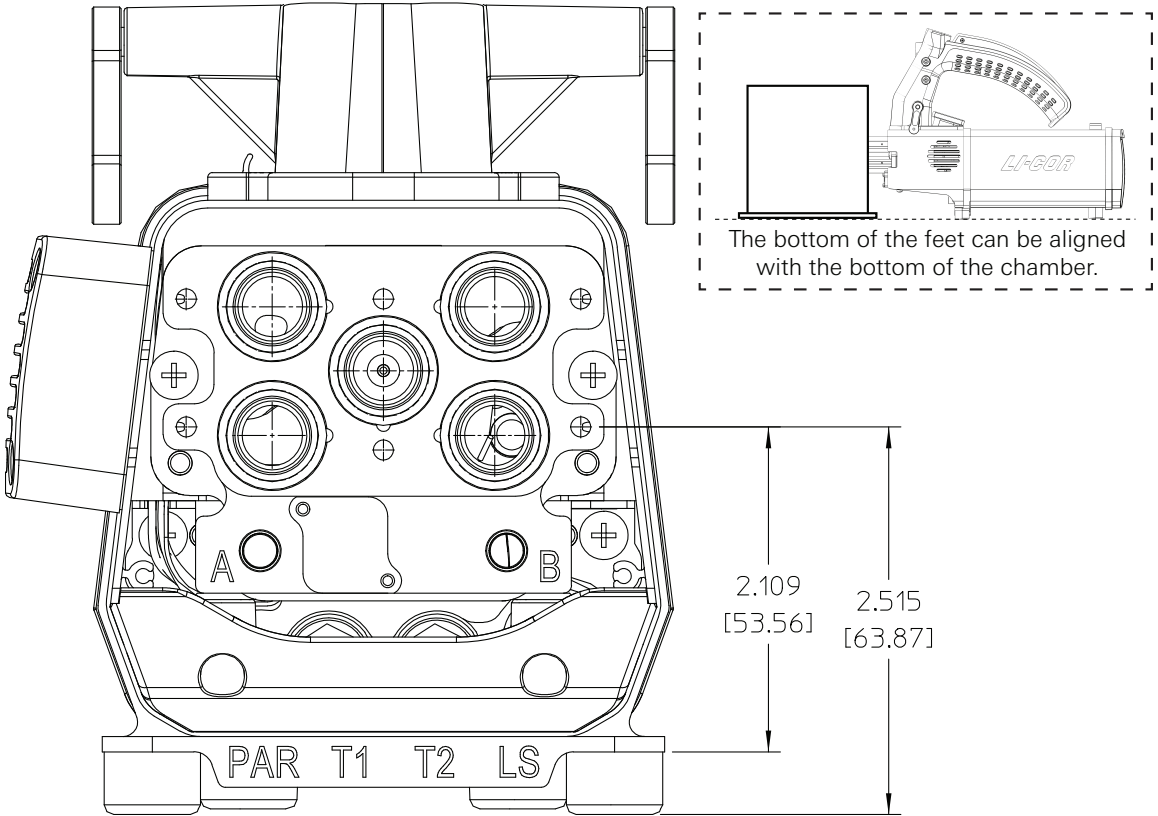


Figure 5-8. When constructing a custom chamber, it may be advantageous to align the bottom of the chamber with the bottom of the sensor head, so that both will sit evenly on a flat surface.

Section 6.

Working with files

Data files, results, and configuration information

Files and directories	6-2
Naming convention	6-2
Directories	6-2
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Working with files

This section describes how to work with data that are logged on the LI-6800. Files are accessible under the **Log Files** tab.

Files and directories

Data files are always logged as tab-delimited text files, and if selected, Microsoft® Excel® files. Text files can be opened in any text editor or imported into a spreadsheet application (you may need to change the file extension to `.txt` before importing). Text files, in contrast with the Excel files, do not have any mechanism to embed equations.

Naming convention

File names on the LI-6800 can be as long as you want to make them, and can consist of any combination of numbers, letters, spaces, and punctuation, except the forward slash character(/). File names can be upper and lower case, so the following could be distinct files:

```
logdata
LogData
LOGDATA
log_data
```

Dots (full stops) can be used as well:

```
log.data
```

Directories

Directories provide a mechanism for grouping files logically (or illogically, if you prefer). Directories can be named in any way described previously. Directories that are used for measurements are named **logs** and **firevents**.

Navigating directories

Tap a folder to open that folder. Tap **New Folder** to create a new one under the selected folder, and **New File** to create a new file in the selected directory. There is a home button that takes you to the top level.

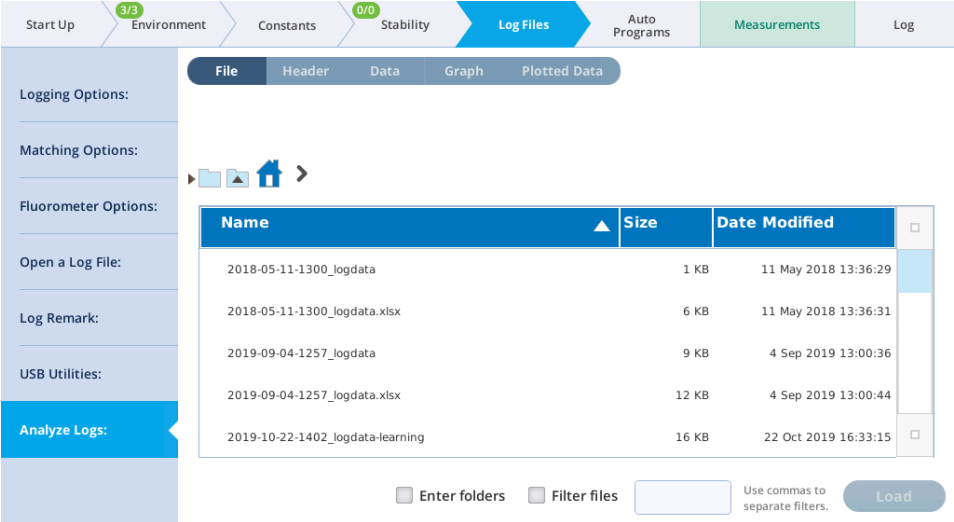
Sorting folders and files

In any of the file views, you can sort the contents of a folder by name, size, and date by tapping the corresponding sort filter. If you tap the filter again, you can sort by descending order, tap again and return to ascending order.

Managing logged data on the console

Data that are stored on the LI-6800 can be viewed at any time on the device console.

1 View files under Log Files > Analyze Logs.



Name	Size	Date Modified
2018-05-11-1300_logdata	1 KB	11 May 2018 13:36:29
2018-05-11-1300_logdata.xlsx	6 KB	11 May 2018 13:36:31
2019-09-04-1257_logdata	9 KB	4 Sep 2019 13:00:36
2019-09-04-1257_logdata.xlsx	12 KB	4 Sep 2019 13:00:44
2019-10-22-1402_logdata-learning	16 KB	22 Oct 2019 16:33:15

2 Select a file, then tap the View button.

3 After the file is opened, you can view:

- File: Tap this to view the files.
- Header: The header information from the open file.
- Data: The actual data values.
- Graph: Where you can plot variables from the data file.
- Plotted Data: The values of only the graphed data.

4 Plot the data.

Tap the **Graph** button to get a graphical view. Select the variables to plot on the horizontal axis, left axis, and right axis.

File management

A **Trash Folder** is now used to contain all deleted files and folders. Moving unwanted items to the trash can be done from any file viewer that has a **Delete** button, or by a new utility that allows you to target files using name and date filters. The trash can be browsed, and selected content easily recovered, automatically returned to its place of origin. To recover space, items in the trash can be erased, either all at once or selectively.

A new **Backup/Recovery** utility makes it easy to copy data, settings, and programs to a USB drive. To do a backup, simply insert a drive, select an optional time filter (all files before or after a certain date, or between two dates), and the desired categories: user data, configuration data, and diagnostics. Backups are archived by console serial number and date/time, making it easy to find and browse backed up data. Restoring from a backup is simple and flexible. You can select an entire backup, or narrow it down as far as you like, all the way to a particular folder or file. You also have control over what happens when restoring a file if the original still exists: overwrite it, skip it, or rename it.

For general purpose file copying between a **USB** drive and the LI-6800, there is a utility screen that lets you browse both file systems system side by side, and pick files and folders to copy, putting them exactly where you want them.

Trash

In version 1.4, when a file or directory is deleted, it is moved to a Trash Folder (`/home/licor/trash`). When items are added to the trash, folders are created as needed so that the deleted item's location in the trash matches its original location. For example, the file

```
/home/licor/logs/mydata/lightcurves/mydata.xlsx
```

would be moved to

```
/home/licor/trash/logs/mydata/lightcurves/mydata.xlsx
```

with all of the intermediate folders (`trash/logs/mydata/lightcurve`) created as needed. Mirroring the directory structure in the trash has two advantages: 1) when browsing the trash, you know where to look to find a file you deleted, and 2) all you have to do to recover an item is to select it; the system knows where to move it based on its location in the trash.

When a file is moved to the trash it will undergo a slight name change if there is a name conflict with something already there. For example, deleting `abc.txt` when there is already a previously deleted `abc.txt` in the trash at the same location will result in the moved item being renamed `abc.txt_0` (or `_1`, `_2`, etc.) to make the name unique.

Moving files to the trash

Any file viewer with a **Delete** button can move selected files to the trash (*Figure 6-1* below).

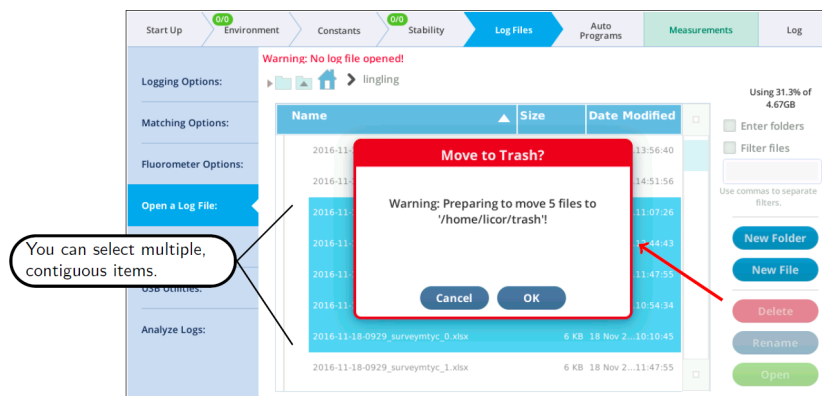


Figure 6-1. Delete buttons in file viewers move items to the trash.

Suppose you want to remove all fluorometer event `.xlsx` files that are older than some date. These are distributed in daily folders in the `/home/licor/logs/flrevents` folder, so stepping through removing them by the **Delete** button method would be very tedious.

Fortunately, there is a new utility for just this sort of purpose: it is found in the **Manage Files** screen, accessed by **Log Files > Manage Files > Trash** (see *Figure 6-2* on the next page).

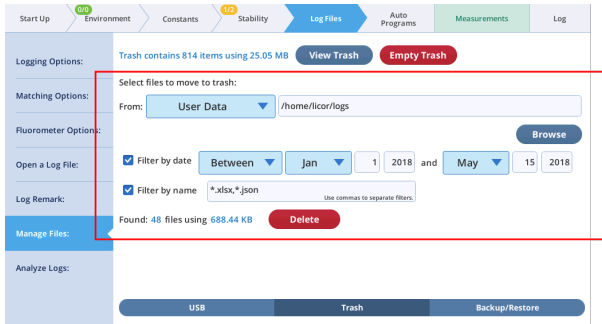
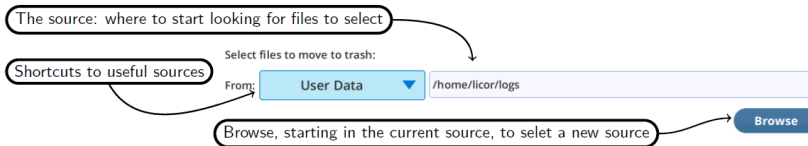


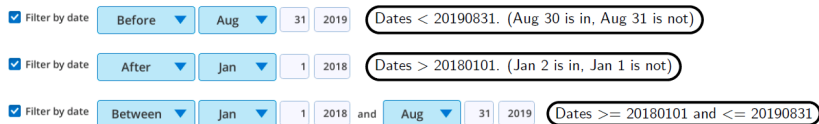
Figure 6-2. A utility for selecting files by name and date filter to move to the trash.

You can target a folder (including all sub-folders), and specify name and date filters. As you edit the target and filters, the system automatically re-scans and indicates the number of files that meet the criteria. The **Delete** button brings up a dialog that summarizes what will happen, giving you a chance to cancel or proceed.

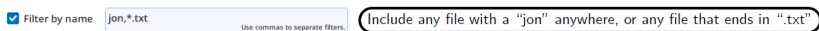
The **source folder** can be selected by editing, picking from a list, or browsing.



The **time filter** can be set to select before, after or between. The before and after are not inclusive (i.e., the date itself is excluded), and the latter is inclusive (the dates are included).



The **name filter** can use Linux wildcards (* and ?). If no wildcards are used, files that contain the filter string anywhere in their name are included. Name filters are not case sensitive. Multiple filters (they are "or-ed") are separated by comma.



The following table illustrates how the name filter behaves.

Filter(s)	File names					
	abc	xyz.txt	Ab.TXT	stXtx	aBXyxT	abc.xlsx
*	✓	✓	✓	✓	✓	✓
.		✓	✓			✓
*.txt		✓	✓			
txt		✓	✓	✓		
ab	✓		✓		✓	✓
xy, *.txt		✓	✓		✓	
????xt			✓		✓	
???.*		✓				✓

Configured for selecting: a) fluorometer event files b) older than Aug 31 c) ending in .xlsx (Excel files)

The screenshot shows a software interface with a sidebar on the left containing options like 'Logging Options', 'Matching Options', 'Fluorometer Options', 'Open a Log File', 'Log Remark', 'Manage Files', and 'Analyze Logs'. The main area displays 'Trash contains 244 items using 219.14 KB' with 'View Trash' and 'Empty Trash' buttons. Below this, 'Select files to move to trash:' is shown with a dropdown menu set to 'Fluor Events' and a path '/home/licor/logs/flrevents'. The 'Filter by date' section is set to 'Before Aug 31 2019', and 'Filter by name' is set to '*.xlsx'. A 'Delete' button is visible next to the text 'Found: 158 files using 1.33 MB'. An arrow points from the 'Delete' button to a 'File Alert' dialog box. The 'File Alert' dialog asks 'OK to move files to Trash?' and shows 'File count: 158', 'Starting directory: /home/licor/logs', 'Date filter: before 2019-8-31', and 'Name filter: *.xlsx'. It has 'Cancel' and 'Move to Trash' buttons. An arrow points from the 'Move to Trash' button to a 'Move Files To Trash' dialog box. The 'Move Files To Trash' dialog shows 'Done Moving Files', 'Files moved: 158/158', and 'Errors: 0'. It has an 'OK' button.

Figure 6-3. A utility for selecting files by name and date filter to move to the trash.

Recovering trash items

To recover items that have been sent to the trash, tap the **View Trash** button. This opens a viewer from where you can select items to recover. Multiple selection (contiguous items) is supported. Note that you can choose what to do in the case of a name duplication.

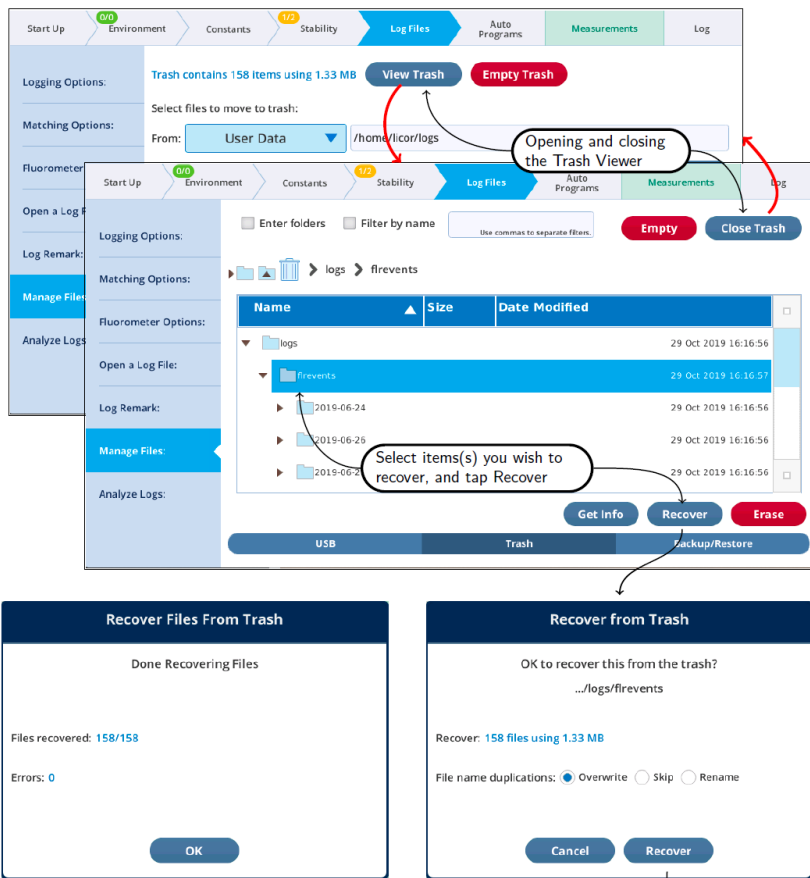


Figure 6-4. The Trash Viewer lets you view and recover items.

Depending on how you select items to be recovered from the trash, you may see empty folders afterwards (Figure 6-5 on the facing page).

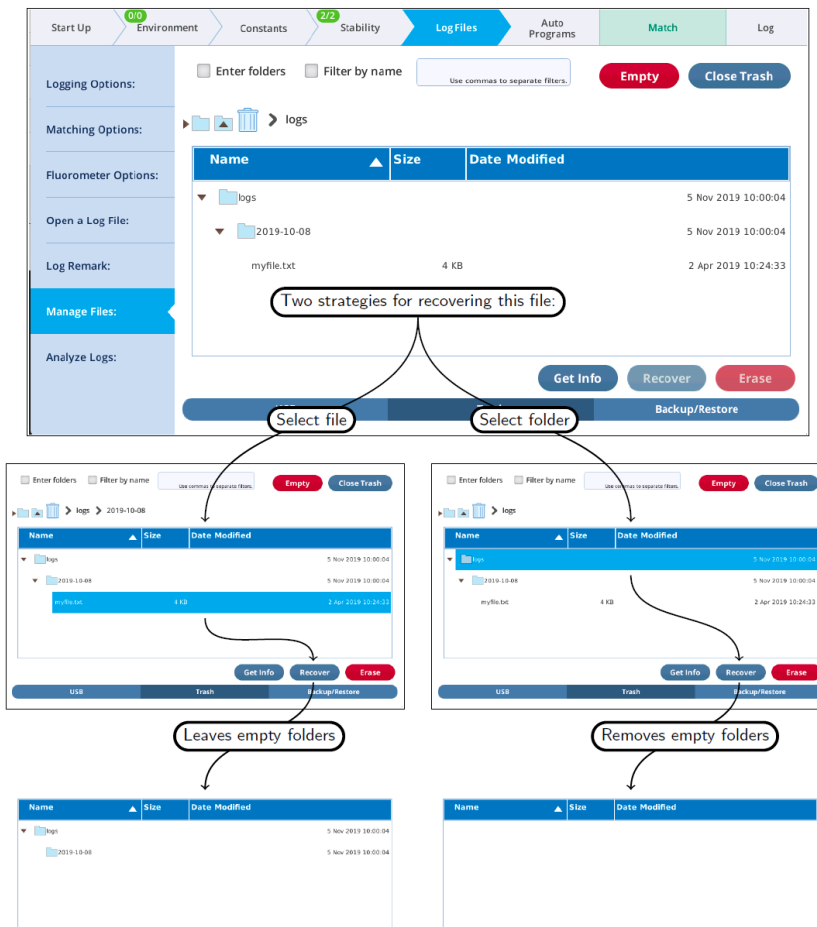


Figure 6-5. Folders emptied when trash is recovered are removed, but only if they are the targeted selection, or a child thereof.

You can remove empty folders by **Recover** or **Erase**, although they do not count as files in the ensuing dialogs ("Recover 0 files using 0 bytes"). What actually happens when "recovering" an empty folder is that the system first moves the contents (there are none, it is empty), then erases the folder if it is empty (it is).

Emptying the trash

Moving items to the trash does not free up space on the file system. To do that, items need to be erased from the trash. **Once an item is erased it can never be recovered.** To erase everything, use the **Empty Trash** button (Figure 6-6 below, top). Partial erasures can also be done (Figure 6-6 below, bottom).

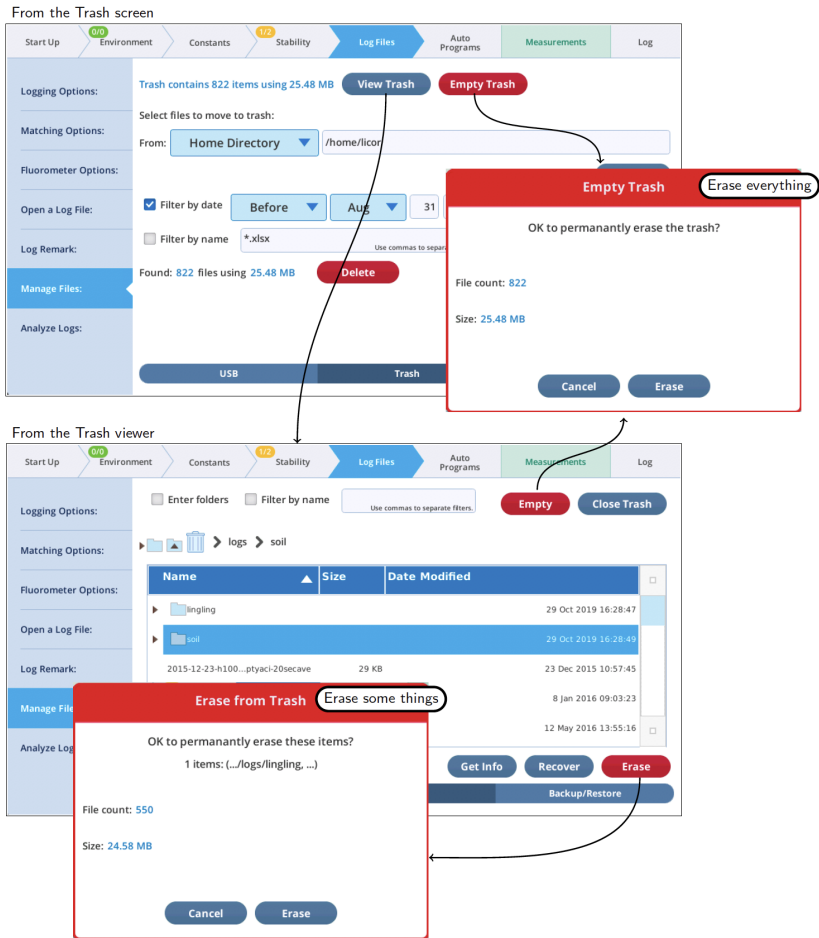


Figure 6-6. Erasing all or some of the trash. **Warning:** This can't be undone.

Another method of selectively erasing trash is to point the **Move to Trash** utility to the trash folder, and set the filters to select the files you want to permanently get rid of.

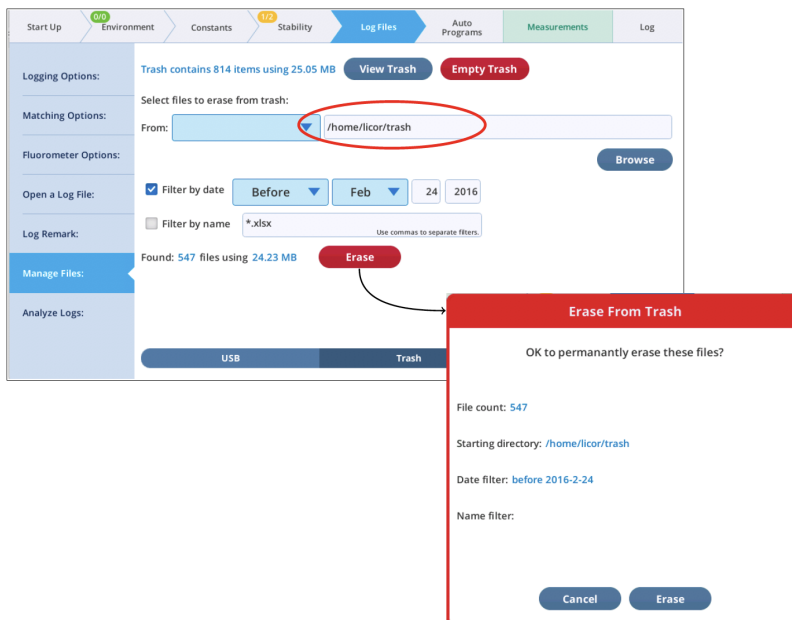


Figure 6-7. Using the move to trash utility to selectively erase items from the trash.

What's using the space?

You can get information on where file space is being used by the **Get Info** button available in many of the file browsers. For example, in the **Move to Trash** utility, set the target to the home folder (`/home/licor`), tap **Browse**, and then tap **Get Info**.

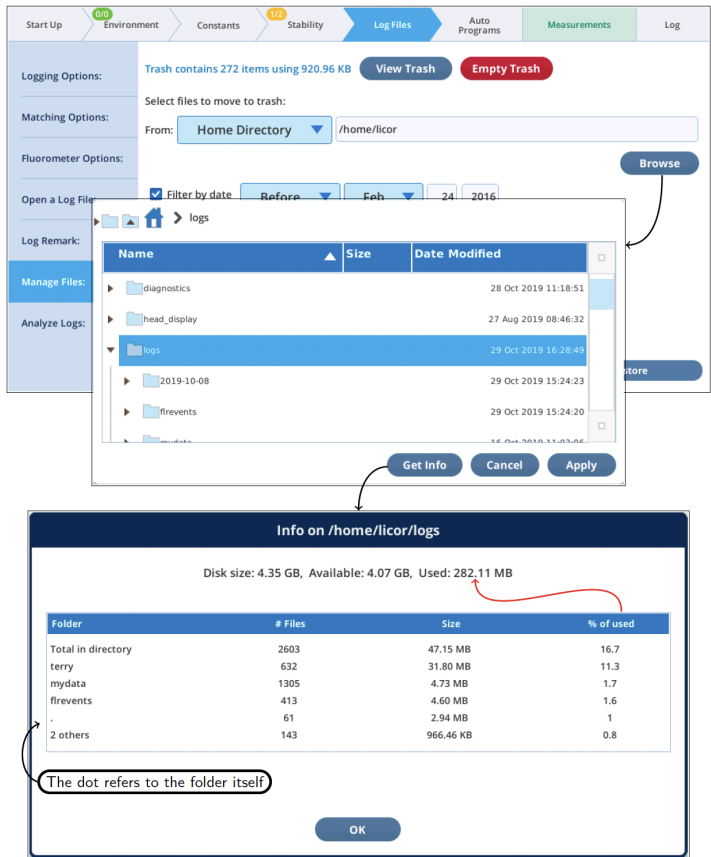


Figure 6-8. The **Get Info** display for a folder ranks its subfolders folders by size.

Backup and restore

The **Backup/Restore** page provides utilities for backing up to USB, and restoring files a USB backup to the console. Since backups created on USB are easy to navigate (folder structure is maintained), backing up is also a quick and easy way to get data files from the console to your computer.

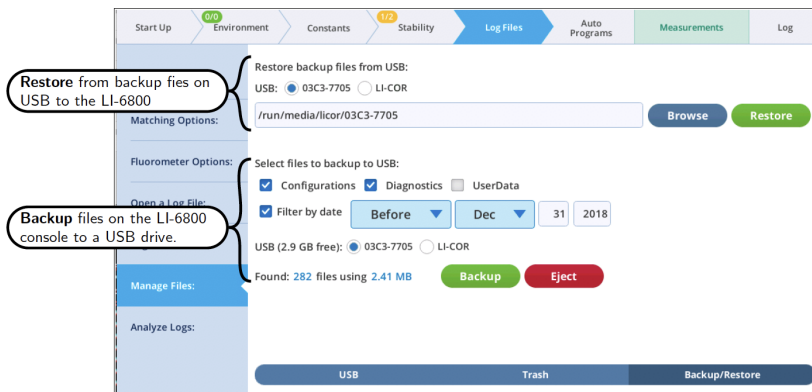


Figure 6-9. The Backup/Restore file management screens.

All backup and restore file operations are "copy", never "move". That is, backing up a file does not remove it from the console, and restoring a file from a backup does not remove it from the USB drive.

For purposes of backup, all LI-6800 console files that might need backing up are divided into three categories: Configurations, Diagnostics, and UserData.



Backups never include factory files, so - in the future - restoring from a backup with a different software version should be safe.

Backups

An example backup is illustrated in *Figure 6-10* on the next page. In this case, all categories are included, but only those files that have changed prior to the specified date.

To do a complete backup, simply a) clear the time filter box, and b) check all three file categories.

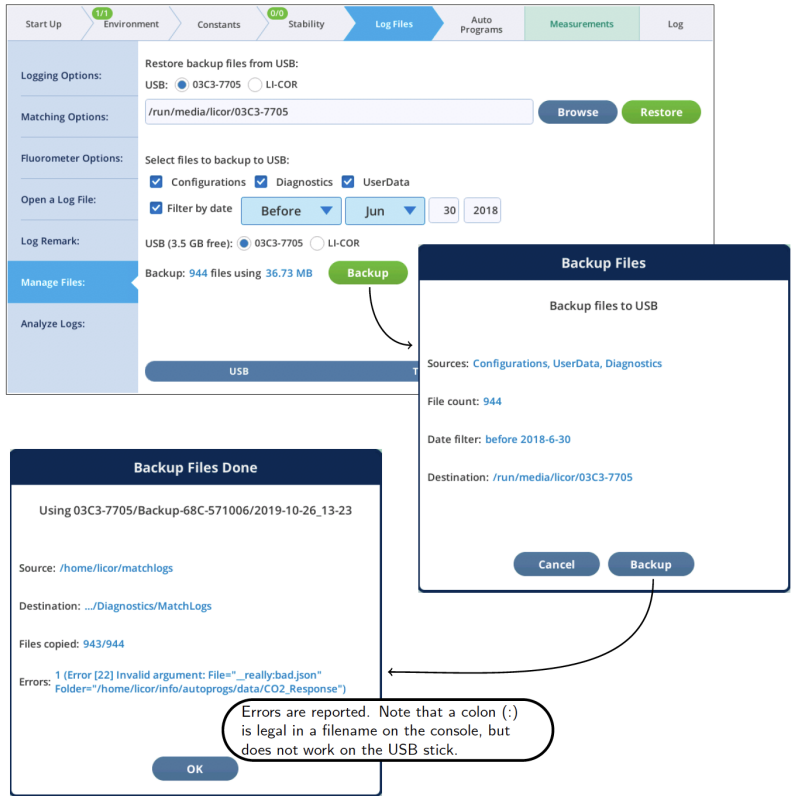
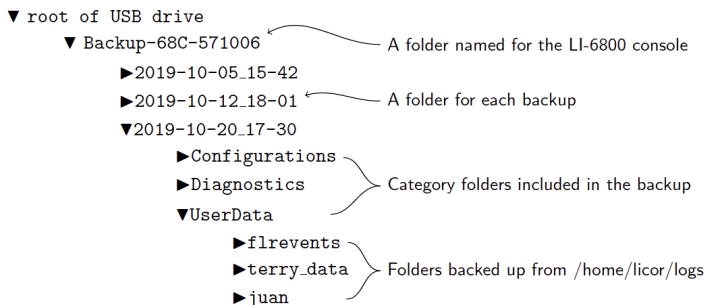


Figure 6-10. Backup and restore.

Every backup goes into a folder on the root of the selected USB drive. The folder is named for the console serial number. For example, backing up console 68C-571006 would cause a folder Backup-68C-571006 to be created on the USB drive, into which all backup would be written. Each backup event is stored in a subfolder named for the date and time the backup was started. Inside that folder will be a folder for each selected category.

Thus, after a few backups, the folder structure on the USB drive might look like the example below:



Restoring files from backup

To restore from an entire backup, tap **Restore** (Figure 6-11 below). If no Backup folders are found, then you will be notified.

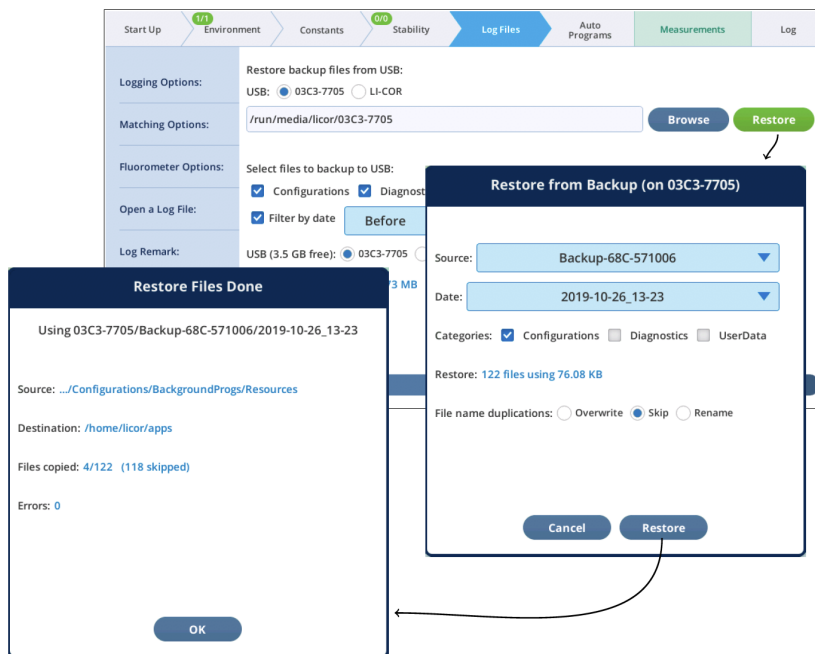


Figure 6-11. Restoring from a USB drive.

To more narrowly focus the restore from backup, follow these steps (Figure 6-12 on the next page):

- 1 Use **Browse** to open a viewer
- 2 Navigate to the desire folder or file and select it
- 3 Tap **Apply**
- 4 Tap **Restore**

Note that if you navigate in step 2 to somewhere that is not within the parentage of a backup folder, your selection will be ignored.



Figure 6-12. Selecting a specific folder or file to restore.

Transferring files to a computer

You can transfer files by copying them to a USB storage device, or you can transfer them to a computer or a computer network using the Ethernet connection, which puts the LI-6800 on a network as though it is a computer. Data files can be downloaded from the LI-6800 console using either Windows Explorer on Windows computers, or using the Finder on Macs. You can also use SFTP software to transfer files.

Using a USB storage device

The simplest way to transfer files is to copy them onto a USB storage device. You'll need a USB flash drive with enough empty space to save the files (external hard drives may work but we have not tested them and do not promise compatibility).

- 1 Plug the USB device into one of the USB ports on the back of the LI-6800.
- 2 Under **Log Files > Export Logs**, tap the **Refresh** button.

Now you can select individual files to copy or select all files.

- 3 Tap **Copy to USB**.

After a moment the files will be visible on the USB device list. This indicates that they have been copied successfully.

- 4 Eject the USB device after all the files are copied.

Using a network connection

File transfer over a network is compatible with:

- Windows 7, 8, 8.1, 10
- Mac OS 10 and newer

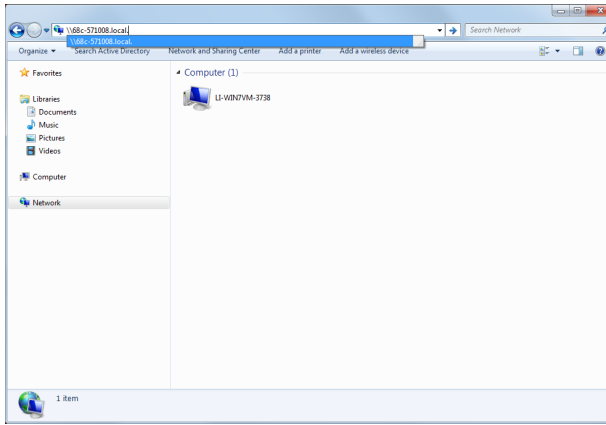
To transfer data, prepare the following:

- Locate the console serial number (labeled SR. NO.) on the label behind the CO₂ cartridge holder.
- Connect the instrument to your computer or network using the Ethernet cable.
- Power on the instrument.

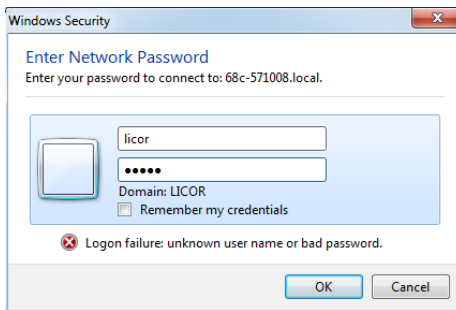
On the Windows operating system

- 1 Open an **Explorer** window. Double click, for example, on **Computer**.
- 2 Enter two backslashes (\) followed by the console serial number into the address bar. You'll enter **\\68c-#####**.

In some network configurations you may need to use **\\68c-#####.local**. instead.

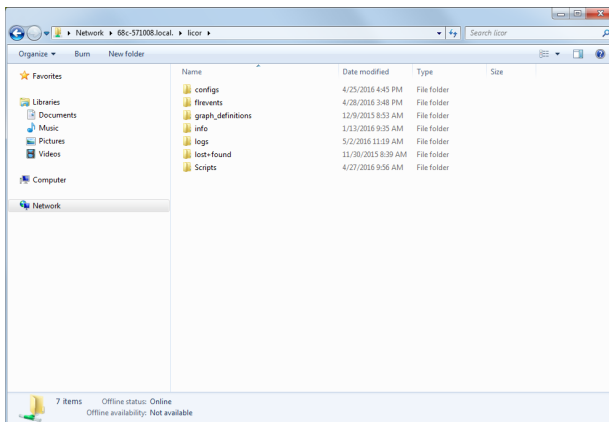


- 3 Log in when prompted.
 - **User name:** licor
 - **Password:** licor



- 4 There is one folder called 'licor'.

You can open it by double clicking, or map it to a drive letter.

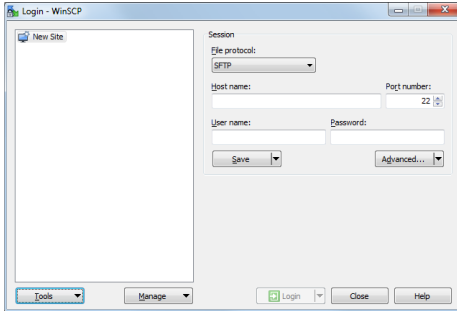


- Gas exchange data are in the **logs** folder (see *What's in the logs folder* on page 6-25).
- Fluorescence data are in the **firevents** folders (see *What's in the firevents folder* on page 6-26).

With WinSCP

WinSCP is a freely downloadable Windows application that supports the secure file transfer protocol (SFTP). It is a reliable alternative if you are having trouble making Windows recognize the LI-6800. For example, when we plug an LI-6800 into the LAN here at LI-COR, it can sometimes take several minutes before it becomes “visible” in the Microsoft Windows Network. WinSCP, on the other hand, will find it immediately. You'll need to download the WinSCP software and install it on your computer. It is available at <http://winscp.net/eng/download.php>.

1 Open the WinSCP program.



2 Select **New Site** and enter the following.

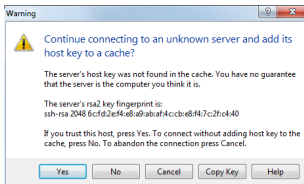
- **File protocol:** SFTP
- **Host name:** Enter the instrument serial number.
The serial number is similar to **68c-#####**.
In some network configurations you may need to use **68c-#####.local**. instead.
- **Port number:** 22
- **User name:** licor
- **Password:** licor

Save the connection if you'll be using it a lot.

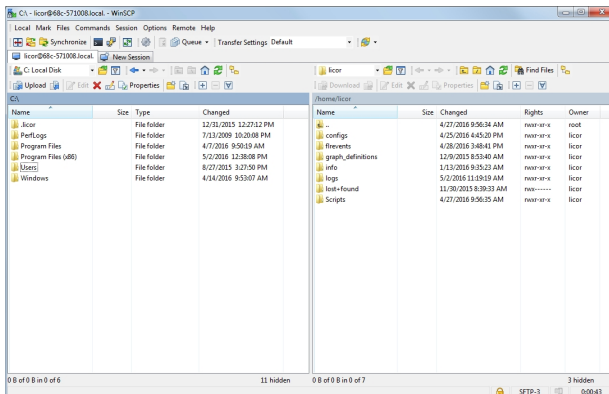
3 Click **Login**.

The login screen will momentarily appear. If WinSCP requires another password (if you saved the connection but not the password), enter the password and click **OK**.

4 If a warning about an unknown server appears, click **Yes**.



5 Data are in the **logs** and **firevents** folders.

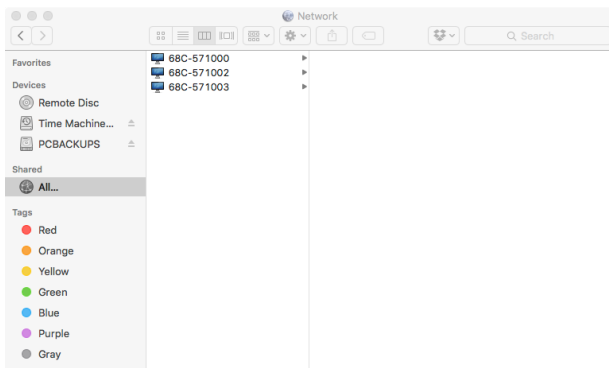


You can map it to a drive letter and it will show up in the same place as your other drives.

- Gas exchange data are in the **logs** folder (see *What's in the logs folder* on page 6-25).
- Fluorescence data are in the **firevents** folders (see *What's in the firevents folder* on page 6-26).

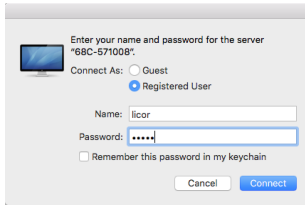
On the Mac operating system

- 1 Open a new Finder window and select **All...** from the **Shared** section.



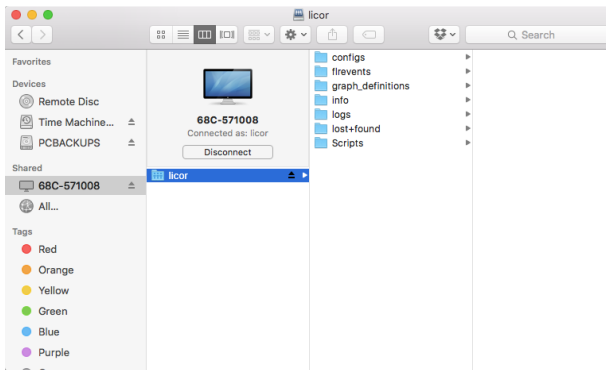
You'll see a list of the instruments on your network. Names are 68C-##### or something similar.

- 2 Select the instrument that you want to access.
- 3 Log in when prompted.



- **User name:** licor
- **Password:** licor

It may take a few minutes for the directories to be visible on your computer.



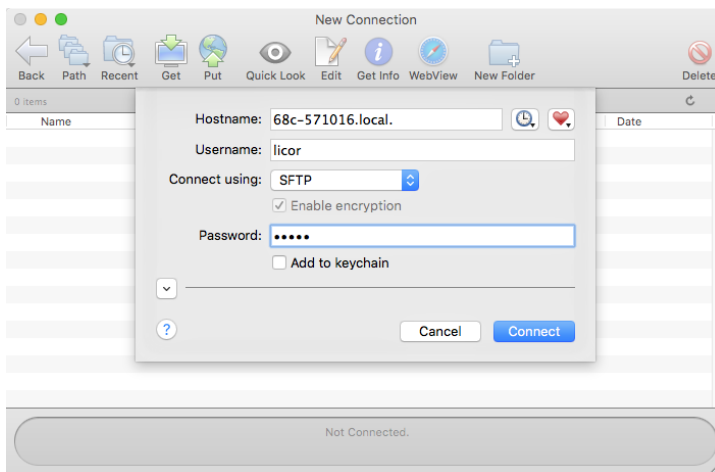
- Gas exchange data are in the **logs** folder (see *What's in the logs folder* on page 6-25).
- Fluorescence data are in the **frevents** folders (see *What's in the frevents folder* on page 6-26).

With Fetch FTP

Fetch is an inexpensive file transfer application for Mac OS that supports SFTP through a simple graphical interface. Other SFTP applications, such as CyberDuck, are configured similarly.

1 Run Fetch.

The **New Connection** window is where you establish the connection.



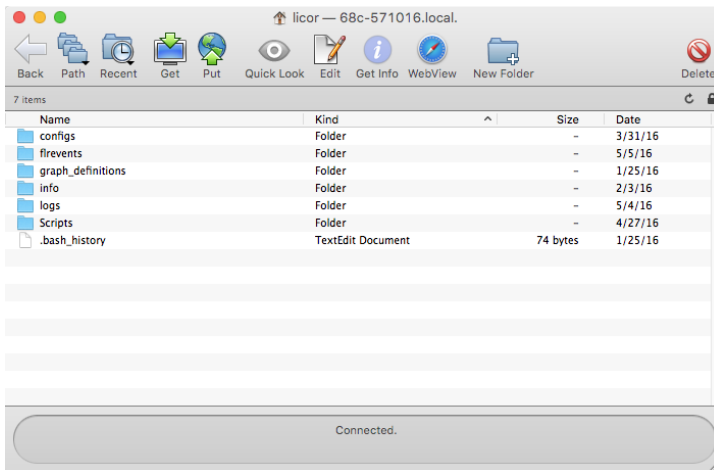
2 Enter the following.

- **Host name:** Enter the instrument serial number.
The serial number is similar to **68c-#####**.
In some network configurations you may need to use **68c-#####.local**. instead.
- **Username:** licor
- **Connect using:** SFTP
- **Password:** licor

3 Click Connect.

4 Explore, Drag, Drop, etc.

Once you are logged in, the LI-6800's file system will appear in the Fetch window. You can drag, drop, and otherwise manipulate files.



- Gas exchange data are in the **logs** folder (see *What's in the logs folder* on the facing page).
- Fluorescence data are in the **firevents** folders (see *What's in the firevents folder* on page 6-26).

Folders

The LI-6800 stores data files and configuration information in folders, similar to a typical computer operating system. You can create, rename, and delete files and folders directly from the instrument console; and you can create, rename, delete, and copy files and folders when connected to the instrument with a computer.

For the well-being of your instrument (and for good relationships with others who use the instrument), we recommend that you leave the files and folders alone unless you know exactly why they are on the instrument. You can freely delete any data files in the **logs** and **firevents** folders, however, if you share the instrument with other people or labs, be aware that your haphazard labmates might be storing the only copy of precious data on the instrument. So, use discretion when deleting unknown files. Once you delete a file, it is gone and not retrievable. Gone. For good. Like last year. You can't get it back. So be careful and backup your files regularly.

The other folders contain configuration files, graph definitions, and important system-related files. You can delete the configuration and graph definition files, but

again, your labmates might be annoyed if you dispose of their hard work. As a good practice, regularly back up your own configurations and graph definitions for safe-keeping from any compulsive file cleaners. The **info** and **scripts** folders are related to the operating system and instrument. Leave them alone.

What's in the logs folder

Data files are tab-delimited text files, and if you selected the option, .xlsx files. Each file includes a header—which is information that describes the rest of the file. The contents of the header will depend on whether you selected variables in **Check to log as a row in Logging options** (see *Check to log as a row* on page 3-84). Underneath the header is the data from each measurement. Here again, the columns included depend on the variables you choose. You will find some or all of the following:

Header label	Description	Variables table
Const	Gas exchange constants	Table 6-4 on page 6-31
FLR	Fluorometer computations	Table 6-5 on page 6-31 ¹
FlrLS	Fluorometer light source data	Table 6-6 on page 6-32 ¹
FlrStats	Fluorometer statistics	Table 6-7 on page 6-33
GasEx	Gas exchange variables	Table 6-8 on page 6-33
LQConst	Light source constants	Table 6-10 on page 6-34
LTConst	Leaf temperature constants	Table 6-11 on page 6-35
LeafQ	Light	Table 6-12 on page 6-35
MPF	MultiPhase flash values	Table 6-13 on page 6-35 ^{1,2}
Match	Match data	Table 6-16 on page 6-37 ²
Meas	Measured variables	Table 6-17 on page 6-37
Meas2	Additional measured data	Table 6-18 on page 6-38 ²
QConst	Light source constants	Table 6-19 on page 6-38
Raw	Raw data	Table 6-20 on page 6-39 ²
Stability	Stability parameters	Table 6-21 on page 6-39 ²
Status	Status data	Table 6-22 on page 6-40
Status2	Additional status information	Table 6-23 on page 6-41 ²
Sys	System variables	Table 6-24 on page 6-42
UserDef	User defined constants	

¹Included if the Multiphase Flash Fluorometer is connected.

²Included if selected under Logging Options.

Remarks in the log files

- **In text data files:** Remarks are recorded in the text data file, either in the header information or inline with the other measurements. Remarks will always include the time stamp.
- **In Excel files:** Remarks are in a separate tab from the measurements. The tab is called Remarks. Remarks will always include the time stamp.

What's in the flrevents folder

Data files for each flash made by the fluorometer are stored in the instrument's memory. Inside the folder you'll see text files, and possibly .xlsx files with names like the following examples:

```
RECT-3-20160217-16_54_29  
MPF-4-20160126-16_45_11  
INDUCTION-5-20160202-21_42_17  
DARK-6-20160202-21_42_23  
RCRDG-6-20161112-11_24_53
```

In each of these file names, the first set of capital letters indicates the flash type.

- **RECT** is for rectangular flash
- **MPF** is for a multiphase flash
- **INDUCTION** is an induction event
- **DARK** is for the dark pulse.
- **RCRDG** is the continuous fluorescence trace.

The first digit is the flash number. Each flash made by the fluorometer is assigned a number, and the number increments once per measurement. The remaining numbers are the date and time stamp, following this convention: `yyyymmdd-hh_mm_ss`. Year is indicated by `yyyy`, `mm` is the month, `dd` is day, `hh` is hour, `mm` is minute, `ss` is second.

Interpreting a flash data set

If you open a rectangular flash file in Microsoft Excel, you will see something like this:

	A	B	C	D	E	F	G	H
Fluorometer	1	EVENT_ID	2	SECS	FLUOR	DC	PFD	CODE
Serial Number	2	DEVICE	MPF-551005	0	0.614	-109.986	24.9985	2
	3	DATE	20160217	0.002	-0.61	-107.434	24.9985	2
	4	TIME	16:54:24	0.004	-0.758	-106.01	24.9985	2
Fluorometer	5	TYPE	RECT	0.006	-1.532	-106.308	24.9985	2
Settings	6	OUTRATE	500	0.008	0.66	-108.618	24.9985	2
	7	MARGIN	5	0.01	-0.426	175.348	4082.08	3
	8	DURATION	1000	0.012	-1.116	185.838	4081.94	3
	9	Q_RED_SETPOINT	4000	0.014	0.35	188.132	4081.89	3
	10	D_RED_PERCENT	100	0.016	-0.376	186.402	4081.81	3
	11	FMAX	1.574	0.018	0.16	185.968	4081.77	3
	12	QMAX	4082.08	0.02	0.28	187.678	4081.72	3
	13	MODRATE	250000	0.022	-0.296	190.32	4081.7	3
	14			0.024	-0.342	188.28	4081.7	3
	15			0.026	-1.202	187.078	4081.58	3
	16			0.028	-0.418	188.396	4081.55	3
	17			0.03	-1.484	190.476	4081.52	3
	18			0.032	-0.608	188.992	4081.48	3
	19			0.034	-1.3	187.15	4081.46	3

The left two columns are information about the flash event. Columns D to H are the fluorescence data.

Table 6-1. Each flash will have its own file, which includes the following pieces of information.

Column	Meaning
Secs	Time
Fluor	
DC	Actinic light
PFD	Photon Flux Density
Code	The sequence of the flash that is underway at the time. <ul style="list-style-type: none"> • 2 is the beginning margin • 3 is the rectangular flash • 4 is Phase 1 of a multiphase flash • 5 is Phase 2 of a multiphase flash • 6 is Phase 3 of a multiphase flash • 7 is the end margin

If you open a typical multiphase flash data set, expect something like this:

	A	B	C	D	E	F	G	H
1	EVENT_ID		1	SECS	FLUOR	DC	PPD	CODE
2	DEVICE	MPF-551005		0	-0.664	252.43	24.9985	2
3	DATE	20160209		0.002	-0.894	253.38	24.9985	2
4	TIME	21:26:27		0.004	0.312	251.944	24.9985	2
5	TYPE	MPF		0.006	0.082	251.244	24.9985	2
6	OUTRATE	500		0.008	-0.632	251.966	24.9985	2
7	MARGIN	5		0.01	1.144	838.506	8169.37	4
8	PHASE1_DURATION	300		0.012	-1.214	852.652	8169.15	4
9	PHASE2_DURATION	300		0.014	-0.788	853.55	8168.91	4
10	PHASE3_DURATION	300		0.016	-0.31	854.67	8168.69	4
11	Q_RED_SETPOINT	8000		0.018	-0.132	856.482	8168.51	4
12	D_RED_PERCENT	0		0.02	-1.14	857.452	8168.36	4
13	RAMP	30		0.022	-0.386	855.74	8168.17	4
14	FMAX	-0.265718		0.024	-1.202	857.65	8168.01	4
15	QMAX	8169.37		0.026	-0.452	856.572	8167.83	4
16	MODRATE	250000		0.028	0.768	857.064	8167.7	4
17	P1_MAXF	1.466		0.03	1.332	855.452	8167.51	4
18	P2_SLP	-0.0371314		0.032	0.49	855.024	8167.33	4
19	P2_INT	-0.265718		0.034	-0.504	856.074	8167.16	4
20	P2_R2	4.25728E-05		0.036	-0.566	856.202	8166.99	4
21	P2_SLP_SE	0.467773		0.038	0.38	854.53	8166.88	4
22	P2_INT_SE	0.685604		0.04	-0.868	853.964	8166.67	4
23	P2_QDDT	-0.00815425		0.042	1.4	854.162	8166.54	4
24	P3_Q	8155.94		0.044	-0.436	853.882	8166.36	4
25	P3_MAXF	1.692		0.046	-0.692	853.674	8166.25	4
26	P3_PREDF	-0.270271		0.048	0	852.872	8166.11	4
27	P3_DELTAF	1.96227		0.05	-1.708	854.192	8165.95	4
28				0.052	-0.75	852.644	8165.83	4
29				0.054	0.162	851.76	8165.66	4

The continuous fluorescence trace

When you record the fluorometer trace (**Environment > Fluor > Measuring**), the resulting data file will be stored in the `/home/licor/flrevents` directory and the name will begin with `RCRDG...` followed by the date and time that recording began. The resulting file is tab-delimited. The data frequency will be about 2 Hz, except during flash or dark events, in which case it is determined by the output frequency that you specified for the event.

```

CODE TIME FLUOR DC PFD RED BLUE FARRED REDMODAVG
1 1478561051.8 1.80625 113.31 100.013 89.996998089 10.016001911 0.0 1.99973
1 1478561052.4 1.6265 113.091 99.983 90.004796583 9.978183420399999 0.0 1.99973
1 1478561053 1.699 113.041 100.022 90.005496854 10.016503146 0.0 1.99973
1 1478561053.6 2.296 112.79 100.015 89.998797795 10.016202205 0.0 1.99973
1 1478561054.2 2.14475 112.784 99.9829 90.0047065629 9.978173440519999 0.0
1.99973
1 1478561054.8 1.388 113.628 100.01 89.994298530000001 10.01570147 0.0 1.99973
1 1478561055.4 2.45675 113.051 99.9943 90.0146688714 9.97968112575 0.0 1.99973
1 1478561056 1.97575 113.334 99.9834 90.0051566634 9.97822333992 0.0 1.99973
1 1478561056.6 1.7955 112.926 99.9909 90.0119081709 9.978971830919999 0.0
1.99973
:

```

Listing 6-1. The start of the RCRDG... file.

Table 6-2. Items in a fluorescence recording (RCRDG...) file.

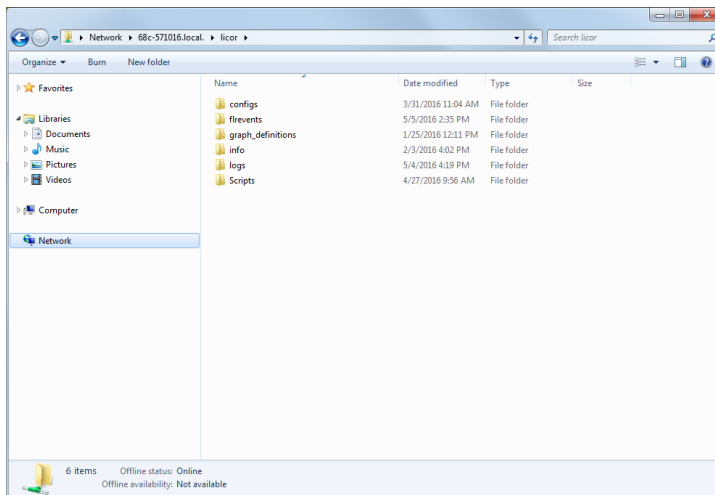
Label	Description
CODE	Record identifier (see <i>Table 6-3</i> below)
TIME	Seconds
FLUOR	AC fluorescence
PDF	Total photon flux density ($\mu\text{mol m}^{-2} \text{s}^{-1}$)
RED	Red (non-modulated) photon flux density ($\mu\text{mol m}^{-2} \text{s}^{-1}$)
BLUE	Blue photon flux density ($\mu\text{mol m}^{-2} \text{s}^{-1}$)
FARRED	Far red flux fluorescence ($\mu\text{mol m}^{-2} \text{s}^{-1}$)
REDMODAVG	Averaged photon flux density contributed by the modulation ($\mu\text{mol m}^{-2} \text{s}^{-1}$)

Table 6-3. CODE values in a fluorescence recording file.

CODE	Description
0	Off
1	Normal measuring
2	Pre-flash
3	Rectangle flash
4	MPF, phase 1
5	MPF, phase 2
6	MPF, phase 3
7	Post-flash
11	Pre-dark
12	Dark pulse, Far Red ON (actinic still on)
13	Dark pulse, Actinic off (far red still on)
14	Dark pulse, all off
15	Post-dark

Configuration files, graph definition files, and assorted miscellany

In addition to the data files, the LI-6800 file system includes configuration files, graph definitions, and other files that are either logged information, settings (saved as plain text files), and instructions for the instrument (saved as .py and .json files). You can access all of these when connected to the instrument's file system.



We're interested in the settings—the contents of the **configs** folder. Since these are plain text files, you can open and edit them in any text editor, so feel free to explore them on your own if you are so inclined.

Copying configuration files is just like moving files in any other computer file system. You'll save the files to your computer, and then load them onto another instrument by placing them in the same directory that they were in originally.

Snapshots

Snapshots are stored in the directory `/home/licor/info/snapshots`, and are named for the date and time when they occurred. They are .json files, consisting of the latest values for each of a number of topics at the time of the snapshot.

Summary of symbols

The software presents a scheme for identifying and categorizing variables into groups. Variables displayed on the screen or in a data file will be labeled with a group name and a display label. The groups and display labels are shown below. Labels are only used for display or log file purposes, so they can be changed without breaking any internal software.

Gas exchange constants

Table 6-4. Gas exchange constants (called **Const** in the data file). For each constant, you can specify whether it is logged in a data file column, or as part of the header.

Display Label	Units	Description	Actual Name
ChType		Chamber	ChType
Custom	$\text{mol m}^{-2} \text{s}^{-1}$	1-sided boundary layer conductance for custom	CustomBLC
Geometry		Chamber	Geometry
K		Stomatal ratio	K
Oxygen	percent	Oxygen concentration	Oxygen
S	cm^2	Leaf area	S

Fluorometer variables

Table 6-5. Light source variables for the fluorometer (called **FLR** in the data file).

Display Label	Units	Description	Actual Name
A_fs	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Assimilation rate corresponding with the last Fs measurement	A_fs
Adark	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Dark assimilation rate before Fm	Adark
DarkAdaptID		ID of latest dark-adapted flash	DarkAdaptedID
DarkPulseID		ID of latest dark pulse	DarkPulseID
ETR	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Electron transport rate	ETR
Fm		Ratio	
Fm'		Maximal F, light-adapted	Fmp
Fo		Ratio	Fo
Fo'		Minimal F, light-adapted	Fop
Fs		Steady state F	Fs

Table 6-5. Light source variables for the fluorometer (called FLR in the data file). (...continued)

Display Label	Units	Description	Actual Name
Fv		Variable fluorescence Fm - Fo	Fv
Fv/Fm			Fv/Fm
Fv'			Fv'
Fv'/Fm'			Fv'/Fm'
LightAdaptedID		ID of the latest light-adapted flash	LightAdaptedID
NPQ		(Fm - Fm')/Fm'	NPQ
PS2/1		Ratio of PS2 to PS1	PS2/1
PhiCO2		(A - Ao)/Qabs	PhiCO2
PhiPS2		1 - Fs/Fm'	PhiPS2
Qabs_fs	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Absorbed light corresponding to the last Fs measurement	Qabs
qN		(Fm - Fm')/(Fm - Fo)	qN
qN_Fo		(Fm - Fm')/Fm - Fo	qN_Fo
qP		(Fm' - Fs)/(Fm' - Fo)	qP
qP_Fo		(Fm' - Fs)/(Fm' - Fo)	qP_fo

Fluorometer light source variables

Table 6-6. Light source values for the source attached to the console (called FirLS in the data file). If a light source is attached when a data file's first observation is logged, then all of this group is logged in data file columns.

Display Label	Units	Description	Actual Name
f_blue	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Fraction blue; $f_{blue} = \frac{Q_{blue}}{Q_{red} + Q_{blue} + Q_{modavg}}$	f_blue
F_dc		DC fluorescence	DC
f_farred	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Fraction far red	FARREDFRAC
F		Demodulated fluorescence	FLUOR
Q	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Fluorometer photon flux density	PFD
f_red	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Fraction red; $f_{red} = \frac{Q_{red} + Q_{modavg}}{Q_{red} + Q_{blue} + Q_{modavg}}$	f_red
Q_modavg	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Modulated red contribution to Q	REDMODAVG
Q_red	$\mu\text{mol m}^{-2} \text{s}^{-1}$		
Q_blue	$\mu\text{mol m}^{-2} \text{s}^{-1}$		
Q_farred	$\mu\text{mol m}^{-2} \text{s}^{-1}$		

Fluorometer statistics

Table 6-7. Statistics related to the fluorometer (referred to as **FlrStats** in the data file).

Display Label	Units	Description	Actual Name
F_avg		Average F	F_avg
F_dc_avg		Average F_dc	F_dc_avg
dF/dt	min ⁻¹	Rate of change of F	dF/dt
dF_dc/dt	min ⁻¹	Rate of change of F_dc	dF_dc/dt
period	s	Period for statistics	period

Gas exchange variables

Table 6-8. Computed gas exchange variables (called **GasEx** in the data file). All of these are logged in data file columns.

Display Label	Units	Description	Actual Name
A	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Assimilation rate (corrected for leaks)	A
Ci	$\mu\text{mol mol}^{-1}$	Intercellular CO ₂	Ci
Ca	$\mu\text{mol mol}^{-1}$	Ambient CO ₂ (in the chamber)	Ca
E	$\text{mol m}^{-2} \text{s}^{-1}$	Transpiration rate (corrected for leaks)	E
Pca	Pa	Chamber CO ₂	Pca
Pci	Pa	Intercellular CO ₂	Pci
RHcham	%	Relative humidity in the chamber	RHcham
Rabs	W m^{-2}	Absorbed short wave radiation	Rabs
SVPcham	kPa	Saturation vapor pressure in the chamber	SVPcham
SVPlleaf	kPa	Saturation vapor pressure at leaf temperature	SVPlleaf
Time	s	Epoch time; seconds since January 1970	TIME
TleafCnd	°C	Leaf temperature used for gas exchange computations	TleafCnd
TleafEB	°C	Leaf temperature from energy balance	TleafEB
VPDleaf	kPa	Vapor pressure deficit at leaf temperature	VPDleaf
VPcham	kPa	Vapor pressure in the chamber	VPcham
blfa_1		Parameter 1 for boundary layer as a function of area	blfa_1

Table 6-8. Computed gas exchange variables (called GasEx in the data file). All of these are logged in data file columns. (...continued)

Display Label	Units	Description	Actual Name
blfa_2		Parameter 2 for boundary layer as a function of area	blfa_2
blfa_3		Parameter 3 for boundary layer as a function of area	blfa_3
gbw	$\text{mol m}^{-2} \text{s}^{-1}$	Boundary layer conductance to water vapor	gbw
gsw	$\text{mol m}^{-2} \text{s}^{-1}$	Stomatal conductance to water vapor	gsw
gtc	$\text{mol m}^{-2} \text{s}^{-1}$	Total conductance to CO_2	gtc
gtw	$\text{mol m}^{-2} \text{s}^{-1}$	Total conductance to water vapor	gtw

Leak variables

Table 6-9. The leak variables are always logged in a file called **Leaks**.

Display Label	Units	Description
Leak	$\mu\text{mol s}^{-1}$	Leak from chamber ($50 \mu\text{mol s}^{-1}$)
LeakPct	%	Leak as a percentage of the flow
CorrFact		Correction factor (K in equations below)
CorrFactPct	%	Correction factor expressed as a percentage
Mix		Chamber-based correction factor to tune the leak correction
Fan	$\mu\text{mol s}^{-1}$	Fan flow rate

More light source constants

Table 6-10. Constants pertaining to leaf absorption (called **LQConst** in the data file). Always part of a data file header.

Display Label	Units	Description	Actual Name
abs_ambient		Leaf absorptance to Sun+Sky	abs_ambient
abs_blue		Leaf absorptance to blue	abs_blue
abs_green		Leaf absorptance to green	abs_green
abs_red		Leaf absorptance to red	abs_red
abs_white		Leaf absorptance to white	abs_white
k_ambient	$\text{J } \mu\text{mol}^{-1}$	Conversion factor for ambient	k_ambient
k_blue	$\text{J } \mu\text{mol}^{-1}$	Conversion factor for blue	k_blue
k_green	$\text{J } \mu\text{mol}^{-1}$	Conversion factor for green	k_green

Table 6-10. Constants pertaining to leaf absorption (called **LQConst** in the data file). Always part of a data file header. (...continued)

Display Label	Units	Description	Actual Name
k_red	J μmol^{-1}	Conversion factor for red	k_red
k_white	J μmol^{-1}	Conversion factor for white	k_white

Temperature constants

Table 6-11. Constants pertaining temperature, called **LTConst** in the data file. Always part of a data file header.

Display Label	Units	Description	Actual Name
deltaTw	$^{\circ}\text{C}$	Wall-air temp difference	deltaTw
fT1		Fraction Tleaf	fT1
fT2		Fraction Tleaf2	fT2
fT3		Fraction TleafEB	fTeb

Incident and absorbed light variables

Table 6-12. Values pertaining to light incident on and absorbed by the leaf in the chamber (called **LeafQ** in the data file). All of these are logged in data file columns.

Display Label	Units	Description	Actual Name
Qin	$\mu\text{mol m}^{-2} \text{s}^{-1}$	PPFD incident on the leaf	Qin
alpha		Weighted leaf absorption	alpha
Qabs	$\mu\text{mol m}^{-2} \text{s}^{-1}$	PPFD absorbed by the leaf	Qabs
convert	J μmol^{-1}	Weighted conversion from PPFD to W m^{-2}	convert

MultiPhase Flash variables

Table 6-13. Values pertaining to the multiphase flashes (called **MPF** in the data file). All of these are logged in data file columns.

Display Label	Units	Description	Actual Name
ID		ID of this MPF flash	EVENT_ID
P1_Fmax		Fmax of phase 1	P1_MAXF
P2_dQdt	$\text{mol m}^{-2} \text{s}^{-1}$	Phase 2 dQ/dt	P2_DQDT
P3_ΔF		Fmeasured - Fpredicted Phase 3	P2_DELTAF
P1_dur	ms	Phase 1 duration	PHASE1_DURATION

Table 6-13. Values pertaining to the multiphase flashes (called MPF in the data file). All of these are logged in data file columns. (...continued)

Display Label	Units	Description	Actual Name
P2_dur	ms	Phase 2 duration	PHASE2_DURATION
P3_dur	ms	Phase 3 duration	PHASE3_DURATION
P1_Qmax	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Qmax in phase 1	QMAX

Induction kinetics variables

The log group **FastKntcs** allows induction curve analysis to be added to log files. The two inflection points (F1 and F2) are found by curve fitting the modulated fluorescence data.

Table 6-14. Items contained in the **FastKntcs** group. Times are measured from the start of the actual flash.

Item	Description
EVENT_ID	ID of flash
DURATION	Flash duration
Fo	Initial value of modulated fluorescence (average of pre-flash values)
F1	Fluorescence value at first inflection point
F2	Fluorescence value at second inflection point
FMAX	Maximum value of modulated fluorescence
T@HIR	Time (secs) of "half initial rise", that is, when $F = F_o + (F1 - F_o) / 2$
T@F1	Time (secs) of F1 point
T@F2	Time (secs) of F2 point
T@Fmax	Time (secs) of Fmax point

Miscellaneous induction kinetics items

Table 6-15. Other items from induction kinetics.

Item	Description
DCmax	Maximum value of DC fluorescence
DCo	Initial (extrapolated back) value of DC fluorescence
InitSlope	Initial slope of DC fluorescence
T_OFFSET	Time (seconds) in flr event file when flash occurred

Match variables

Table 6-16. Values related to matching (called **Match** in the data file).

Display Label	Units	Description	Actual Name
co2_adj	$\mu\text{mol mol}^{-1}$	CO ₂ match adjustment factor	co2_adj
co2_at	$\mu\text{mol mol}^{-1}$	CO ₂ at last match	co2_at
co2_cv	%	CV of last CO ₂ match	co2_cv
count		Match count number	count
h2o_adj	mmol mol^{-1}	H ₂ O match adjustment factor	h2o_adj
h2o_at	mmol mol^{-1}	H ₂ O at last match	h2o_at
h2o_cv	%	CV of last H ₂ O match	h2o_cv
hhmmss		Time of last match	hhmmss
Time	secs	Time of last match (Epoch time, seconds since Jan 1970)	time

Measured variables

Table 6-17. Measured variables (called **Meas** in the data file). Basic input measurements in engineering units. A subset of these are logged in the data file.

Display Label	Units	Description	Actual Name
CO2_r	$\mu\text{mol mol}^{-1}$	Reference cell CO ₂ concentration	CO2_r
CO2_s	$\mu\text{mol mol}^{-1}$	Sample cell CO ₂ concentration	CO2_s
Fan_speed	rpm	Chamber fan rotation rate	Fan_speed
Flow	$\mu\text{mol s}^{-1}$	Flow rate to chamber	Flow
H2O_r	mmol mol^{-1}	Reference cell H ₂ O concentration	H2O_r
H2O_s	mmol mol^{-1}	Sample cell H ₂ O concentration	H2O_s
Qamb_in	$\mu\text{mol m}^{-2} \text{s}^{-1}$	In-chamber ambient PPFD	PPFD_in
Qamb_out	$\mu\text{mol m}^{-2} \text{s}^{-1}$	External quantum sensor	PPFD_out
ΔP_{cham}	kPa	Chamber overpressure	Pchamber
Pa	kPa	Atmospheric Pressure	Press
Time	s	Epoch time, seconds since Jan 1970	TIME
Tair	°C	Chamber air temperature	Tchamber
Tleaf	°C	Leaf thermocouple #1	Tleaf
Tleaf2	°C	Leaf thermocouple #2	Tleaf2

Measured variables (the second set)

Table 6-18. Measured variables (called **Meas2** in the data file). Basic input measurements in engineering units. A subset of these are logged in the data file.

Display Label	Units	Description	Actual Name
CO2_a	$\mu\text{mol mol}^{-1}$	A cell CO ₂ concentration	CO2_a
CO2_b	$\mu\text{mol mol}^{-1}$	B cell CO ₂ concentration	CO2_b
ΔCO_2	$\mu\text{mol mol}^{-1}$	Sample - Reference CO ₂	CO2_d
CO2_r_d	$\mu\text{mol mol}^{-1}$	Reference cell CO ₂ , dry mole fraction	CO2_r_d
CO2_s_d	$\mu\text{mol mol}^{-1}$	Sample cell CO ₂ , dry mole fraction	CO2_s_d
Flow_a	$\mu\text{mol s}^{-1}$	A cell flow rate	Flow_a
Flow_b	$\mu\text{mol s}^{-1}$	B cell flow rate	Flow_b
H2O_a	mmol mol^{-1}	A cell H ₂ O concentration	H2O_a
H2O_b	mmol mol^{-1}	B cell H ₂ O concentration	H2O_b
$\Delta\text{H}_2\text{O}$	mmol mol^{-1}	Sample - Reference H ₂ O	H2O_d
Td_r	°C	Reference cell dew point	Td_r
Td_s	°C	Sample cell dew point	Td_s
e_r	kPa	Reference cell vapor pressure	e_r
e_s	kPa	Sample cell vapor pressure	e_s

Light source constants

Table 6-19. Constants pertaining to light sources (called **QConst** in the data file). These are always part of a data file header.

Display Label	Units	Description	Actual Name
fQ_Amb_in		Fraction Q from Amb_in	fQ_Amb_in
fQ_Amb_out		Fraction Q from Amb_out	fQ_Amb_out
fQ_ConsoleLS		Fraction Q from ConsoleLS	fQ_ConsoleLS
fQ_Flr		Fraction Q from Flr	fQ_Flr
fQ_HeadLS		Fraction Q from HeadLS	fQ_HeadLS

Raw data variables

Table 6-20. Raw voltage signals underlying many of the measured values (called **Raw** in the data file). If logged, they will appear in data file columns.

Display Label	Units	Description	Actual Name
ADC_CH1	V	Spare channel 1	ADC_CH1
ADC_CH2	V	Spare channel 2	ADC_CH2
ADC_CH3	V	Spare channel 3	ADC_CH3
ADC_CH4	V	Spare channel 4	ADC_CH4
ADC_CH5	V	Spare channel 5	ADC_CH5
ADC_CH6	V	Spare channel 6	ADC_CH6
ADC_CH7	V	Spare channel 7	ADC_CH7
ADC_CH8	V	Spare channel 8	ADC_CH8
Flow_a_v	V	Exhaust flow signal cell A	Flow_a_v
Flow_b_v	V	Exhaust flow signal cell B	Flow_b_v
Vflow	V	Flow meter signal	Vflow
VPchamber	V	Chamber pressure sensor signal	VPchamber
Wc_a	V	Sample counts, CO ₂ a	Wc_a
Wc_b	V	Sample counts, CO ₂ b	Wc_b
Wco_a	V	Reference counts, CO ₂ a	Wco_a
Wco_b	V	Reference counts, CO ₂ b	Wco_b
Ww_a	V	Sample counts, H ₂ O a	Ww_a
Ww_b	V	Sample counts, H ₂ O b	Ww_b
Wwo_a	V	Reference counts, H ₂ O a	Wwo_a
Wwo_b	V	Reference counts, H ₂ O b	Wwo_b
abs_c_a	V	CO ₂ absorptance cell a	abs_c_a
abs_c_b	V	CO ₂ absorptance cell b	abs_c_b
abs_h_a	V	H ₂ O absorptance cell a	abs_h_a
abs_h_b	V	H ₂ O absorptance cell b	abs_h_b
leaf2_t_v	V	Leaf 2 temperature thermocouple V	leaf2_t_v
leaf_t_v	V	Leaf temperature thermocouple V	leaf_t_v

Stability

Table 6-21. Parameters related to stability criteria (called **Stability** in the data files).

Display Label	Units	Description	Actual Name
A:MN	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Mean of A	A.GasEx:MN

Table 6-21. Parameters related to stability criteria (called Stability in the data files). (...continued)

Display Label	Units	Description	Actual Name
A:OK		Stable status of A	A.GasEx:OK
A:SD	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Standard deviation of A	A.GasEx:SD
A:SLP	$\mu\text{mol m}^{-2} \text{s}^{-1} \text{min}^{-1}$	Rate of change of A	A.GasEx:SLP
CO2_s:MN	$\mu\text{mol mol}^{-1}$	Mean of CO2_s	CO2_s.Meas:MN
CO2_s:OK		Stable status of CO2	CO2_s.Meas:OK
CO2_s:SD	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Standard deviation of CO2_s	CO2_s.Meas:SE
CO2_s:SLP	$\mu\text{mol m}^{-2} \text{s}^{-1} \text{min}^{-1}$	Rate of change of CO2_s	CO2_s.Meas:SLP
H2O_s:MN	mmol mol^{-1}	Mean of H2O_s	H2O_s.Meas:MN
H2O_s:OK		Stable status of H2O	H2O_s.Meas:OK
H2O_s:SD	$\text{mmol m}^{-2} \text{s}^{-1}$	Standard deviation of H2O_s	H2O_s.Meas:SE
H2O_s:SLP	$\text{mmol m}^{-2} \text{s}^{-1} \text{min}^{-1}$	Rate of change of H2O_s	H2O_s.Meas:SLP
Stable		Number of stable items	Stable
State		stable # / total #	State
Total		Number of items tracked	Total

Status variables

Table 6-22. Status variables are quantities that reflect the state of the system (called Status in the data file). A subset of these are logged in data file columns.

Display Label	Units	Description	Actual Name
CO2_percent	%	CO ₂ Injector amount on	CO2_percent
CO2_r_setpoint	$\mu\text{mol mol}^{-1}$	CO ₂ mixer algorithmic setpoint	CO2_r_setpoint
DIAG	-	Diagnostic word	DIAG
Desiccant_percent	%	Desiccant valve position	Desiccant_percent
Flow_r	$\mu\text{mol s}^{-1}$	Flow through reference cell	Flow_r
Flow_s	$\mu\text{mol s}^{-1}$	Flow through sample cell	Flow_s
H2O_r_setpoint	mmol mol^{-1}	H ₂ O control algorithmic setpoint	H2O_r_setpoint
Heatx_setpoint	°C	Heat exchange algorithmic setpoint	Heatx_setpoint
Humidifier_percent	%	Humidifier valve position	Humidifier_percent
MatchCO2	$\mu\text{mol mol}^{-1}$	CO ₂ match adjustment factor	MatchCO2
MatchH2O	mmol mol^{-1}	H ₂ O match adjustment factor	MatchH2O

Table 6-22. Status variables are quantities that reflect the state of the system (called Status in the data file). A subset of these are logged in data file columns.
(...continued)

Display Label	Units	Description	Actual Name
MatchValveR	%	Reference match valve position, -100 to +100	MatchValveR
MatchValveS	%	Sample match valve position, -100 to +100	MatchValveS
SS_s	%	Junction temperature	SS_a
SS_r	%	Junction temperature	SS_b
Ts	°C	A cell inlet temperature	Ta
Tr	°C	B cell inlet temperature	Tb
Tchopper	°C	IRGA chopper temperature	Tchopper
Tirga	°C	IRGA block temperature	Tirga_block
Txchg	°C	Heat exchanger temperature	Txchg

Status variables (the second set)

Table 6-23. Status variables are quantities that reflect the state of the system (called Status2 in the data file). A subset of these are logged in data file columns.

Display Label	Units	Description	Actual Name
AccCO2_soda	mg	Absorbed CO ₂ (mg) by soda lime	AccCO2_soda
AccH2O_des	mg	Absorbed H ₂ O (mg) by desiccant	AccH2O_des
AccH2O_hum	mg	Added H ₂ O (mg) by humidifier	AccH2O_hum
CO2_hrs	hrs	CO ₂ cartridge hours	CO2_hrs
Console_H2O	mmol mol ⁻¹	H ₂ O after humidifier	Console_H2O
Console_RH	%	RH after humidifier	Console_RH
Console_T	°C	Temperature after humidifier	Console_T
Fan_percent	%	Fan power in percent	Fan_percent
Flow_percent	%	Flow split position, -100 to +100	Flow_percent
Pump		Pump status: 0=off, 0.5 minimum, 1=low, 2=med, 3=high	Pump
TleafJunction	°C	Reference junction 1 temperature	TleafJunction
TleafJunction2	°C	Reference junction 2 temperature	TleafJunction2
diag_12v	V	12 V test point	diag_12v
diag_20v	V	20 V test point	diag_20v
diag_3_3v	V	3.3 V test point	diag_3_3v

Table 6-23. Status variables are quantities that reflect the state of the system (called Status2 in the data file). A subset of these are logged in data file columns.
(...continued)

Display Label	Units	Description	Actual Name
diag_5_4v	V	5.4 V test point	diag_5_4v
diag_5v	V	5 V test point	diag_5v
Flow_b	$\mu\text{mol s}^{-1}$	B cell flow rate	Flow_b
heatx_t_pwm_v	V	Heat exchanger drive voltage	heatx_t_pwm_v

System variables

Table 6-24. System variables are related to the time and observation number.

Display Label	Units	Description	Actual Name
date	s	Date and time of last observation	date
elapsed	s	Seconds since first observation	elapsed
obs		Number of observations logged	obs
time	s	Epoch time, seconds since Jan 1970	time

Note: In a data file, a value of `sys::time` and `GasEx::Time` will be different if the data was logged with extra averaging time. The time for `sys::time` is always the actual time data was logged, while `GasEx::time` lags behind by an amount that is equal to $T/2$, where T is the averaging time.

Section 7.

System tests, messages, & troubleshooting

When things go wrong

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Warmup tests	7-3	always being overridden	7-37
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Warmup and system tests

With software version 1.4, the System and Warmup Tests are modified to differentiate errors (serious issues) and warnings (things you may or may not need to address). When errors or warnings occur, there will always be one or more suggestions shown to help you address the problem, or determine if it can be ignored.

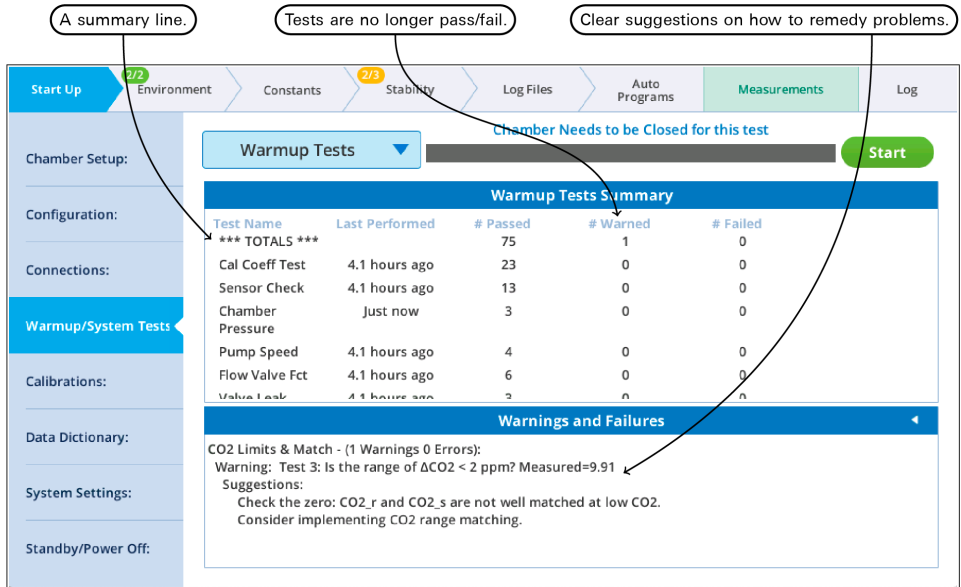


Figure 7-1. The warmup tests summary page.

The silent test

All of the system tests that rely on flow rate contain a silent test that checks the state of the flow system. ("Silent" means that output is logged only if there is a problem.) If there is problem with flow, the system test is abandoned, and reasons given.

If the issue is a console leak, the following results:

```
Chamber_Pressure started at 2019-09-25 13:15:34
Overpressure test, Pump=high
*** Fail *** Test 1: Abandoning Test. System Flow Fault.
Suggestion:
Console Flow Leak. Desiccant or Humidifier tube cap loose?
```

```
Chamber_Pressure stopped at 2019-09-25 13:15:46
```

If the issue is unexpectedly low flow, the following results:

```
Chamber_Pressure started at 2019-09-25 13:18:35
Overpressure test, Pump=high
*** Fail *** Test 1: Abandoning Test. Expected flow > 100. Measured -0.0.
  Suggestions:
    Is the flow tube from the console firmly connected to the head?
    Is the pump functioning?
Chamber_Pressure stopped at 2019-09-25 13:18:46
```

Warmup tests

The warmup tests are designed for daily use, to catch errors and faults that might compromise your measurements. If everything passes, you can be reasonably sure (but not guaranteed) that the system is working as expected. Common failures are due to worn out desiccant or an empty CO₂ cylinder. Other failures may be simply because a setting or measurement is just outside the expected range, in which case you have to decide if it needs attention or can be ignored. The descriptions below should help you make those decisions.

Calibration coefficients test

The calibration coefficient test tests each current user calibration setting to see if it falls within an expected range. If a value falls outside that range, it is marked as failed. This does not necessarily mean something is wrong, but it does bear further investigation. For example, if a zeroing parameter failed, it is OK if that value is what it really takes to properly zero that sensor, but make sure that the sensor hasn't been mis-zeroed instead.

```
Pass Test 1: Cal check: Is oxygen between 2 and 22? Currently=21.0
Pass Test 2: Cal check: Is co2azero between 0.8 and 1.2? Currently=0.90977
Pass Test 3: Cal check: Is co2aspan1 between 0.8 and 1.2? Currently=1.003
Pass Test 4: Cal check: Is co2aspan2 between -0.3 and 0.3? Currently=-0.002
Pass Test 5: Cal check: Is co2bzero between 0.8 and 1.2? Currently=0.911449
Pass Test 6: Cal check: Is co2bspan1 between 0.8 and 1.2? Currently=1.0
Pass Test 7: Cal check: Is co2bspan2 between -0.3 and 0.3? Currently=0.0
Pass Test 8: Cal check: Is h2oazero between 0.8 and 1.2? Currently=1.01078
Pass Test 9: Cal check: Is h2oaspan1 between 0.8 and 1.2? Currently=1.01456
Pass Test 10: Cal check: Is h2oaspan2 between -0.3 and 0.3? Currently=-0.116034
Pass Test 11: Cal check: Is h2obzero between 0.8 and 1.2? Currently=0.988944
Pass Test 12: Cal check: Is h2obspan1 between 0.8 and 1.2? Currently=1.02657
Pass Test 13: Cal check: Is h2obspan2 between -0.3 and 0.3? Currently=-0.263223
Pass Test 14: Cal check: Is tazero between -0.5 and 0.5? Currently=-0.223923
```

```
Pass Test 15: Cal check: Is tbzero between -0.5 and 0.5? Currently=-0.174402
Pass Test 16: Cal check: Is flowmeterzero between 0.5 and 1.4?
    Currently=0.991608
Pass Test 17: Cal check: Is flowazero between 0 and 0.8? Currently=0.39242
Pass Test 18: Cal check: Is flowbzero between 0 and 0.8? Currently=0.22801
Pass Test 19: Cal check: Is chamberpressurezero between 2 and 3?
    Currently=2.55412
Pass Test 20: Cal check: Is ssa_ref between 20000 and 55000? Currently=34092.0
Pass Test 21: Cal check: Is ssb_ref between 20000 and 55000? Currently=33927.5
Pass Test 22: Chamber Info: Is pca = 0? Currently=0
Pass Test 23: Chamber Info: Is pcb = 0? Currently=0
```

h2oazero, h2obzero, co2azero, co2bzero

Zero values for the IRGAs (a = sample, b = reference) are typically between 0.8 and 1.2. These values are set from the calibration menu, either by flowing zero air through the appropriate cell and pressing a button, or by direct editing. If one of them is out of range, visit the calibration menu, and make sure the zero was properly done.

Example: suggestion when warning about co2azero:

Verify that CO2_s is properly zeroed.

h2oaspan1, h2obspan1, co2aspan1, co2bspan1

Span 1 values for the IRGAs (a = sample, b = reference) are typically between 0.9 and 1.2, but expect them to be very close to 1.0. Departures from 1.0 may be real, or may be due to spanning at an inappropriate time, or with a gas that is not actually at the assumed concentration (or for H₂O, an issue with the LI-610 dew point generator: lack of water, overpressure, etc.) These values are set from the calibration menu, either by flowing calibrated air through the appropriate cell and pressing a button, or by direct editing. Hint: manually setting these values to 1.0 can sometimes be the best action for span related issues. If you do this, also set the associated span 2 value (next) to 0.

Example: suggestion when warning about co2aspan:

Verify that CO2_s is properly spanned.

h2oaspan2, h2obspan2, co2aspan2, co2bspan2

Span 2 values for the IRGAs (a = sample, b = reference) are typically between -0.3 and +0.3. A value of 0 means there is no secondary span. Values outside the expected range should be treated with suspicion, and are likely due to improper calibration. These values are set from the calibration menu, either by flowing calibrated air through the appropriate cell and pressing a button, or by direct editing. Hint: manually setting these values to 0 can sometimes be the best action for span related issues.

Example: suggestion when warning about co2aspan2:

Verify that CO₂_s is properly spanned.

tazero, tbzero

These are offsets to correct the IRGA exhaust temperature sensors (a = sample, b = reference) to the IRGA temperature thermistor (T_{irga}). These are typically between -0.5 and +0.5 °C. The test for an appropriate setting is to compare T_s and T_r - they should be quite close under stable temperature conditions and similar flows going through the cells. These temperatures can be matched in software version 1.4 and above, by using the BP (background program) named ZeroTaTb.py found in /home/licor/apps/tech/.

Example: suggestions when warning about tazero:

Verify that sample cell temp T_s is close to T_{irga} .
Use the apps/tech/ZeroTaTb.py BP to make them match.

ssa_ref, ssb_ref

These values are factory-set reference readings used to compute the IRGA cell signal strength SS_s and SS_r. (ssa_ref is for sample, and ssb_ref used for reference). Expected values are between 22000 and 55000.

Example: suggestion when warning about ssa ref

This only affects sample cell signal strength SS_s.

chamberpressurezero

This value is the signal from the chamber overpressure sensor when overpressure is 0. The expected value is between 2 and 3 V. This can be set in the flow zeroing calibration screen. A value out of that range would indicate that zeroing occurred when it should not have (there was overpressure), or perhaps a sensor fault.

Suggestion when warning about chamberpressurezero:

Verify that chamber pressure is properly zeroed.

flowmeterzero

This value is the offset of the main flow meter. Expected value is between 0.5 and 1.4 V. This can be set in the flow zeroing calibration screen. A value out of that range would indicate that zeroing occurred when it should not have (there was flow), or a sensor fault.

Suggestion when warning about flowmeterzero:

Verify that Flow is properly zeroed.

flowazero, flowbzero

These are offset signals to zero the IRGA exhaust flow sensors (a = sample, b = reference). The expected range is between 0 and 0.8 V. These can be set in the flow zeroing calibration screen. A value out of that range would indicate that zeroing occurred when it should not have (there was flow), or a sensor fault.

Example: suggestion when warning about flowazero:

Verify that Flow_s is properly zeroed.

pca, pcb

These values are used to correct chamber overpressure for fan speed and flow rate in chambers that have apertures (and a modified internal flow path). The expected values are based on what chamber type the system thinks is attached. The values are available on the Chamber Setup screen.

The screenshot shows the 'Options for the Attached Chamber 6800-01A Fluorometer' configuration screen. The 'Chamber Info' panel on the right contains the following fields:

- id: 6800-01A
- sn: MPF-551000
- rev: 1
- ppfd cal: 0
- vol: 81
- pca: 4.3321e-10 (highlighted with a red circle)
- pcb: 1.0215e-05 (highlighted with a red circle)

A 'Load Factory' button is located below the fluorometer icon, and a 'Lock' button is at the bottom of the Chamber Info panel. A caution note at the bottom right states: 'Caution: values stored in chamber memory.'

Table 7-1. Values for pressure correction coefficients for each chamber type.

Chamber	pca	pcb
6800-01 Fluorometer	0	0
6800-01A Fluorometer	4.3321E-10	1.0215E-5
6800-12 3x3	0	0
6800-12A 3x3	2.6520E-10	1.3064E-5
6800-13 6x6	4.3998E-11	1.4792E-5
6800-17 Small Plant	0	0
6800-19 Custom	0	0

Suggestion when warning about pca or pcb:

Set proper value in Chamber Setup.

Oxygen

This test checks the system constant for a reasonable value for oxygen (2 to 22%). The value is settable in system constants.

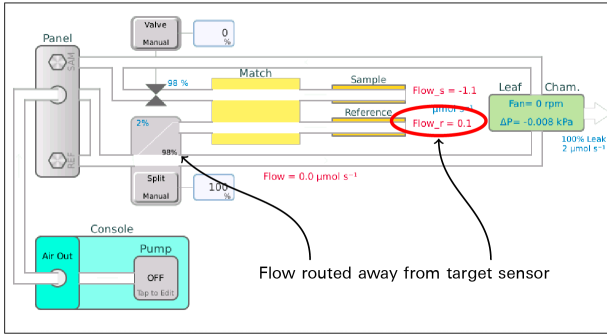
Suggestion when warning about Oxygen:

Set as needed in system constants.

Sensor check

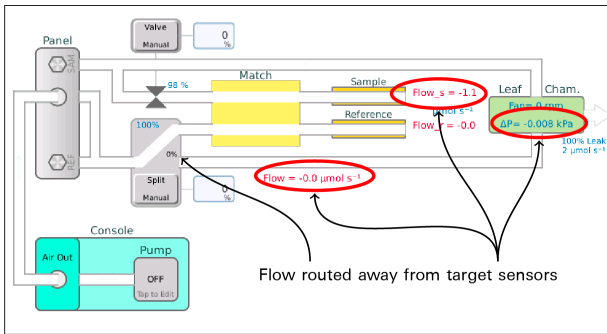
This test checks some sensors for proper zero, and some other sensors and settings for reasonable values.

With the pump off, mixing fan off, and temp control off, all flow (if there is any) is routed to the chamber, and after a 10 second wait, the reference cell IRGA flow is tested for zero.



Pass Test 1: Is Flow_r within 5 of zero? Measured=0.1μmol s⁻¹

Flow is then routed to the Reference IRGA, and after a 5 second wait, the following zeros are checked:



Pass Test 2: Is Flow within 2 of zero? Measured=-0.2μmol s⁻¹

Pass Test 3: Is Pchamber within 0.05 of zero? Measured=0.0008kPa

Pass Test 4: Is Flow_s within 5 of zero? Measured=-0.4μmol s⁻¹

Falling outside of the stated tolerance is a warning. It becomes an error if the sensor is out by more than 4 times the tolerance. The remedy is always:

Zero the sensor

Tests 5 and 6 check that the IRGA's internals are being temperature controlled to 30C. They should be correct by default, but they are changeable through alternative interfaces.

```
Pass Test 5: IRGA temperature control on? Setting= on
Pass Test 6: IRGA temperature control on target (30.0)? Measured 30.0
```

A failure on either of these IRGA control temperature tests is an error, with the suggestions:

```
Try restarting the system.
Contact LI-COR.
```

A sanity check on temperature sensors follows. At least one leaf temperature sensor is expected (unless configured for the soil chamber). Any other failure in this group would indicate a failed sensor, or that you are in conditions out of the operating range of the instrument.

```
Pass Test 7: At least one Leaf temp sensor? Tleaf=27.33, Tleaf2=999.9
Pass Test 8: Is Tchamber between -5C and 60C? 27.51
Pass Test 9: Is Txchg between -5C and 60C? 25.82
Pass Test 10: Is Ta between -5C and 60C? 27.53
Pass Test 11: Is Tb between -5C and 60C? 27.57
Pass Test 12: Is Tirga_block between -5C and 60C? 27.74
Pass Test 13: Are Ta, Tb, and Tirga_block within 0.5C? 0.21
```

A failure on Test 7 is a warning, with the suggestion:

```
Plug in a working leaf temperature thermocouple, or use energy balance.
```

A failure on Tests 8 through 12 is an error, with the suggestion:

```
Contact LI-COR.
```

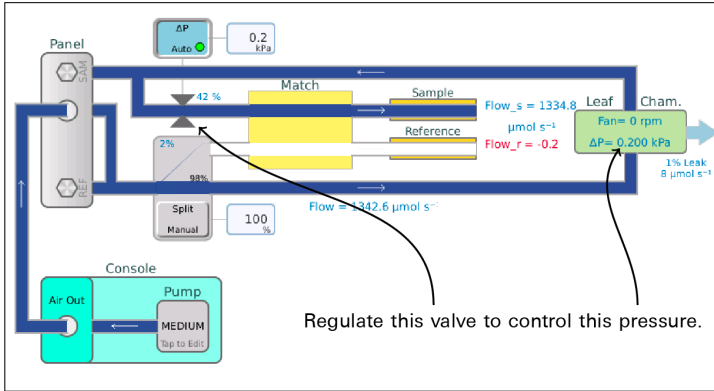
A failure on Test 13 (Ta, Tb, and Tirga block matching) is a warning, with the suggestion:

```
Use the apps/tech/ZeroTaTb.py BP to make them match.
```

Chamber pressure

The chamber pressure test attempts to hold the chamber overpressure at 0.2 kPa with the pump on high, then medium, then low. If there is a chamber leak, the lower the pump speed, the harder it is to hold that high an overpressure. If the

issue is not a chamber leak, but a failed pressure control valve, then the test would fail at all pump speeds.



A normal chamber:

Pass Test 1: Can Pchamber be held at 0.2 kPa with pump on high? Measured 0.2 kPa with valve @ 37.0%

Pass Test 2: Can Pchamber be held at 0.2 kPa with pump on medium? Measured 0.2 kPa with valve @ 47.0%

Pass Test 3: Can Pchamber be held at 0.2 kPa with pump on low? Measured 0.2 kPa with valve @ 52.0%

A leaky chamber:

Pass Test 1: Set Pchamber to 0.2 kPa, pump= high? Measured 0.201 kPa, valve= 56.0%

Pass Test 2: Set Pchamber to 0.2 kPa, pump= medium? Measured 0.201 kPa, valve= 59.0%

*** Warning *** Test 3: Set Pchamber to 0.2 kPa, pump= low? Measured 0.164 kPa, valve= 98.0%

Suggestion:
Check the chamber for leaks.

Failed valve (or open chamber):

*** Fail *** Test 1: Set Pchamber to 0.2 kPa, pump= high? Measured 0.007 kPa, valve= 98.0%

Suggestion:
Is the chamber closed?,"

*** Fail *** Test 2: Set Pchamber to 0.2 kPa, pump= medium? Measured 0.008 kPa, valve= 98.0%

Suggestion:
Is the chamber closed?,"

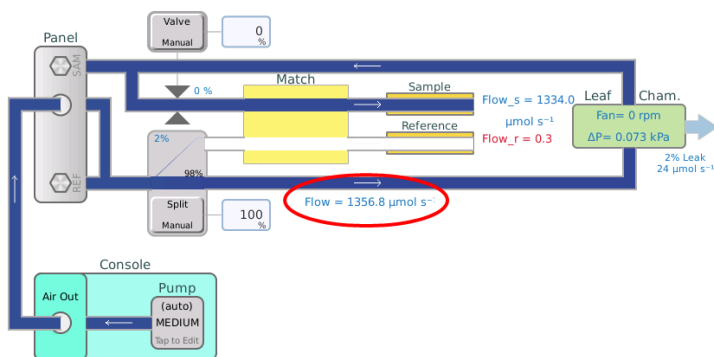
```

*** Fail *** Test 3: Set Pchamber to 0.2 kPa, pump= low? Measured 0.009 kPa,
valve= 98.0%
Suggestions:
Is the chamber closed?
Is the overpressure sensor zeroed correctly?
Is the pressure valve even working?

```

Pump speed

The pump speed checks that the 4 active pump settings (minimum through high) generate the flow expected, within 20%, adjusting for ambient pressure.



```

Pass Test 1: At Pump=minimum, Flow expected to be within 20% of 685.0? Measured
655.265 (-4.3% low)
Pass Test 2: At Pump=low, Flow expected to be within 20% of 1028.0? Measured
981.342 (-4.5% low)
Pass Test 3: At Pump=medium, Flow expected to be within 20% of 1370.0? Measured
1325.79 (-3.2% low)
Pass Test 4: At Pump=high, Flow expected to be within 20% of 1713.0? Measured
1654.25 (-3.4% low)

```

A failure is an error, with these suggestions:

```

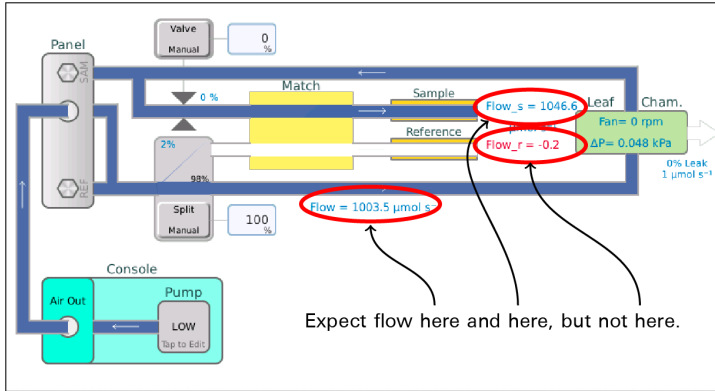
Check for leaks in the flow connections between the console and sensor head.
If it is a pump calibration issue, run the BP
apps/tech/CalibratePumpSettings.py,

```

Flow valve functionality

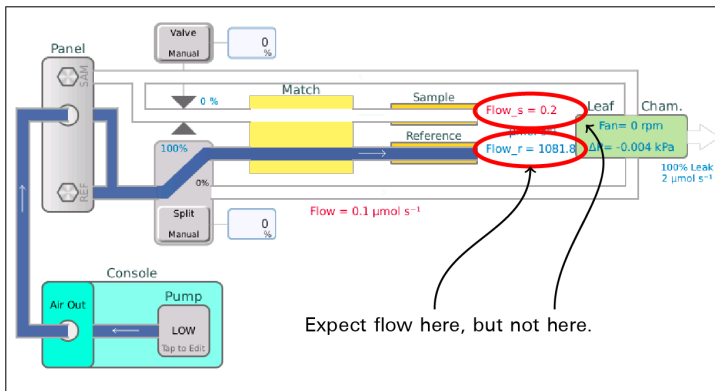
The flow valve functionality test checks for basic functionality of the match/flow split control unit by doing a simple sequence and looking at the flow meters:

- With all the air routed to the chamber and the match valve normal, measure Flow. That value becomes the target for this test. Flow_s (if the chamber is closed and not leaking badly) should be within 30% of the target value, and Flow_r would be 0.



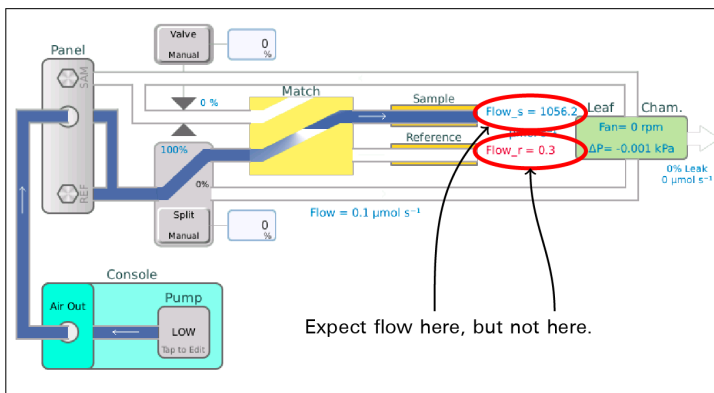
Pass Test 1: With FlowSplit="To Chamber" and Match="Normal": is Flow_s = 1003.5 +/- 30%? Measured=1048.6
 Pass Test 2: With FlowSplit="To Chamber" and Match="Normal": is Flow_r = 0 +/- 7.0? Measured=-0.2

- Now route all the flow to the reference cell. Flow_r should read close to the target, and Flow_s should be 0.



Pass Test 3: With FlowSplit="To IRGA" and Match="Normal": is Flow_r = 1003.5 +/- 30%? Measured=1081.8
 Pass Test 4: With FlowSplit="To IRGA" and Match="Normal": is Flow_s = 0 +/- 7.0? Measured=0.2

- 3 With all the flow still going to the IRGAS, route it to the sample.
Flow_s should be close to the target, and Flow_r should be 0.



Pass Test 5: With FlowSplit="To IRGA" and Match="Crossed": is Flow_s = 1003.5 +/- 30%? Measured=1056.2

Pass Test 6: With FlowSplit="To IRGA" and Match="Crossed": is Flow_r = 0 +/- 7.0? Measured=0.3

If a flow rate is off by more than 30% of expected, the following are suggested:

Determine why the flow is not getting to the expected location.
Worst case is a nonfunctioning valve, which would require LI-COR service.

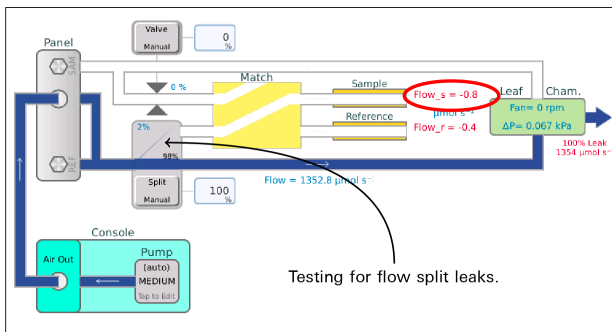
On failures of the no-flow sensor being off by more than $7 \mu\text{mol s}^{-1}$, the following are suggested:

This could be a flow zeroing issue (refer to 'Sensor Check' test).
This could be a leaking valve issue (refer to the 'Valve Leak' test).

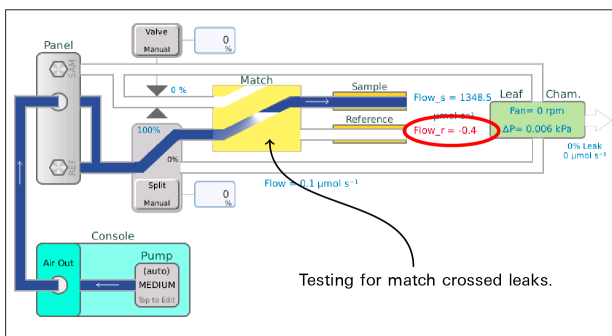
Valve leak

The valve leak test does a systematic leak detection test on the flowsplit / match valve. The methodology is to put the valve into a particular state, then ramp the pump from low to high, and look for any trends up in the flow meters that should NOT be getting any flow. Specifically, on a plot of leaked flow plotted as function of main flow, we are looking for slopes of less than 0.2%. More than that might be a leak.

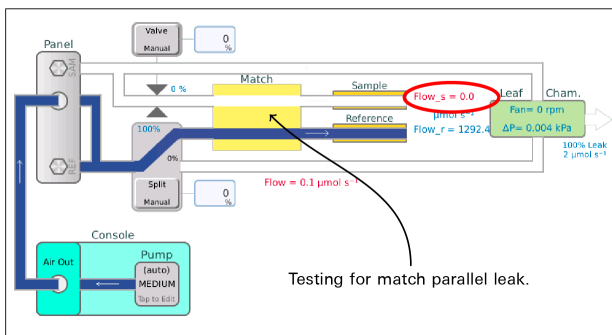
Step 1 has all the flow going to the chamber, with the match valve crossed to return air from the chamber is vented to the case. Flow_s should be getting nothing.



Step 2 routes all flow to IRGA and Flow_s. Flow_r would be getting nothing.



Step 3 routes all flow to IRGA and Flow_r. Flow_s should be getting nothing.




```

Step 1/3 (Flow Split Valve): All flow to chamber, leaks to Flow_s
Pass Test 1: Is Flow split valve leak response slope < 0.002 ? Measured -0.001
Step 2/3 (Match Crossed): All flow to Flow_s, leaks to Flow_r
Pass Test 2: Is Match X valve leak response slope < 0.002 ? Measured -0.0
Step 3/3: Match Straight: All flow to Flow_r, leaks to Flow_s
Pass Test 3: Is Match || valve leak response slope < 0.002 ? Measured -0.001

```

Failures of the rst two tests are warnings, since they don't really compromise measurement integrity. The suggestions are:

```

Slight leaks in this flow configuration do not compromise measurements.
Fixing this issue requires LI-COR service.

```

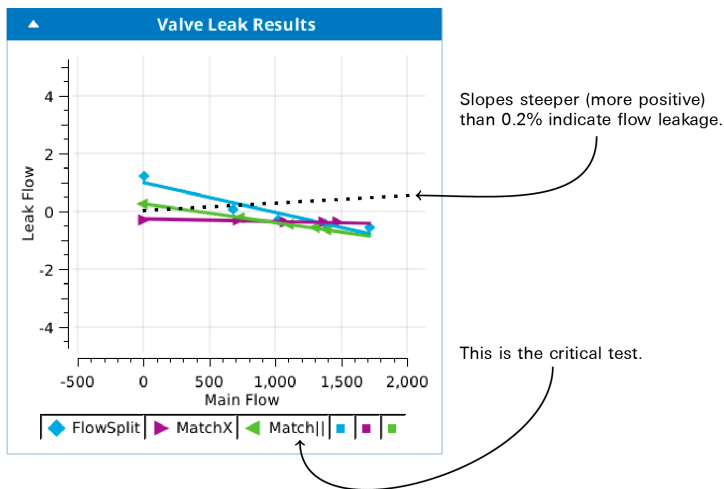
A failure of the last test is serious, and an error. The suggestions are:

```

Leaks with the match valve in normal position DO compromise measurements.
Fixing this issue requires LI-COR service.

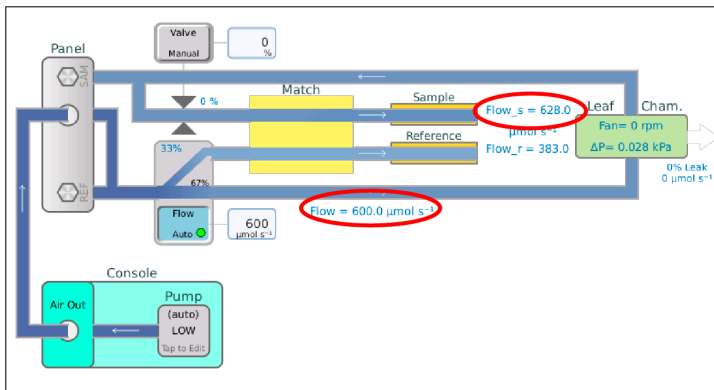
```

A leak in the flow split (step 1) wouldn't matter for normal operation, and a leak while matching (step 2) wouldn't matter, since the valve is not fully closed, but directing air to both cells. However, the critical one is the third test, with the match valve in normal operating position. A leak here represents potential cross contamination between cells, which will corrupt your measurement. A leak in step 3 needs to be dealt with.



Sample cell flow meter calibration

The LI-6800 does real time chamber leak detection by comparing the main flow meter to the sample cell flow meter. Since the latter is of much lower performance, it usually needs to be calibrated to the main flow meter. (This calibration only comes into play when computing leak corrections - it does NOT change the displayed / logged values of Flow_s.)



```

Step 1: Testing for chamber leak: Bypassing chamber
Step 2: Testing for chamber leak: Through chamber
Pass Test 1: Is Flow_s > 400 ? Measured 1081.9
Pass Test 2: Is preliminary leak < 2% ? Measured 1.0
Step 3: Matching at 8 flows
Flow= 0.0, Flow-Flow_s= -1.0
Flow= 102.0, Flow-Flow_s= 14.0
Flow= 407.0, Flow-Flow_s= 9.0
Flow= 600.0, Flow-Flow_s= -28.0
Flow= 801.0, Flow-Flow_s= -65.0
Flow= 1005.0, Flow-Flow_s= -45.0
Flow= 1200.0, Flow-Flow_s= -45.0
Flow= 1401.0, Flow-Flow_s= 19.0

```

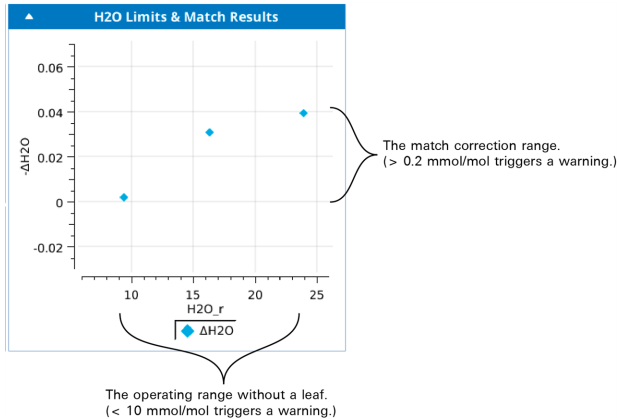
If a preliminary leak test indicates a leak of less than 2%, an error is triggered, with the suggestion

Is the chamber closed?

H₂O limits & match

This test checks the control range for water, and thus checks the state of the desiccant and humidifier tubes. It also checks how well the IRGAs are matched for water over that range at three points: driest, ambient, wettest.

The final graph is a view of the H₂O operating range, and what the match correction looks like over that range.



With working chemicals:

```

Step 1: Full desiccant
Step 2: Bypass all chemicals
Pass Test 1: Is desiccant effect > 80% of expected? Measured 93% (18.4 mmol/mol)
Pass Test 2: Is lowest H2O_r between -1 and 3? Measured 1.26781
Step 3: Full Humidifier
Pass Test 3: Is Humidifier adding to ambient? (Change = 4.1 mmol/mol)
Pass Test 4: Is the range of ΔH2O < 0.2 ppm? Measured=0.03
Pass Test 5: Is the max |ΔH2O| < 0.1 mmol/mol? Measured=0.03

```

With bad desiccant:

```

Step 1: Full desiccant
Step 2: Bypass all chemicals
*** Warning *** Test 1: Is desiccant effect > 80% of expected? Measured 42% (6.9
mmol/mol)
Suggestion:
Check the desiccant
*** Warning *** Test 2: Is lowest H2O_r between -1 and 3? Measured 9.39568
Suggestions:
Check the desiccant

```

If the desiccant is good, perhaps the H2O sample and reference are not zeroed properly.

Step 3: Full Humidifier

Pass Test 3: Is Humidifier adding to ambient? (Change = 7.6 mmol/mol)

Pass Test 4: Is the range of $\Delta H_2O < 0.2$ ppm? Measured=0.037

Pass Test 5: Is the max $|\Delta H_2O| < 0.1$ mmol/mol? Measured=0.039

With a dry humidifier tube:

Step 1: Full desiccant

Step 2: Bypass all chemicals

Pass Test 1: Is desiccant effect > 80% of expected? Measured 93% (15.6 mmol/mol)

Pass Test 2: Is lowest H2O_r between -1 and 3? Measured 1.10174

Step 3: Full Humidifier

*** Fail *** Test 3: Is Humidifier adding to ambient? (Change = 1.0 mmol/mol)

Suggestion:
 Add water to the humidifier tube.

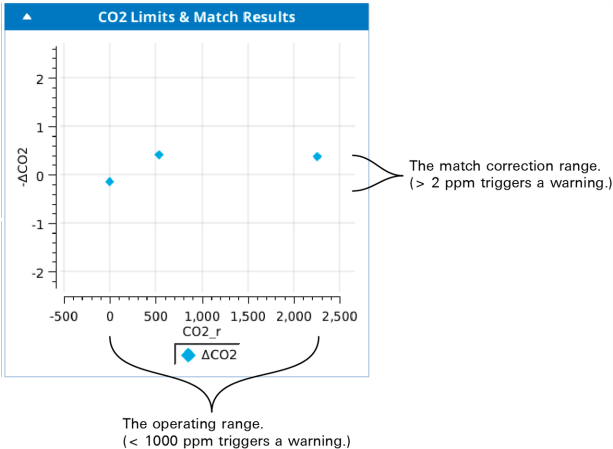
Pass Test 4: Is the range of $\Delta H_2O < 0.2$ ppm? Measured=0.053

Pass Test 5: Is the max $|\Delta H_2O| < 0.1$ mmol/mol? Measured=0.046

CO₂ limits & match

This test checks the control range for CO₂, and thus checks the state of the soda lime and CO₂ cartridge. It also checks how well the IRGAs are matched for CO₂ over that range at three points: scrubbed, ambient, full CO₂.

The final graph is a view of the CO₂ operating range, and what the match correction looks like over that range.



An open or extremely leaky chamber will produce an error with these suggestions:

```

*** Fail *** Test 1: Abandoning test because Flow_s < 100. Measured=-0.653923
  Suggestions:
    Is the chamber closed?
    Is the leaf thermocouple installed?",

```

Test results when everything works as expected:

```

Step 0: Flushing desiccant and humidifier
Step 1: Ambient
Step 2: Full scrub
Pass Test 1: Is lowest CO2_r between -2 and 5? Measured -0.0357313
Step 3: Full CO2
Pass Test 2: Is highest CO2_r > 1500 ppm? Measured 2251.7
Pass Test 3: Is the range of ΔCO2 < 2 ppm? Measured=0.54
Pass Test 4: Is the max |ΔCO2| < 1 ppm? Measured=0.4

```

Test 1 catches zeroing issues...

```

*** Warning *** Test 1: Is lowest CO2_r between -2 and 5? Measured -2.53
  Suggestion:
    Check the CO2 zero for reference and sample.

```

... and soda lime issues:

```

*** Warning *** Test 1: Is lowest CO2_r between -2 and 5? Measured 9.8
  Suggestion:
    Check the soda lime.
    If the soda lime is good, perhaps the CO2 sample and reference are not
    zeroed properly.

```

Test 2 catches empty CO₂ cylinders:

```

*** Warning *** Test 2: Is highest CO2_r > 1500 ppm? Measured -0.1
  Suggestion:
    Replace the CO2 cylinder.

```

If Test 3 fails, and the offset looks like a zeroing issue:

```

*** Warning *** Test 3: Is the range of ΔCO2 < 2 ppm? Measured=8.7
  Suggestions:
    Check the zero: CO2_r and CO2_s are not well matched at low CO2.
    Consider implementing CO2 range matching.

```

If Test 3 fails, and the offset looks like a spanning issue:

```

*** Warning *** Test 3: Is the range of ΔCO2 < 2 ppm? Measured=8.7
  Suggestions:
    Check the spans: CO2_r and CO2_s are not well matched at high CO2.
    Consider implementing CO2 range matching.

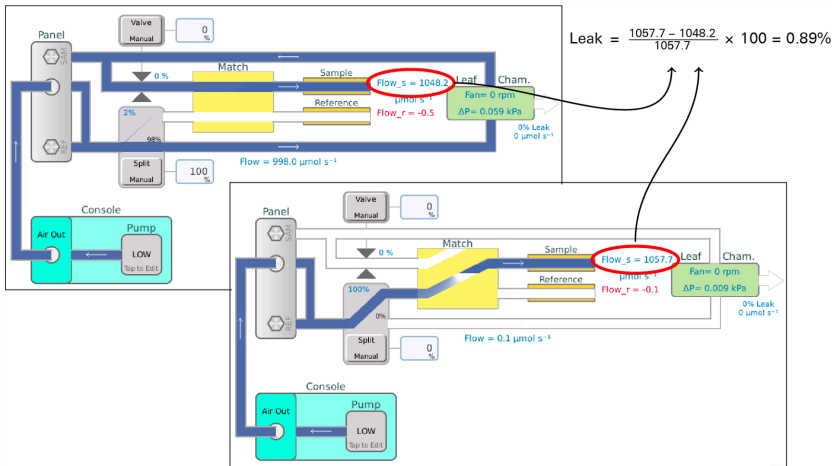
```

If Test 3 passes, there will be a Test 4 to catch an obvious match need:

*** Warning *** Test 4: Is the max |ΔCO2| < 1 ppm? Measured=2.4
 Suggestions:
 Matching will take care of this.

Chamber leak

The chamber leak test is a two-part leak test, comparing the Flow_s readings with all air going through the chamber, with all air going directly to the sample cell bypassing the chamber. This is potentially more accurate than the live leak indicator, since the former uses the same flow meter, while the latter uses separate flow meters and depends on their inter calibration.



Leaks less than 1% are pass the test; between 1 and 2% will generate a warning, and above 2% is an error,

Step 1: All flow bypasses the leaf chamber
 Bypass flow = 1057.7
 Step 2: All flow passes through the leaf chamber
 Chamber flow = 1048.2
 Pass Test 1: Leak rate < 1% ? Measured=0.9

Leaks below 50% merit these suggestions:

*** Fail *** Test 1: Leak rate < 1% ? Measured=22.6
 Suggestions:
 Check the gaskets.
 Improve sealing around any stems.

Above 50% produces these suggestions:

```

*** Fail *** Test 1: Leak rate < 1% ? Measured=99.3
  Suggestions:
  Is the chamber closed?
  Is the leaf temperature thermocouple installed?

```

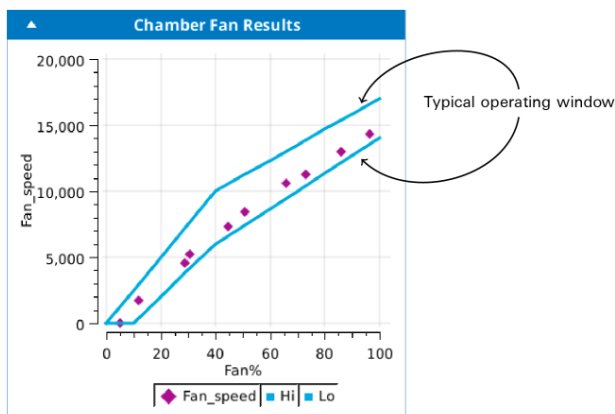
Chamber fan

The chamber fan test sets the fan to a series of 10 set points, each randomly chosen in a fixed interval. For each set point, there is a typical expected range of resulting fan speeds. If your results have a point or two just outside the expected range, you can safely ignore it. If all or most points are well out (such as all at 0), then there is clearly a problem. If there seems to be a problem with the fan not coming on at lower power settings (or at any setting), then power the head off, remove the chamber, and look for debris obstructing the fan.

```

Pass Test 1: 5.2% produces 5 rpm. Is that between 0.0 and 1300.0 rpm?,
Pass Test 2: 12% produces 1680 rpm. Is that between 400.0 and 3000.0 rpm?,
Pass Test 3: 28.6% produces 4560 rpm. Is that between 3720.0 and 7150.0 rpm?,
Pass Test 4: 30.7% produces 5210 rpm. Is that between 4140.0 and 7675.0 rpm?,
Pass Test 5: 44.6% produces 7295 rpm. Is that between 6613.0 and 10537.0 rpm?,
Pass Test 6: 50.7% produces 8410 rpm. Is that between 7427.0 and 11248.0 rpm?,
Pass Test 7: 65.9% produces 10585 rpm. Is that between 9453.0 and 13022.0 rpm?,
Pass Test 8: 73.1% produces 11285 rpm. Is that between 10413.0 and 13862.0 rpm?,
Pass Test 9: 85.8% produces 12960 rpm. Is that between 12107.0 and 15343.0 rpm?,
Pass Test 10: 96.5% produces 14335 rpm. Is that between 13533.0 and 16592.0
rpm?,

```



If the fan does not appear to be running at any power above 30%, an error with this suggestion is produced:

Power off, remove chamber, check for debris blocking the fan.

Any other violation is a warning with this suggestion:

Fan speeds slightly out of the envelope can be ignored.

Heat exchanger

The heat exchanger test does a three set point test using the heat exchanger, to make sure it is working. If there are failures, contact LI-COR.

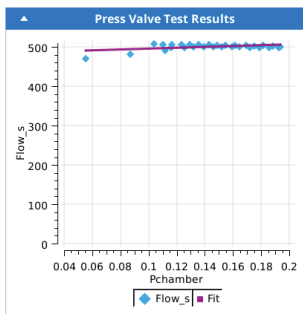
Doing 3 random Txchg setpoints,
Pass Test 1: Is Txchg within 0.2 of 33.3%? Measured 33.22 after 71.0 s,
Pass Test 2: Is Txchg within 0.2 of 23.3%? Measured 23.332 after 75.5 s,
Pass Test 3: Is Txchg within 0.2 of 28.3%? Measured 28.212 after 50.5 s,

Press valve test

This tests attempts to detect harmonic vibrations set up in the pressure control valve that might influence the Flow_s flow sensor. The pressure control is ramped up to 0.2 kPa and back, while the Flow_s sensor is monitored. Normally there is little or no effect on Flow_s, but a vibrating valve can cause a sudden apparent drop in the Flow_s reading.

A normal valve looks like this:

Pass Test 1: Is Slope of Flow_s with Pchamber > -500?: Measured=103.0

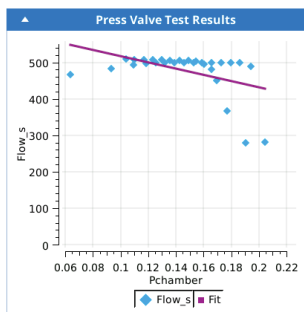


A vibrating valve looks like this:

```

*** Warning *** Test 1: Is Slope of Flow_s with Pchamber > -500? : Measured=-
850.0
Suggestions:
The pressure valve seems to be resonating and interfering with the Flow_s
sensor.
You can avoid this by operating at low (or no) overpressure, or by putting a
very slight flow restriction on the exhaust A port of the IRGA head.
This is fixable, but requires servicing by LI-COR.

```



An open or extremely leaky chamber will produce an error with these suggestions:

```

*** Fail *** Test 1: Abandoning test because Flow_s < 100. Measured=-0.653923
Suggestions:
Is the chamber closed?
Is the leaf thermocouple installed?,

```

Soil chamber test

The soil chamber test is only available when the soil chamber is attached. The test checks for chamber opening and closing, and circulation between the chamber and head.

Testing Open/Close

Pass Test 1: Chamber closing time expected to be 12 s +/- 6? Measured 12.0

Pass Test 2: Chamber opening time expected to be 12 s +/- 6? Measured 9.0

Testing Circulation Pump

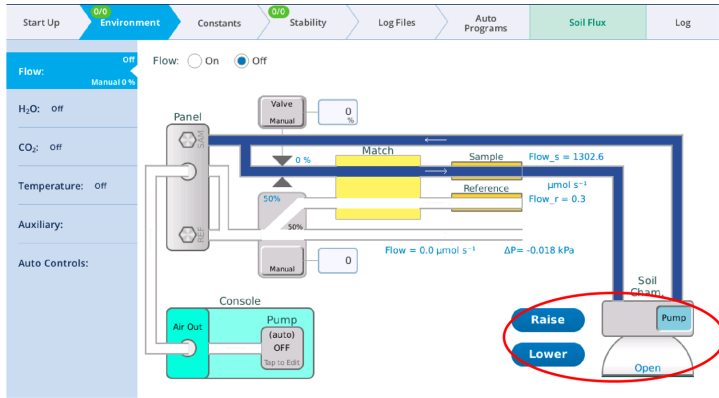
Pass Test 3: Max circulation flow > 1000? Measured 1367.26

Pass Test 4: Flow leakage (Flow_r+Flow) < 5 ? Measured 0.724502

These actions can also be tested manually from the **Environment > Flow** page.

Problems here are usually due to improper plumbing, kinked tubing, etc., and produce these suggestions:

Incorrect soil chamber plumbing?
Flow from IRGA obstructed?



Controller tests

The controller tests serve the following purpose: if you observe that the instrument is not controlling some parameter (e.g., CO₂_s) in an expected manner even after passing the warmup tests (i.e. chemicals checked, zeroes checked, etc.), then run the associated controller test for that parameter.

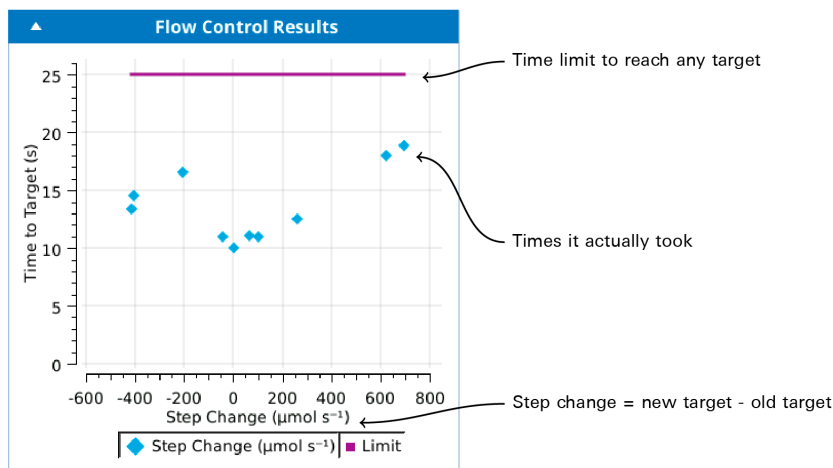
The controller tests have a common protocol: do a series of set points, randomly chosen, and note how long it takes the system to equilibrate at the new target. Failures result when the system is unable to stabilize at the new target in some pre-scribed amount of time.

There may be occasional failures, depending on the step change the happens to be come along, so don't be concerned if that happens now and again. Consistent and repeated failures would indicate a problem, however.

Sample results are below, from the Flow Controller test.

Pass Test 1:	Is Flow within 2 of 694 μmol s ⁻¹ ?	Measured 693.867 after 18.9 s
Pass Test 2:	Is Flow within 2 of 759 μmol s ⁻¹ ?	Measured 759.14 after 11.1 s
Pass Test 3:	Is Flow within 2 of 762 μmol s ⁻¹ ?	Measured 762.038 after 10.0 s
Pass Test 4:	Is Flow within 2 of 355 μmol s ⁻¹ ?	Measured 355.008 after 14.5 s
Pass Test 5:	Is Flow within 2 of 978 μmol s ⁻¹ ?	Measured 977.994 after 18.0 s
Pass Test 6:	Is Flow within 2 of 563 μmol s ⁻¹ ?	Measured 563.21 after 13.4 s
Pass Test 7:	Is Flow within 2 of 823 μmol s ⁻¹ ?	Measured 822.936 after 12.5 s
Pass Test 8:	Is Flow within 2 of 927 μmol s ⁻¹ ?	Measured 927.192 after 11.0 s
Pass Test 9:	Is Flow within 2 of 721 μmol s ⁻¹ ?	Measured 720.918 after 16.6 s

Pass Test 10: Is Flow within 2 of 677 $\mu\text{mol s}^{-1}$? Measured 676.836 after 11.0 s



Failed controller tests produce warnings with these suggestions:

An occasional failure, especially after a large step change, can be ignored. Consistent and repeated failures should be investigated.

Fluorometer tests

Cal values

The fluorometer calibration value tests factory calibration factors. The first 12 tests check each of the calibration values against a range of expected values, and warns about outliers. If there is a warning here, check the cal sheet to be sure the entered value is correct. If it is, trust the calibration sheet over the expected value, and ignore the warning.

```

Pass Test 1: Is F_red = 0.2 +/- 0.15 ? Measured= 0.12818
Pass Test 2: Is F_blue = 0.2 +/- 0.15 ? Measured= 0.174674
Pass Test 3: Is F_farred = 0.2 +/- 0.19 ? Measured= 0.175743
Pass Test 4: Is F_modred = 0.2 +/- 0.19 ? Measured= 0.202304
Pass Test 5: Is u_red = 2.5 +/- 2.0 ? Measured= 2.54922
Pass Test 6: Is u_blue = 0.2 +/- 0.15 ? Measured= 0.203029
Pass Test 7: Is u_farred = 0.003 +/- 0.002 ? Measured= 0.003662
Pass Test 8: Is u_modred = 0.03 +/- 0.029 ? Measured= 0.009615
Pass Test 9: Is dac_red_zero = 600 +/- 300 ? Measured= 653.0
Pass Test 10: Is dac_blue_zero = 600 +/- 300 ? Measured= 742.0
Pass Test 11: Is dac_farred_zero = 800 +/- 300 ? Measured= 752.0

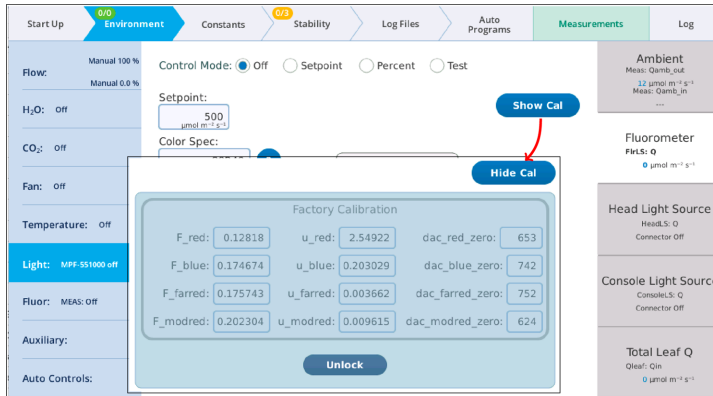
```

Pass Test 12: Is dac_modred_zero = 600 +/- 300 ? Measured= 624.0

If a value falls outside the expected range, then this suggestion is produced:

Make sure this is what is on the fluorometer's calibration sheet.

Fixing a value involves a visit to the fluorometer calibration dialog:



The next two tests check the farred LEDs, the actinic photodiode, and the fluorescence photodiode. We look at the effect of far red LEDs on the actinic detector (PC) and the fluorescence detector (DC). The slope of the response (counts per percent far red power) is logged and compared with expected values.

Checking farred response

Pass Test 13: Do farred LEDs affect PC? Expect PC/% > 1. Measured 2.3

Pass Test 14: Do farred LEDs affect DC? Expect DC/% > 50. Measured 115.0

If one slope fails, it is a warning:

Is the face of the fluorometer dirty and blocking light?'

If both slopes are near zero, then an error is produced with this suggestions:

Far red LEDs may not be working. Contact LI-COR

Tests 15-18 are a firmware check of the fluorometer, but will fail if the modulation LEDs can't hit a peak value of $100 \mu\text{mol m}^{-2} \text{s}^{-1}$, which corresponds to a average output (REDMODAVG) of $5 \mu\text{mol m}^{-2} \text{s}^{-1}$ when modulated at 50kHz, and $0.01 \mu\text{mol m}^{-2} \text{s}^{-1}$ when modulated at 100Hz.

Testing autoselection of modulation rate

```

Pass Test 15: With Q=172.0, is REDMODAVG=5.0 0.01? Measured 5.0
Pass Test 16: With Q=0.0, is REDMODAVG=0.01 0.01? Measured 0.01
Pass Test 17: With Q=165.0, is REDMODAVG=5.0 0.01? Measured 5.0
Pass Test 18: With Q=33.0, is REDMODAVG=0.01 0.01? Measured 0.01

```

The suggestion when not hitting the 5.0 target is:

```

Weak modulation? Modulated cannot reach 100 mol m-2 s-1? Contact LI-COR.

```

The suggestion when not hitting the 0.1 target is:

```

Fluorometer firmware or communications fault. Contact LI-COR.

```

Tests 19 and 20 are another firmware check.

```

Testing REDMODAVG and PFD computations
Pass Test 19: With REDMODAVG=4.99982 and actinic off, does PFD read 4.99982?
    Measured 4.99982
Pass Test 20: With REDMODAVG=0 and actinic off, does PFD read 0? Measured 0

```

An error here produces this suggestion:

```

Fluorometer firmware or communications fault. Contact LI-COR.

```

Actinic 1st response

During operation, when an actinic set point is specified, an active controller (active means it's watching the detector output) drives the light to the target value in typically 5 to 10 seconds depending on the step change. The Actinic 1st response test bypasses that, and takes the light immediately to a "first guess" value, leaving it there. It does this for a series of red, blue, and farred target values, and results are expected to be within 10% of the target value, or 20 $\mu\text{mol}/\text{m}^2/\text{s}$, which ever is larger).

```

Pass Test 1: farred. Is Q = 5.0 +/- 20? Measured=5.4
Pass Test 2: farred. Is Q = 10.0 +/- 20? Measured=10.6
Pass Test 3: farred. Is Q = 20.0 +/- 20? Measured=20.5
Pass Test 4: blue. Is Q = 20.0 +/- 20? Measured=17.7
Pass Test 5: blue. Is Q = 100.0 +/- 20? Measured=90.8
Pass Test 6: blue. Is Q = 500.0 +/- 50.0? Measured=481.1
Pass Test 7: blue. Is Q = 800.0 +/- 80.0? Measured=778.6
Pass Test 8: blue. Is Q = 1000.0 +/- 100.0? Measured=977.2
Pass Test 9: red. Is Q = 20.0 +/- 20? Measured=21.2
Pass Test 10: red. Is Q = 800.0 +/- 80.0? Measured=807.9
Pass Test 11: red. Is Q = 1500.0 +/- 150.0? Measured=1516.8
Pass Test 12: red. Is Q = 5000.0 +/- 500.0? Measured=5072.5
Pass Test 13: red. Is Q = 10000.0 +/- 1000.0? Measured=10134.2

```

Pass Test 14: red. Is $Q = 1500.0 \pm 1500.0$? Measured=15112.8
 Pass Test 15: red. Is $Q = 200.0 \pm 20.0$? Measured=202.2

Failures here would indicate that something has shifted since the factory calibration. It could be in the relation between controller current and light output, or in the relation between light output and photodiode signal. Or both. Slight failures can probably be ignored, since the active controller for set point control never uses first guesses. Large failures may merit returning the fluorometer to LI-COR for a re-calibration.

Values out of range are a warning. If the target intensity is <100 , a zeroing suggestion is produced.

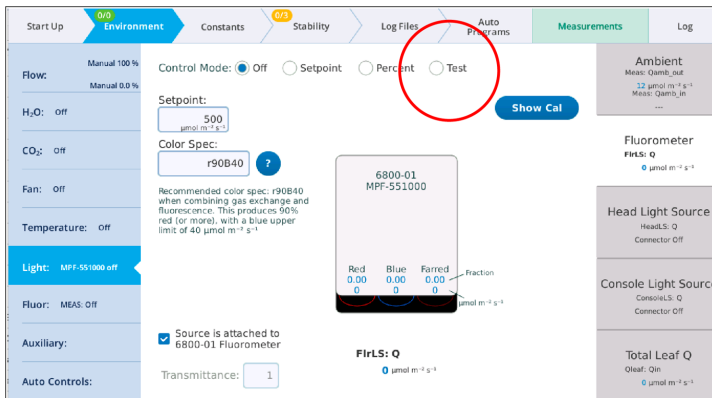
Check the PC (actinic photodiode) zero.

For higher intensities, specific cal values are targeted for checking. For example, if it is red light:

Compare cal sheet to factory cal. Specifically, the F_{red} , u_{red} , and dac_{red_zero} values.

Fluorometer may need recalibration. Contact LI-COR.

Note that this first response mode is available for use or manual testing by selecting the "Test" option in the fluorometer actinic control page.



Actinic control

The actinic control test does a controller test on actinic control. A series of random set points and colors are used for set points, and the time it takes for the

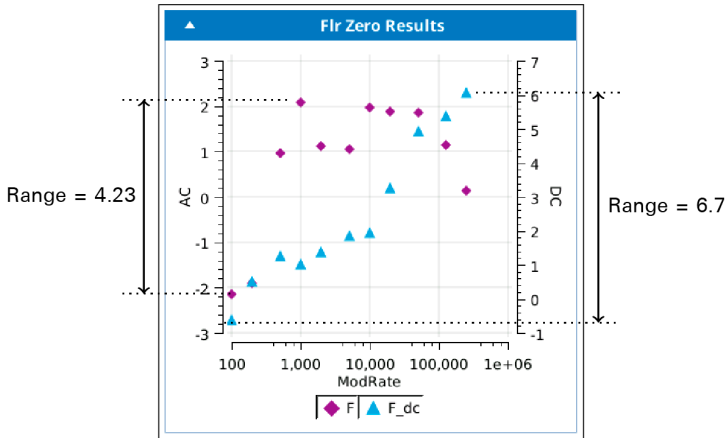
fluorometer to stabilize at the new target is recorded.

```
Pass Test 1: Is Q within 1.0 of 1032 umol m-2 s-1? Measured 1031.65 after 13.5s
Pass Test 2: Is Q within 1.0 of 794 umol m-2 s-1? Measured 794.424 after 10.5s
Pass Test 3: Is Q within 1.0 of 1406 umol m-2 s-1? Measured 1405.967 after 12.5s
Pass Test 4: Is Q within 1.0 of 1145 umol m-2 s-1? Measured 1145.747 after 9.5s
Pass Test 5: Is Q within 1.0 of 1562 umol m-2 s-1? Measured 1561.34 after 9.5s
Pass Test 6: Is Q within 1.0 of 617 umol m-2 s-1? Measured 617.342 after 12.0s
Pass Test 7: Is Q within 1.0 of 880 umol m-2 s-1? Measured 879.114 after 9.0s
Pass Test 8: Is Q within 1.0 of 951 umol m-2 s-1? Measured 950.459 after 8.0s
Pass Test 9: Is Q within 1.0 of 1049 umol m-2 s-1? Measured 1048.883 after 9.5s
Pass Test 10: Is Q within 1.0 of 383 umol m-2 s-1? Measured 383.375 after 11.5s
```

Fluorometer zero

The fluorometer zero test measures the effect of modulation rate on the AC and DC (i.e., modulated and non-modulated) fluorometer zero. The range of measured values is compared with an expected typical range.

```
Testing 100 Hz modulation rate
Testing 200 Hz modulation rate
Testing 500 Hz modulation rate
Testing 1000 Hz modulation rate
Testing 2000 Hz modulation rate
Testing 5000 Hz modulation rate
Testing 10000 Hz modulation rate
Testing 20000 Hz modulation rate
Testing 50000 Hz modulation rate
Testing 125000 Hz modulation rate
Testing 250000 Hz modulation rate
Pass Test 1: Is range of F < 10? Measured 4.23
Pass Test 2: Is range of F_dc < 20? Measured 6.7
```



Warnings for modulated fluorescence (F):

Is the chamber closed and empty? And clean inside?
 To assess if this range of zero shift is significant, compare:
 a) the zero difference between the flash and measuring modulation rates.
 b) the typical magnitude of F during a flash.

Warnings for dc fluorescence (F_dc):

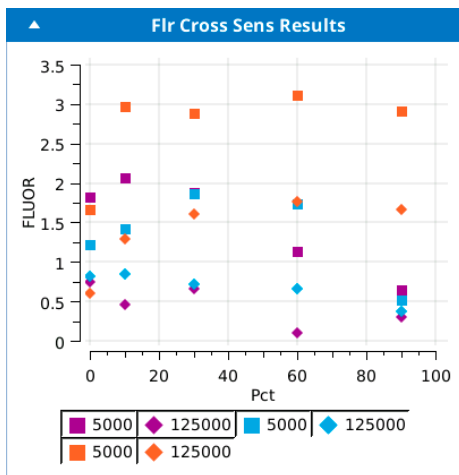
This might indicate an above-average temperature sensitivity of the fluorescence detector.

If there is no problem with the F zero shift, the following is included in the suggestion list:

The F zero shift is ok, so you can probably ignore this.

Fluorometer cross sensitivity

The fluorometer cross sensitivity test checks modulated fluorescence for any cross sensitivity with actinic light output. Red, blue, and farred are tested individually by ramping each from 0 to 90 percent of full power. The slope and intercept of the modulated fluorescence as a function of light intensity is then determined. This is done for two modulation rates, 5000 Hz and 125 kHz. Intercept values should be near 0, and slopes should be flat. A significant positive slope would indicate a cross sensitivity problem.



Checking cross sensitivity to red

Pass Test 1: red 5000Hz: Regressed intercept - measured < 5? Measured 0.5

Pass Test 2: red 5000Hz: Response slope < 0.05? Measured -0.0187

Pass Test 3: red 125000Hz: Regressed intercept - measured < 5? Measured -0.17

Pass Test 4: red 125000Hz: Response slope < 0.05? Measured -0.004

Checking cross sensitivity to blue

Pass Test 5: blue 5000Hz: Regressed intercept - measured < 5? Measured 0.71

Pass Test 6: blue 5000Hz: Response slope < 0.05? Measured -0.0114

Pass Test 7: blue 125000Hz: Regressed intercept - measured < 5? Measured 0.1

Pass Test 8: blue 125000Hz: Response slope < 0.05? Measured -0.0056

Checking cross sensitivity to farred

Pass Test 9: farred 5000Hz: Regressed intercept - measured < 5? Measured 1.3

Pass Test 10: farred 5000Hz: Response slope < 0.05? Measured 0.0001

Pass Test 11: farred 125000Hz: Regressed intercept - measured < 5? Measured 0.77

Pass Test 12: farred 125000Hz: Response slope < 0.05? Measured 0.0045

Faults are warnings, with these suggestions:

Is the chamber closed and empty? And clean inside?

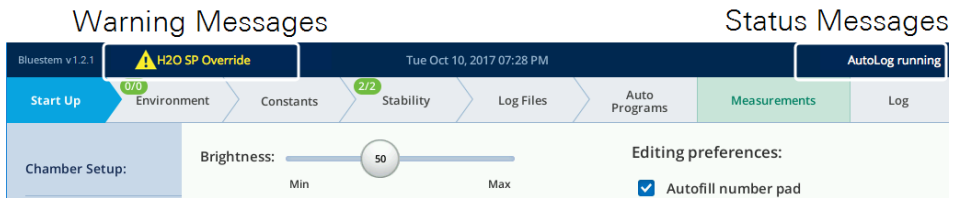
Otherwise, contact LI-COR.

Passing these tests is primarily a product of optical filter choice and electronic design - things that stay constant through the life of the fluorometer, so failures are not expected. Should one occur, it may be because of something simple, such as the presence of fluorescing material in the chamber, or - the worst case - it could mean that a fault had developed (contamination, breakage, etc.) with the optics or electronics, in which case LI-COR service is needed.

If there is a warning that can't be fixed, and factory service is not an immediate option, then you should evaluate the impact of the cross sensitivity on your measurements: Cross sensitivity to red is probably most serious, since F will be different during a flash (high red) than during a measurement (low red) just due to cross sensitivity. Try an empty chamber flash and see what the effect on F is, then determine if that effect warrants post-measurement correction. Cross sensitivity to far red will affect dark pulse readings, but only while the far red is on, and the $F_{0'}$ value is recorded after the far red is off, so if that is the only time you use far red, then it may not be an issue. Cross sensitivity to blue is hard to explain outside of an electronic fault, but if your unit has a blue issue, and if it is stable, then it would not be an issue for fluorescence measurements at a constant (actinic) light, but would be an issue when comparing (F_s , for example) two different actinic levels.

Messages

The LI-6800 is equipped with ample self-testing combined with status messages and warnings shown in the upper left corner of the display, which alert you to issues in the system. Pay attention to these. An ignored error message can lead to poor experimental outcomes and poor-quality data. Errors are color-coded to indicate severity. A **Red** message indicates a problem and **Yellow** indicates potential concern. The potential errors are given in *Table 7-2* below.



Status messages

Table 7-2. Status messages given on the display.

Message	What it means
Log Pending	Displayed during a log event, which can be very short, or a few seconds if it has to wait for a fluorescence event, or several seconds if matching has to happen.

Table 7-2. Status messages given on the display. (...continued)

Message	What it means
<autoprogram name> paused	When an Auto Program is active, but paused.
<autoprogram name> running	When an Auto Program is active.
Auto CO2	The various automatic environmental controls that can be active. These messages indicate which auto control is active. When more than one control is active the messages will cycle across the display.
Auto Fan	
Auto Flow	
Auto H2O	
Auto Light	
Auto Temp	
Auto Match	When the instrument is running the automatic match algorithm.
Manual Match	When the instrument is in the manual match mode, waiting for you to do the match and exit. (To exit this mode, press the OK button.)
SysTest: <name>	Warmup or system test is running.
AutoDry	AutoDry is an automatic drying routine that kicks in if the humidity gets > 99% in the chamber or one of the IRGAs.
PostFlash	PostFlash is a “post fluorescence flash stabilization routine”. If you do a fluorescence flash while doing leaf temperature control, and/or leaf VPD water control, right before the flash these controls are “locked” so they don’t react to the pulse in leaf temperature that will result from the flash. It lasts for 30 seconds.

Warning messages

The following are warning messages that might appear on the display.

Table 7-3. Warning messages given on the display.

Message	What it means
<List of one or more temperatures>	When a temperature sensor is reading $>80\text{ }^{\circ}\text{C}$ or $<20\text{ }^{\circ}\text{C}$, it appears in this warning list. The temperature sensors that are checked (besides Tleaf and Tleaf2) are Ts (sample cell), Tr (ref cell), Tirga (IRGA block), and Tair (chamber air temp).
Chopper Not Locked	Unable to record position of the optical chopper. Triggered by head status bit 0x2000. To resolve it, try putting the instrument into sleep mode and take it out of sleep mode. If that doesn't work, restart the instrument. If that doesn't work, call technical support.
Close Cmbr for <xxx> ctrl	When doing CO ₂ _s or H ₂ O_s control, the chamber needs to be closed. If it is not, this message appears. Close the chamber to resolve it.
CO ₂ Cartridge Low/Empty	These messages are triggered from the value of CO ₂ _FLOW_STATUS value being <0.05 . If it is in this condition with a low CO ₂ setpoint, the cartridge is likely empty. Otherwise, it is getting empty, and higher CO ₂ targets are no longer possible.
CO ₂ too high to control	
Console flow leak?	These messages indicate a potential issue with the flow path in the console, either a leak (chemical tube missing, or end cap loose), or a constriction. The specific thing that triggers these messages is the PUMP_PRESSURE_STATUS value: $> 4.9\text{V}$ indicates a constriction, and $< 0.1\text{V}$ indicates a possible leak.
Constricted flow?	
Cooler target less than Td	The target you picked for the cooler is lower than the dew point temperature. To resolve it, pick a warmer target.
Cooler Td limited	The cooler cannot lower the temperature as fast as it normally would, because that would require driving it to a temperate below the dew point temperature. This message doesn't necessarily mean condensation is a problem; it just means the cooler is not being driven as hard it could be in drier air.
Fan Fail	Cooling fan or chassis fan is not functioning as expected. Triggered by head status bit 0x0400. Cycle power. If it recurs contact technical support.
Flow_r low	Flow control is ON and F_r or F_s is $<25\text{ }\mu\text{mol s}^{-1}$. The usual reason for the Flow_s Low message is the chamber being open. One or the other of these may appear during Warmup tests.
Flow_s low	

Table 7-3. Warning messages given on the display. (...continued)

Message	What it means
Flr DC Overrange	The fluorometer has over-ranged its modulated signal. This doesn't hurt anything, it just suppresses the modulated fluorescence signal. It is also hard to do: it pretty much needs a direct glimpse of full sun reflected up into the detector to over-range it.
Flr DC Saturated	The fluorometer DC signal is saturated. Diagnostic bit 0x800.
Flr Measuring Off	The fluorometer measuring beam is off (Qmod_avg is 0.0). (The fluorometer is still modulating, but with 0 intensity. This is typically the state when you turn measuring off.)
Flr Mod Off	The fluorometer's modulation is turned off. Diagnostic bit 0x2000. This should be a very temporary condition, because whenever it is detected and the fluorometer is connected to a console, the scripts attempt turn it back on.
Flr Q Saturated	The fluorometer photodiode is saturated. Diagnostic bit 0x400.
Flr Temp Warning	The fluorometer has a temperature issue. Turn it down or off and let it cool for a few minutes. Diagnostic bit 0x1000.
Head Powered Off	The head has been put in a low power state (Power Saver (2)), but it was not done from the console.
High RH: xx% <sensor>	A high humidity warning appears when the RH is higher than 90% in the chamber or either of the IRGA cells. The value of the humidity is shown, and also the location (in terms of the temperature sensor). T_s (or T_a) is sample cell, T_r (or T_b) is reference cell, and the rest (T_{leaf} , T_{air} , T_{xchg}) are the leaf chamber. If RH exceeds 99%, the AutoDry program is launched to bring down the humidity and prevent condensation.
LI-6850 Factory Cal	Indicates that one or more factory calibration values has been altered or that the information cannot be found. Go to Start Up > Calibration and pick Factory Coefficients from the menu.
Low Battery	Install a charged battery or plug in the console power supply
Match Valve Open	A normal measurement has the match valve closed, so sample air goes to the sample IRGA and reference air to the reference IRGA. This message will be displayed anytime the valve is not in that state, which, in normal operation, will be during matching and during some system tests. To resolve it, go to match screen, select manual match, then tap OK to leave manual match.

Table 7-3. Warning messages given on the display. (...continued)

Message	What it means
No Console	This message indicates the sensor head has no "back channel" connection with the console, which means the head will not be able to control the pump, or chemical tube valves, etc. on the console. Flow control, CO ₂ control, H ₂ O control will be adversely affected. This message comes from a diagnostic bit 0x02 from the head. Cycle the power to resolve it.
Power Saver (1)	Power Saver (1) appears while the system is in the Conserve Chemicals mode, with all environmental controls turned off.
Power Saver (2)	Power Saver (2) means it is in the Conserve Power and Chemicals mode, which has Chemical Saver on and the sensor head in a low power state.
TEC Fail	The Thermo Electric Cooler has failed, or more likely tripped its overload protection. The message is triggered by status bit 0x0800. Cycle the power to resolve it.
TLeaf	Both Tleaf and Tleaf2 are missing or reading >80 °C or <-20 °C.
x Ver Flr (need y)	Software versions not matched. It means the head or fluorometer has a software version that is out of sync with the console version.
x Ver Head (need y)	Example. "1.0 Ver Head (need 2.0)"
MQTT Remote Host Closed	The user interface is unable to communicate with the instrument. If you are running an Auto Program , it will probably continue running, so, if you are in the middle of an experiment, wait for the measurement to finish running. You may be able to reconnect under Start Up > Connections , by tapping Disconnect and then Connect . If you are unable to reconnect, restart the instrument.

Troubleshooting

If you are having issues that are not indicated by the messages, look in this section for solutions.

Where are logged remarks saved?

In Excel files, remarks are stored in a different tab than your data values.

- Data are in a tab called **Measurements**.
- Remarks are in a tab called **Remarks**.

The log button on the head does not work

- Log file open?
A log file must be open to record data.
- Pressing long enough?
Press and hold until it logs the value. Holding the log button will only log a single point.

The environmental control setting I select is always being overridden

This can happen under a few circumstances:

- Is the instrument in the middle of the system tests?
You can wait until it is done or cancel the test.
- Is it running an Auto Control?
Disable the Auto Control.
- Is it running an Auto Program?
Disable the Auto Program.
- Has it overridden a humidity setpoint to prevent condensation?
If the humidity gets too high in the chamber, heat exchange, or IRGAs, the instrument will override the setpoint to protect itself from condensation.
- Is it changing the temperature source?
It has detected an implausible reading from the sensor. Check to see that the thermocouple is plugged in. See if it still works.

I enter a setpoint and the instrument does nothing

- Flow on?

Under **Environment > Flow**, be sure the pump is on. It won't necessarily turn on when you start an Auto Program or Auto Control.

Cannot maintain CO₂ setpoint

- Chemicals fresh?

When the soda lime is saturated, you may see oscillations in CO₂_r as small as 2 to 5 $\mu\text{mol mol}^{-1}$, or much greater. You can usually get a few minutes of stable operation by shaking up the columns, but the best solution is to replace the depleted chemical with fresh.

If the Stuttgarter Masse is too wet, you may see unexpected fluctuations in the CO₂ reading.

- Does the cartridge contain a gas other than CO₂?

Some 8-gram cylinders contain gases other than CO₂. If the cylinder is not labeled and you are unable to verify its contents, remove it and install a cylinder that definitely contains CO₂ gas.

Cannot achieve flow setpoint

- Reasonable setpoint?

The instrument will never get over 2000 $\mu\text{mol s}^{-1}$.

- Flow meters need to be zeroed?

Zero the flow meters under **Start Up > Calibrations > Flow/Pressure**.

- Leak in the system?

Run the leak test. If it fails, investigate to find the leak.

If you or someone else recently changed chambers, check that the chamber screws are adequately tight. Be sure the chamber is installed with the correct screws (part number 144-15344). Other non-captive screws will cause leaks.

For each chemical column, remove the cap and clean the O-ring. Wipe the threads to remove dust and debris. Also clean the part of the column that makes contact with the O-ring. Install the caps and tighten them securely.

Check to be sure the chemical columns are installed correctly. Wiggle each one a little. Take them off and reinstall each one, one at a time to ensure that they don't get mixed up.

Check the leaf gaskets to make sure they are not compressed or torn. Replace if necessary.

Chamber closing properly? Open and close the chamber a few times to make sure the mechanism is working correctly.

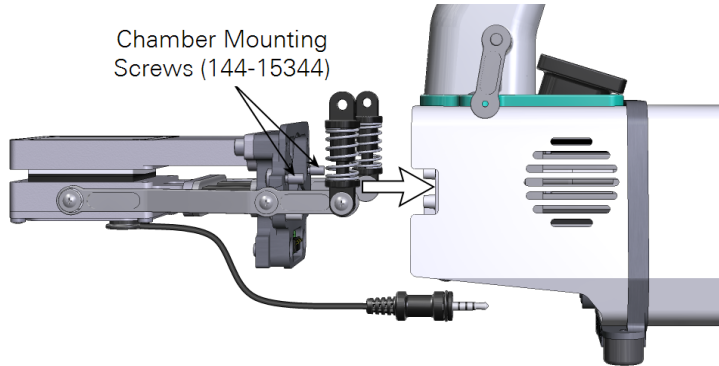
Cannot maintain humidity setpoint

- Substrate too dry?
Check the humidifier column to see if the Pall Stuttgarter Masse is adequately moist. Add 5 to 10 ml of water to each side if it is dry. See *The humidifier column* on page 2-7 for details.
- Humidifier column or desiccant column filters clogged?
Check the filters on the humidifier and desiccant columns. If they are visibly soiled, replace them with filters from the spares kit (see *Column air filters* on page 8-26).

Failing a leak test

Leaks usually come to your attention by causing a leak test, flow test, or pump test failure (**Start Up > Warmup/System Tests**), low **Flow** reading (mass flow to chamber), or a low **Flow_s** or **Flow_r** reading (flow through the sample/reference gas analyzer). Also, one of the following warning messages may appear on the upper left corner of the display: **Console Flow Leak?**, **Flow_s Low**, or **Flow_r Low**.

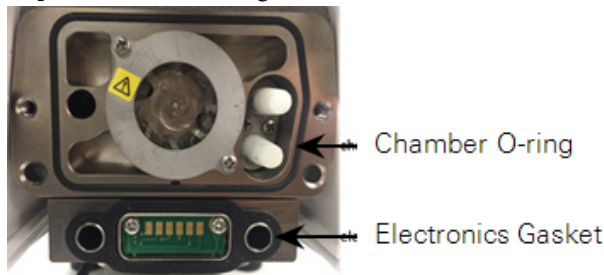
- 1 If you can achieve the **Flow** setpoint, but **Flow_s** is near zero or much lower than the flow to the chamber, it is likely a chamber leak. Check the following things:
 - Make sure that the chamber is installed tightly enough with two Philip screws (part number 144-15344) and adequately closed.



- Check the leaf gaskets on the chamber. If the gaskets are flattened or torn, change them. Black neoprene gaskets can recover if left uncompressed overnight, but the white foam gaskets may not. Advanced polymer gaskets recover more quickly.

Spare gaskets are included in the chamber spares kit. See *Gasket replacement* on page 8-13 for details.

- Make sure that the thermocouple is installed and the O-ring (part number 192-14435) on the chamber bottom that seals the thermocouple is in place.
- The lower chamber may have two flexible black ducts (depends on chamber type, part number 6368-345). They may wear out over time. Change if necessary. Damage to the ducts should rarely happen, but if they are kinked during removal or installation, they may develop leaks. Ensure that the bead of the duct seats in the groove.
- Remove the chamber from the head and you will see the big chamber O-ring (part number 192-14438) and electronics gasket (part number 6568-327). Replace them if damaged.



- If using the leaf chambers that have the removable aperture inserts (6800-12A & 6800-01A), check if the apertures are seated firmly in the top and bottom chambers. Make sure the aperture O-rings are in place and good condition.

Spare O-rings are included in the chamber spares kit. See *Chamber O-rings and gaskets* on page 8-15 for details.

- Change the chamber overpressure (ΔP) setting (**Environment > Flow**) to 0 kPa or 0%, if it was targeting 0.2 kPa or close. If that results in the increase of the **Flow_s** or decrease of the percentage of apparent leak, contact LI-COR.

2 If **Flow_s** / **Flow_r** is low and **Flow** cannot reach its setpoint, check the following:

- Make sure the humidifier and desiccant columns (part number 9968-225) are present and securely attached to the console (they should be removed during transportation and storage). Check if there are any cracks in the walls of the chemical columns.
- Check to see if any chemical debris or anything else has accumulated on the threads of the cap and the tube. This will prevent the cap from getting a good seal. This may especially be the case with the humidification column, where difficult-to-see ceramic particulates can accumulate. Wipe the threads, rim, and O-ring with a moist paper towel to remove any debris. This will also help keep the column and cap from being damaged.

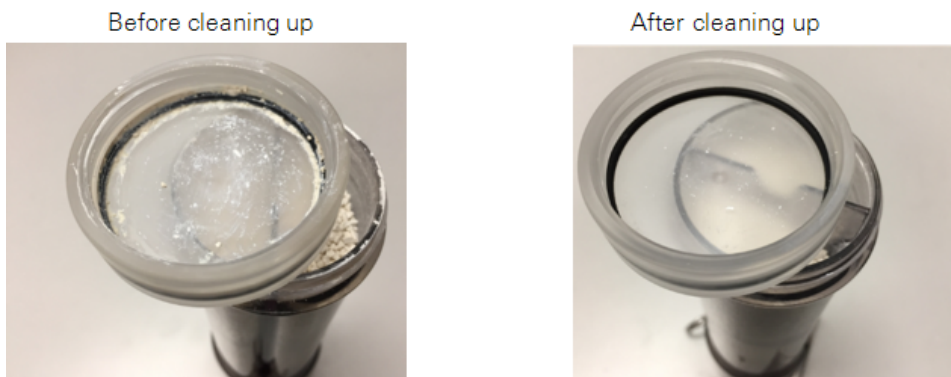


Figure 7-2. This example may be a bit exaggerated but it happens. The column on the left will likely have leaks due to the particles. You may have difficult-to-see particles that cause leak, too.

- To isolate a leak to either the humidifier or desiccant tubes, try replacing the tube in question with the soda lime tube (the soda lime position can remain empty – it is on the vacuum side of the pump so will not cause a leak). If that fixes the leak, you have found the culprit.
- Make sure that the O-ring (part number 192-14541) inside the bottom column cap is in place.
- Each chemical column has two filters (part number 300-14319) held in place with rubber seals (part number 6368-225). Check if any of the two air filters are missing.
- Make sure that the air supply tube is securely connected to the console outlet and sensor head inlet. Hold onto the Bev-a-line tubing (not on the Quick-connect Coupler) and pull. It should not come loose. If it does, firmly reconnect it (make sure the Quick-connect Coupler is pushed in to the end until you hear a click sound). Continued failure may indicate a new Quick-connect Coupler (part number 300-07125) is needed. Check each end of the air supply tube.

If you have done all of the above, but flow problems persist, contact LI-COR.

Unreasonable results

The first indicator of trouble, to those who are new to gas exchange, will typically be the values that the user is paying attention to: photosynthesis, conductance, C_i , or others. The tendency may be to blame the computer, but usually it is just doing what it is told, and the unbelievable results are coming from unbelievable inputs. You will need to look behind the computations and determine which input is unbelievable and why.

Unbelievable photosynthesis rates

If photosynthesis seems to be jumping around, try these suggestions:

- Are you being impatient?
Right after a change in input conditions, such as a big change in the CO₂ control setting, there will be a short period (up to 2 minutes) where the photosynthetic rate will be nonsense until the gas analyzers achieve the new equilibrium values.
- What is the magnitude of the variation?

There will always be some variation in the displayed value of any measured or computed quantity. Is the variation excessive? Is it due to normal noise of the analyzers? Remember that at low photosynthesis rates, the noise in the CO₂ differential will become more and more significant. So, is the variation in ΔCO_2 greater than 0.1 $\mu\text{mol mol}^{-1}$?

- Watch those flow rates.
For purposes of troubleshooting, operate with a fixed flow rate. Disable the **Flow Assist** control (under **Environment > H₂O**).
- Is the input stable?
Watch the reference CO₂ (CO_{2_r}) for 15 seconds. How much does it vary? Expect variations near 0.1 $\mu\text{mol mol}^{-1}$ at ambient concentrations. If it is much more than that, you may have a problem.
- Are the sample measurements (CO_{2_s}) stable?
If the reference values are stable, but the sample values are not, try running the leak test.

Power on problems

There are no user-replacable fuses in the instrument. If you suspect that a fuse has blown contact LI-COR or a LI-COR representative.

Section 8.

Calibration and maintenance

The difference between data and good data

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Calibration and maintenance

This chapter describes the maintenance tasks that may be required in the normal course of operating the LI-6800. Any procedures other than those described here should be performed by LI-COR personnel or a representative.

User calibration

The **Start Up > Calibration** tab shows the type and dates of all previous calibrations. In addition to the **History**, there are four options:

- **IRGAs**, which is where you set the Zero calibrations. See *IRGA zeros* below.
- **CO₂ Spans**, which is where you set the CO₂ span calibrations. See *Carbon dioxide span* on page 8-8.
- **H₂O Spans**, which is where you set the H₂O span calibrations. See *Water vapor spans* on page 8-9.
- **Flow/Pressure**, which is where you set the other flow meter and pressure sensor calibrations. See *Flow meter and chamber pressure sensor zeros* on page 8-11.
- **Diagnostics** provides a summary of information that is valuable when you need to evaluate the overall performance of the instrument. See *Diagnostics* on page 8-12.

IRGA zeros

The **Start Up > Calibrations > IRGA Zeros** page is where you set the zero of the gas analyzers. A zero calibration shifts the entire response curve a fixed amount. For example, if the IRGA reads 2 $\mu\text{mol mol}^{-1}$ with CO₂-free air in the cell, and you do a zero adjustment to make it read 0, then you have changed the response by at least -2 $\mu\text{mol mol}^{-1}$ at *every* concentration (there are some subtleties, it might be -4 at high concentrations, for example).

Caution: Never zero without tank gas or fresh chemicals. Old chemicals, even in new packaging, can expire and lose their ability to scrub gases. If you have doubts about the quality of chemicals or gases, do not zero the LI-6800. If zeroing with chemicals, keep the following in mind:

- Know when and from where the chemicals were purchased.
- Know how and where they were stored.
- Just because you changed the chemicals recently does not necessarily mean they are good.

Checking the zeros

You can check the zeros any time during measurements without entering calibration mode. The chamber can be open or closed, with or without a leaf (although if there is a leaf in the chamber it may be starved of CO₂).

1 Begin with the CO₂ zero.

Zeros are checked under **Start Up > Calibrations**. Tap **Begin Calibration**.

Note that the cell getting flow is the cell that gets zeroed. When checking the zero, flow to the leaf chamber is bypassed. Use the **Change Valves** button to set which cell (Sample or Reference) is getting flow.

- Before checking the zero, check that the soda lime and desiccant are fresh.
- If zeroing CO₂ alone, then scrub CO₂ and bypass H₂O.

The screenshot shows the 'IRGA Zeros' calibration screen. At the top, there are navigation tabs: Start Up, Environment, Constants, Stability, Log Files, Auto Programs, Measurements, and Log. Below the tabs, there's a 'Chamber Setup' section with a dropdown menu set to 'IRGA Zeros' and a 'Change Valves' button. The 'Configuration' section contains instructions: 'If zeroing CO₂ alone, then scrub CO₂ and bypass the desiccant.' and 'If zeroing H₂O alone, then scrub CO₂ and H₂O.' Below this, there are four measurement cards for CO₂-S, CO₂-R, H₂O-S, and H₂O-R, each showing a 'Meas' value and a 'Coeff' value. The 'Coeff' values are 0.87725, 0.85897, 0.97884, and 0.97646. There are also checkboxes for 'Pump On' (checked) and 'Manually set the zero coefficients for H₂O IRGAs (Advanced)'. On the right side, there are five buttons: 'Zero CO₂,S', 'Zero CO₂,R', 'Zero H₂O,S', 'Zero H₂O,R', and 'Exit Calibration'.

2 If everything is working properly, you should see CO₂ values that are very close to zero.

3 Check the H₂O zero.

When checking the H₂O zero, you'll scrub H₂O.

4 Watch the H₂O reading for the cell that is getting flow.

It will approach zero more slowly than the CO₂ reading. Rather than wait the 10 or 20 minutes to get a really good zero, use your judgment. If, after a minute or so, the reading is down to 0.2 or 0.3 mmol mol⁻¹ and falling slowly, that's good enough. Clearly, if it's negative and falling after only 1 minute, it will be going too low, and re-zeroing may be in order.

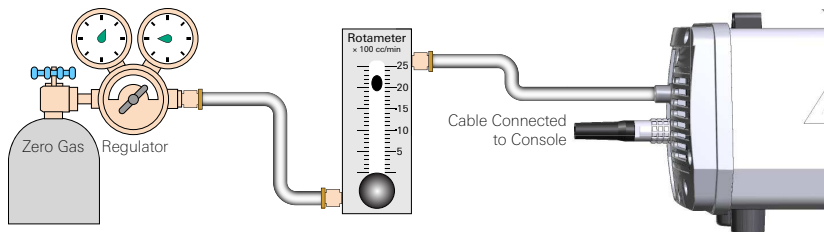
- 5 Be sure to check the zero for both CO₂ and H₂O in both the sample and reference IRGAs.

Zeroing with gases

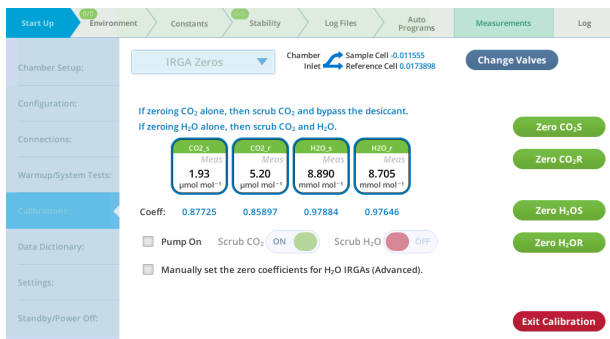
If you have access to a quality calibration gas that has a known concentration of 0 ppm CO₂, you can use that to set the CO₂ and H₂O zeros.

- 1 Connect the head cable between the console and head. Power on the instrument.
The leaf chamber can be open or closed.
- 2 Plumb the zero gas to the analyzer inlet.

The zero gas can be CO₂-free air or nitrogen. Set flow from the tank to about 2 liters per minute (at least 0.5 liters per minute if you want to conserve gas). Be sure to use a flow regulator. There is a piece of Bev-a-line tubing and a quick-connect fitting in the spares kit.



- 3 Begin with the CO₂ zero.
Zeros are set Under **Start Up > Calibrations**. Tap **Begin Calibration**.
The procedure will zero the IRGAs. Note that the cell getting flow is the cell that gets zeroed. During zeroing, flow to the leaf chamber is bypassed.
- 4 Partition the flow so it is through both analyzers.
Tap **Change Valves** so flow goes through both Sample and Reference cells.
- 5 Clear the **Pump On** box, since you are relying on the gas for the flow.
- 6 If everything is working properly, you should see CO₂ and H₂O values that are very close to zero.



7 Check the readings for H2O_r and H2O_s.

When the readings have stabilized to your satisfaction, tap the **Zero H₂OR** and **Zero H₂OS** buttons. Always do H₂O first, since the water vapor affects the CO₂ measurement.

8 Check the readings for CO₂_s and CO₂_r.

When the readings have stabilized to your satisfaction (they should become stable fairly quickly) tap **Zero CO₂S** and **Zero CO₂R**.

9 Exit calibration if you are satisfied with the results.

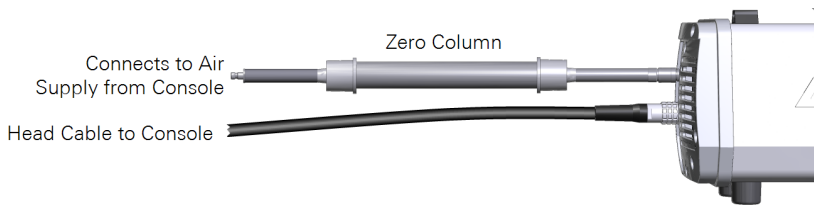
Zeroing with chemicals

Although it is best to set the zero using high-quality calibration gases from a tank, you can zero with the zero kits if calibration gases are not available. Two columns are included—one for soda lime, and one for a desiccant. Follow these good practices to ensure that you achieve a reliable zero.

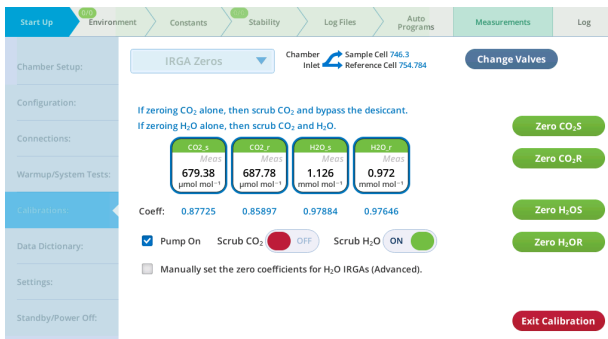
Caution: Never zero without tank gas or fresh chemicals. Old chemicals, even in new packaging, can expire and lose their ability to scrub gases. If you have doubts about the quality of chemicals or gases, do not zero the LI-6800. If zeroing with chemicals, keep the following in mind:

- Know when and from where the chemicals were purchased.
- Know how and where they were stored.
- Just because you changed the chemicals recently does not necessarily mean they are good.

- 1 Connect the cable between the console and head.
Leave the hose disconnected. Power on the instrument. The leaf chamber can be open or closed.
- 2 Fill the zero column (from the spares kit) with either regular or indicating Drierite. **Do not attempt to zero H₂O with Sorbead® Orange CHAMELEON® or silica gel beads.**
Always do water vapor first, since water vapor affects the CO₂ measurement.
- 3 Install the zero column, which is filled with desiccant on the head air inlet.
The zero column goes in line between the console and head.



- 4 Begin with the H₂O zero.
Zeros are set Under **Start Up > Calibrations > IRGA Zeros**. Tap **Begin Calibration**.
Note that the cell getting flow is the cell that gets zeroed. During zeroing, flow to the leaf chamber is bypassed.



- 5 Partition the flow so it is through both analyzers.
Tap **Change Valves** so flow goes through both **Sample** and **Reference** cells.
- 6 If everything is working properly, you should see H₂O values that are very close to zero.
- 7 Tap the **Zero H₂OS** and **Zero H₂OR** buttons.

Check the results to see if they are satisfactory. If you're satisfied with the results, move on to the CO₂ zero.

8 Fill the second zero column with Soda Lime.

9 Connect it in line between the console and head, as before, and begin the zero procedure.

If everything is working properly, you should see CO₂ values that are very close to zero.

10 Tap the **Zero CO₂S** and **Zero CO₂R** buttons when the values are stable.

Start Up Environment Constants Stability Log Files Auto Programs Measurements Log

IRGA Zeros Chamber Inlet Sample Cell 747.661 Reference Cell 754.273 Change Valves

Chamber Setup: Configuration: Connections: Warmup/System Tests: **IRGA Zeros** Data Dictionary: Settings: Standby/Power Off:

If zeroing CO₂ alone, then scrub CO₂ and bypass the desiccant.
If zeroing H₂O alone, then scrub CO₂ and H₂O.

CO ₂ S	CO ₂ R	H ₂ O S	H ₂ O R
0.55 μmol mol ⁻¹	3.93 μmol mol ⁻¹	8.783 mmol mol ⁻¹	8.578 mmol mol ⁻¹
Coeff: 0.87725	0.85897	0.97884	0.97646

Pump On
 Scrub CO₂
 Scrub H₂O
 Manually set the zero coefficients for H₂O IRGAs (Advanced).

Zero CO₂S
Zero CO₂R
Zero H₂O S
Zero H₂O R

Exit Calibration

11 Check the values to confirm that the zero is good.

12 **Exit Calibration** if you are satisfied with the results.

Stow away the zero tubes for future use. You don't need to empty the chemicals from them. Write the date and the chemicals on the tubes, install caps on the ends, and keep them for the next time you need to zero.

Span overview

The CO₂ and H₂O **Spans** are user calibrations in which you check the reading with a known concentration in the cell. What does the span calibration do? Suppose an IRGA reads 990 μmol mol⁻¹ when there is 1000 μmol mol⁻¹ in the cell. If you adjust the span to correct this, you will have increased the response by 10 μmol mol⁻¹ at 1000, but at 500 μmol mol⁻¹, the response will be increased by only 5 μmol mol⁻¹, and at 2000 μmol mol⁻¹, it will be about 20. (The numbers are not quite right, since the span adjustment is linear with IRGA signal, not concentration.) At 0 concentration (or rather, at zero voltage from the IRGA detector), a span adjustment has no effect at all.

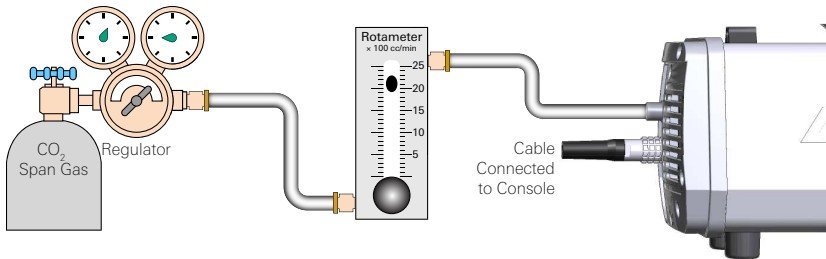
Carbon dioxide span

To set the span of the CO₂ analyzer, *you'll need gas with a known concentration of CO₂*. Generally, this is provided by a tank of compressed CO₂ in air or nitrogen that has been certified, or (even better) that has been measured using a correctly calibrated gas analyzer. The concentration should be at or above where you work most of the time, so 500 μmol mol⁻¹ is fine.

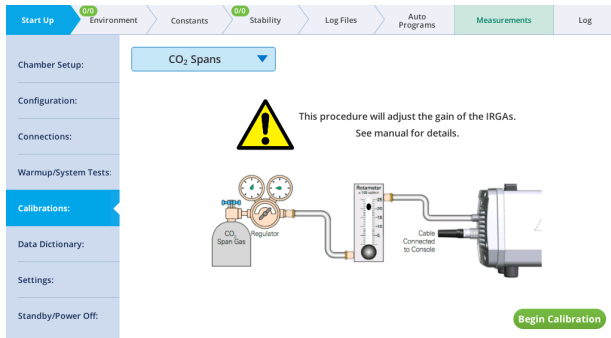
- 1 Connect the head cable between the console and head. Power on the instrument.
The leaf chamber can be open, closed empty, or closed on a leaf (although this is not recommended because it will alter the leaf's response).

- 2 Plumb the span gas to the analyzer inlet.

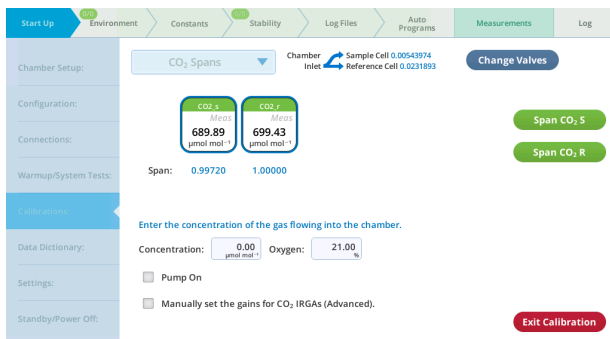
There is a piece of Bev-a-line tube and a quick-connect fitting in the spares kit for this purpose. The span gas can be CO₂ in air or nitrogen. Set flow from the tank to about 2 liters per minute (at least 0.5 liters per minute if you want to conserve gas). Be sure to use a flow regulator.



- 3 Go to **Start Up > Calibrations and select CO₂ Spans. Tap Begin Calibration.**



- 4 Partition the flow so it is through both analyzers (tap Change Values).
- 5 Check the readings for CO₂_r and CO₂_s.



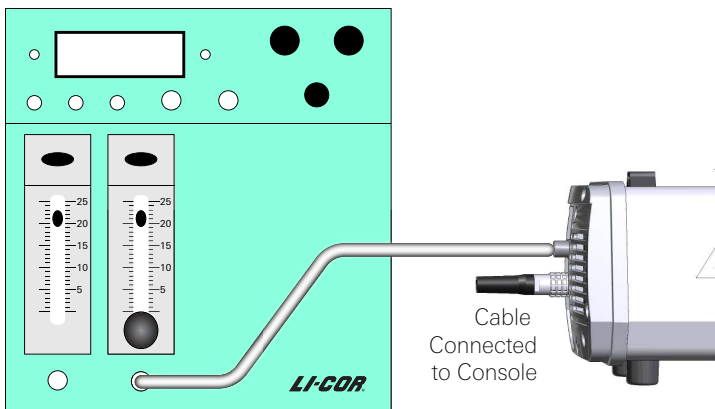
- 6 Enter the **Concentration** of the span gas ($\mu\text{mol mol}^{-1}$).
- 7 Enter the % **Oxygen** in the span gas (21% if your span gas is CO_2 in air, or 0% if using CO_2 in nitrogen).
- 8 When you are satisfied that the readings have stabilized, tap **Span CO_2 S** and **Span CO_2 R**.

Water vapor spans

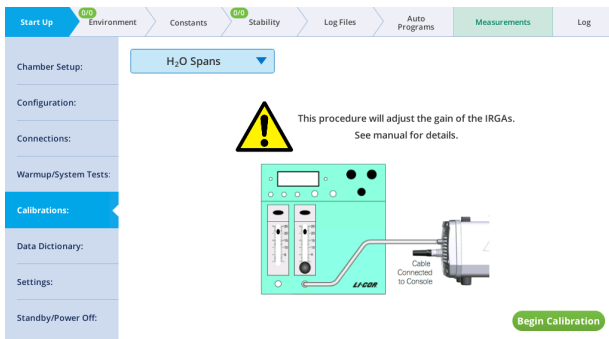
To set the span of the H_2O analyzer, you'll need a gas stream with a known concentration of H_2O . The best choice for this is the LI-610 Dew Point Generator.

Caution: If you don't have an LI-610, or some device of similar accuracy, do not adjust the IRGA span values for water.

- 1 Setup the LI-610 for an appropriate dew point.
Subtract about 5°C from the room temperature and use that for the target dew point temperature. Wait until the condenser's temperature (as monitored on the LI-610) reaches this target. The reason for this 5°C "buffer" is to avoid condensation in the line between the LI-610's condenser and the IRGA. If condensation happens, you will have large errors.
- 2 Set a flow rate of about 0.5 liters per minute from the LI-610.
- 3 Attach the LI-610 air to the IRGA air inlet.

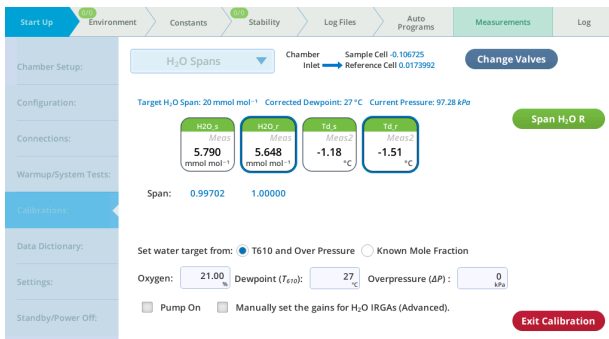


4 Go to Start Up > Calibrations and select H₂O Spans.



5 Tap Begin Calibration.

6 Wait for equilibrium. Watch the values for H₂O_s and H₂O_r. It should take 3 to 5 minutes for equilibration.



- 7** Set the target from: Select either **T610 and overpressure** if you're using the LI-610, or **Known Mole Fraction** if you are using another humidity source.
- 8** Enter the percent oxygen (21% in ambient air).
- 9** Enter the **Dewpoint** temperature (T_{610}) overpressure (ΔP).
- 10** Tap **Span H₂O S** and **Span H₂O R** to set the spans.
You can span the analyzers one at a time or both at the same time. Tap the **Change Valves** button to select a different analyzer or both.

What can go wrong setting the span?

The span factors should be within 0.05 of 1.0 (that is, 0.95 to 1.05). The farther out of this range you go in attempting to make the analyzer read the correct concentration, the more likely it is that something is wrong, such as one of the following:

- **Badly zeroed analyzer?**
Make sure you have a good zero before setting the span.
- **Concentration not what you think it is?**
For CO₂, has the span gas ever been independently checked? Don't believe even the most reputable vendor of calibration gases; after all, someone could have accidentally put the wrong label on the tank sent to you.
For H₂O, is there water in the condenser? Are you asking for a target that is at or above room temperature? (You won't get to that dew point, of course, but you will get in trouble in the form of condensation somewhere in the line.)
- **Leak in the chamber?**
The chamber has to be well sealed for this to work.
- **Plumbing mistake?**

Flow meter and chamber pressure sensor zeros

This is where you zero the flow meter and chamber pressure sensor.

To zero the flow meter:

- 1** Under **Start Up > Calibration**, tap **Flow Meter Zeroing**
- 2** Tap **Begin Calibration**
- 3** Zero the sensors
Tap each of the four buttons to zero the corresponding component.

Diagnostics

The diagnostics tab provides an overview of performance and calibration settings for the sample and reference gas analyzers for both CO₂ and H₂O.

Select Values: <input checked="" type="radio"/> CO ₂ <input type="radio"/> H ₂ O			
CO ₂ _s:	692.03 $\mu\text{mol mol}^{-1}$	CO ₂ _r:	695.86 $\mu\text{mol mol}^{-1}$
CO ₂ _a:	692.03 $\mu\text{mol mol}^{-1}$	CO ₂ _b:	695.86 $\mu\text{mol mol}^{-1}$
Match:	0 $\mu\text{mol mol}^{-1}$		
Ts:	28.78 °C	Tr:	28.63 °C
CO ₂ A Span 1:	0.99720	CO ₂ B Span 1:	1.00000
CO ₂ A Span 2:	0.00000	CO ₂ B Span 2:	0.00000
abs_c_a:	0.128206	abs_c_b:	0.130125
CO ₂ A Zero:	0.87725	CO ₂ B Zero:	0.85897
Wc_s:	29286.9	Wc_r:	31893.3
Wco_s:	29544.3	Wco_r:	31568.7

For the CO₂ Analyzer

Variable	Description
Change Valves:	Changes flow from Sample to Reference.
CO ₂ _s: CO ₂ _r:	CO ₂ measured in the sample (s) and reference (r) analyzers.
CO ₂ _a: CO ₂ _a:	CO ₂ measured in the gas analyzers, after the match is applied.
Match:	The adjustment applied to the sample gas analyzer.
Ts: Tr:	Gas analyzer temperatures for the sample and reference.
CO ₂ A Span 1: CO ₂ B Span 1:	Span value 1 for the reference and sample analyzers.
CO ₂ R Span 2: CO ₂ S Span 2:	Span 2 for the reference and sample analyzers (not used).
abs_c_a: abs_c_b:	Absorption of infrared radiation by CO ₂ .
CO ₂ A Zero: CO ₂ B Zero:	Zero for the CO ₂ reference and sample analyzers.
Wc_a: Wc_b:	
Wco_a: Wco_b:	

For the H₂O Analyzer

Variable		Description
H ₂ O _s :	H ₂ O _r :	H ₂ O for the sample (s) and reference (r) analyzers.
H ₂ O _a :	H ₂ O _b :	H ₂ O for the gas analyzers after the match is applied.
Ts:	Tr:	Gas analyzer temperatures for the sample and reference.
H ₂ O A Span 1:	H ₂ O B Span 1:	Span value 1 for the reference and sample analyzers.
H ₂ O R Span 2:	H ₂ O S Span 2:	Span 2 for the reference and sample analyzers (not used).
abs_h_a:	abs_h_b:	Absorption by H ₂ O
H ₂ O R Zero:	H ₂ O S Zero:	Zero for the H ₂ O reference and sample analyzers.
Ww_a:	Ww_b:	
Wwo_a:	Wwo_b:	

Leaf chambers

This section describes the maintenance of LI-6800 chambers.

Foam gasket care

It is important to take care of the foam gaskets on the leaf chambers. Never latch the chamber closed when it is not in use, as the gaskets will stay compressed if you leave the chamber closed for several hours. Black neoprene will recover after several hours, but the white foam gaskets recover more slowly (if ever). Advanced polymer gaskets are more resilient.

Gasket replacement

The two gaskets on the leaf chamber should be replaced if there is visible damage that interferes with sealing. It is important that you peel off the old gaskets correctly—there is a thin film below the gasket called the *carrier* that holds the adhesive. With a fingernail or the flat edge of a knife, pry up a corner of the carrier. If you only pry up the gasket and adhesive, the carrier will remain on the chamber, and is difficult to scrape off. If you get underneath the carrier, the whole gasket will come off quite readily.

Frequently there is residual adhesive material on the chamber after the gaskets are removed. This can be cleaned up with Goo Gone[®], acetone or another solvent if the surface is painted metal. Never use solvents on the plastic chamber parts.

The replacement gaskets are installed by removing the paper backing over the adhesive. Watch for channels where the adhesive sticks to itself when you remove the backing. If you stretch the gasket slightly, the channels disappear. If you apply the gasket to the chamber with channels, you'll have leaks.

For best results, use advanced polymer gaskets on both the upper and lower chambers. If you use foam gaskets, use white gaskets on the upper chamber and black ones on the lower chamber. The white gasket does not recover nearly as well as black neoprene after being compressed, and it is much more difficult to remove.

Propafilm replacement

The clear-top leaf chamber (3×3 cm), large leaf chamber (6×6 cm), small plant chamber, and sprig blocks use Propafilm (Innovia Films Inc., Atlanta, GA) as the transparent chamber top. You will need to replace the film if it becomes torn, punctured, or excessively dirty. Replacement film and double-sided tape are provided in the spare parts kits for chambers. To replace the Propafilm:

- 1** Remove the light source from the chamber, if necessary.
- 2** Remove the old Propafilm and tape from the chamber.

The tape has a fairly strong adhesive; it does not peel off readily. Use a solvent such as acetone to help dissolve the adhesive. Do not use a blade or other sharp instrument to remove the tape, as you could damage the surface of the chamber, making it difficult to achieve a tight seal with the new Propafilm.

- 3** Prepare the new tape.

Cut a piece of the double-sided tape that is slightly larger than the leaf chamber. Make it 6 × 6 cm for the small chamber or about 10 × 10 cm for the large chamber. Lay on a flat cutting surface, adhesive side up. The tape may be curled slightly; hold the corners down with cellophane tape.

- 4** Attach the tape and trim.

Lower the top surface of the chamber onto the tape and press firmly. Trim the tape around the outer and inner edges of the chamber. To get a clean cut, use a fresh blade and make your first cut as close to the edge as possible.

5 Prepare a piece of Propafilm.

Cut an oversized piece of Propafilm. Stretch the film on a flat, clean surface until taut. You may want to tape the corners with cellophane tape to secure it to the cutting surface.

6 Remove the backing from the tape and attach the propafilm.

Peel the backing from the tape and smooth any bubbles that may have formed. Lay the leaf chamber onto the Propafilm. Turn the chamber over and smooth the film. Bubbles can be lanced and smoothed.

7 Trim to size.

Trim the film around the outer edge of the chamber. Make your first cut as close to the edge as possible.

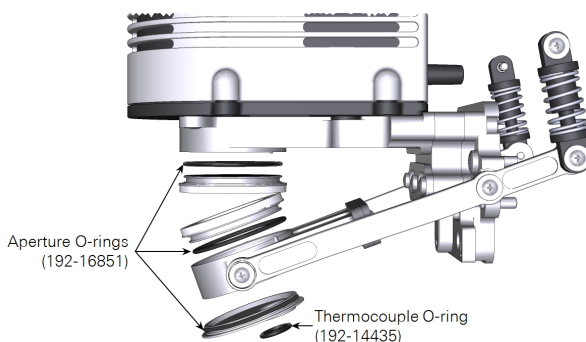
8 Reassemble the leaf chamber.

Chamber O-rings and gaskets

Each chamber includes a spare parts kit with replacement gaskets and O-rings. If the instrument fails a leak test, you may need to replace a damaged gasket or O-ring.

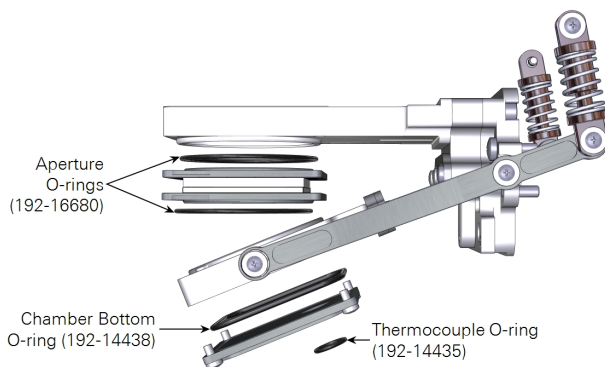
Fluorometer

Spare parts for the fluorometer are included in the fluorometer spares kit (see *Multiphase Flash™ Fluorometer* on page 1-15) or the instrument spares kit.



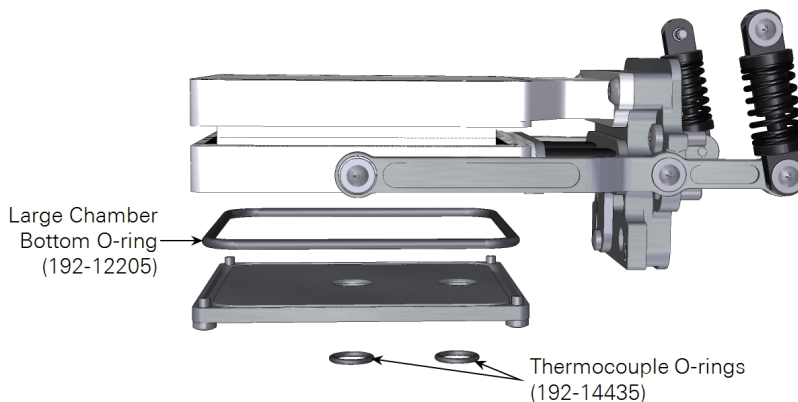
Clear-top leaf chamber

Spare parts for the small leaf chamber are included in the clear-top leaf chamber spares kit (see *Clear-top leaf chamber (3×3 cm)* on page 1-15) or the instrument spares kit.



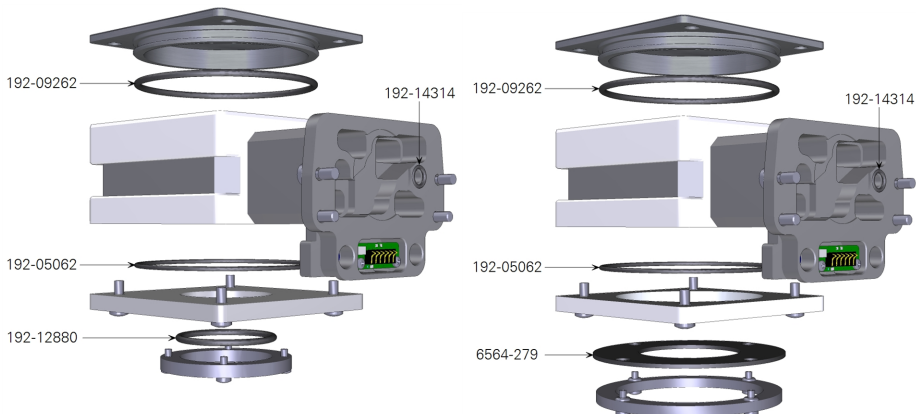
Large leaf chamber

Spare parts for the large leaf chamber are included in the large leaf chamber spares kit (see *6×6 cm clear-top chamber* on page 1-16) or the instrument spares kit.



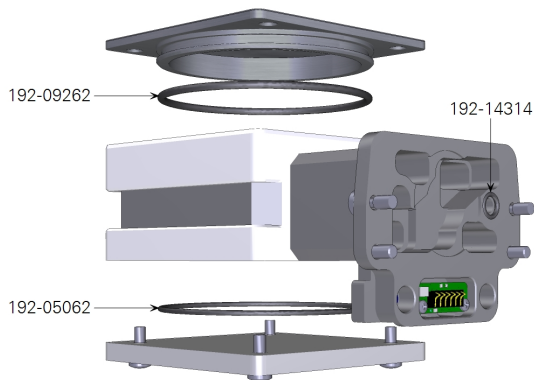
Small plant chamber

Spare parts for the small plant chamber are included in the small plant chamber spares kit (see *Small plant chamber* on page 1-18) or the instrument spares kit.



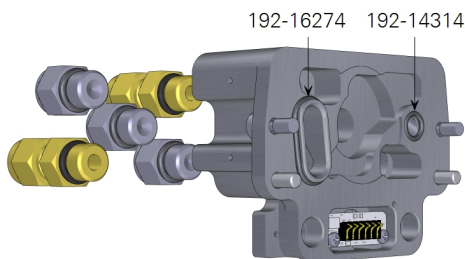
Bryophyte chamber

Spare parts for the bryophyte chamber are included in the bryophyte chamber spares kit (see *Bryophyte chamber* on page 1-18) or the instrument spares kit.



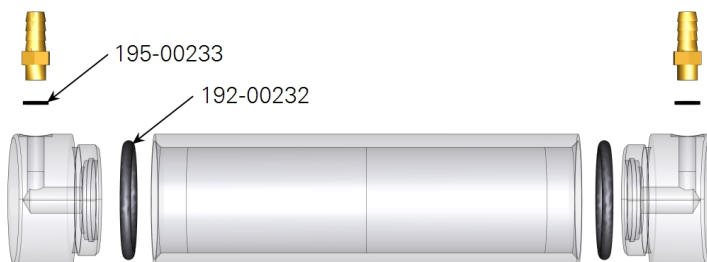
Custom chamber adapter

Spare parts for the custom chamber adapter are included in the custom chamber spares kit (see *Custom chamber adapter* on page 1-19) or the instrument spares kit.



Insect chamber

Spare parts for the custom chamber adapter are included in the insect chamber spares kit (see *Insect respiration chamber* on page 1-19) or the instrument spares kit.



Leaf temperature thermocouple

The leaf temperature thermocouple should require little maintenance. If it is not providing plausible measurements, it may need to be replaced.

Thermocouple maintenance

Readings from the leaf temperature thermocouples should be checked periodically, usually at the beginning of each day. A broken thermocouple is easy to replace, and if you have a broken thermocouple it's better to find out before

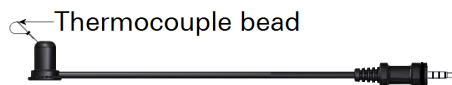


Figure 8-1. Leaf temperature thermocouple and connector.

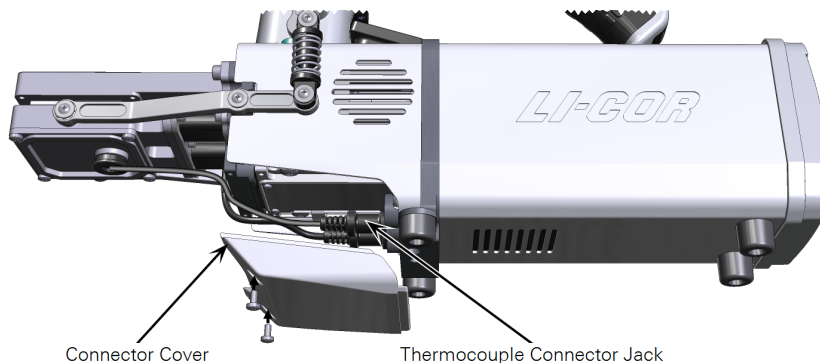
you make your measurements than after. To check it, go to **Start Up > System Tests > Sensor Check** and run the test.

Thermocouple replacement

The leaf temperature thermocouple (part number 9968-245) is mounted in a plastic holder that is inserted from below the bottom half of the leaf chamber. The thermocouple is terminated with a male 1/8" connector. If the thermocouple is damaged, the entire assembly is replaced. A failed thermocouple is indicated by leaf temperature readings around 9999.9.

To replace the leaf temperature thermocouple:

- 1 Remove the connector cover.
It is held in place with two screws.



- 2 Unplug the thermocouple.
Remove the thermocouple connector jack by pulling straight out.
- 3 Remove the thermocouple from the leaf chamber.
Pull straight down and out of the leaf chamber. You may have to wiggle it a bit if it is tight. Do not pull on the thermocouple cable.
- 4 Insert the new thermocouple.
Moisten the thermocouple retainer O-ring slightly, or use a minuscule amount of silicone grease. This will make it easier to install the new thermocouple.

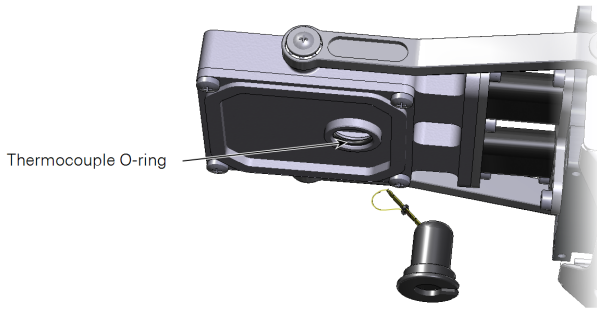


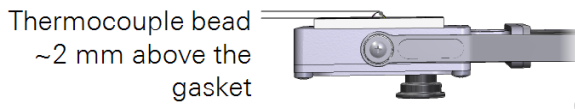
Figure 8-2. Maneuver the thermocouple through the lower chamber plate. Apply a dab of silicone grease to the O-ring to ease installation, if needed.

5 Plug in the connector.

Connect the thermocouple connector to the T1 or T2 connector. A dab of silicone grease will make this easier to install and remove.

6 Position the thermocouple.

Position the thermocouple so that the bead extends just above the lower foam gasket when viewed from the side. This will ensure that the leaf is in contact with the thermocouple when the chamber is closed.



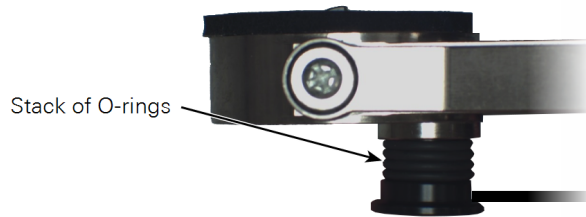
If you are using an energy balance to compute leaf temperature, then position the thermocouple lower, so that it will not touch any leaf material.

7 Install the connector cover.

8 Check the thermocouple reading.

Go to **Start Up > System Tests > Sensor Check** and run the test.

Note: Install 2 to 4 O-rings from the spares kit on the thermocouple shaft, as shown, to maintain the correct position. The required number of O-rings will depend on the angle of the thermocouple bead.

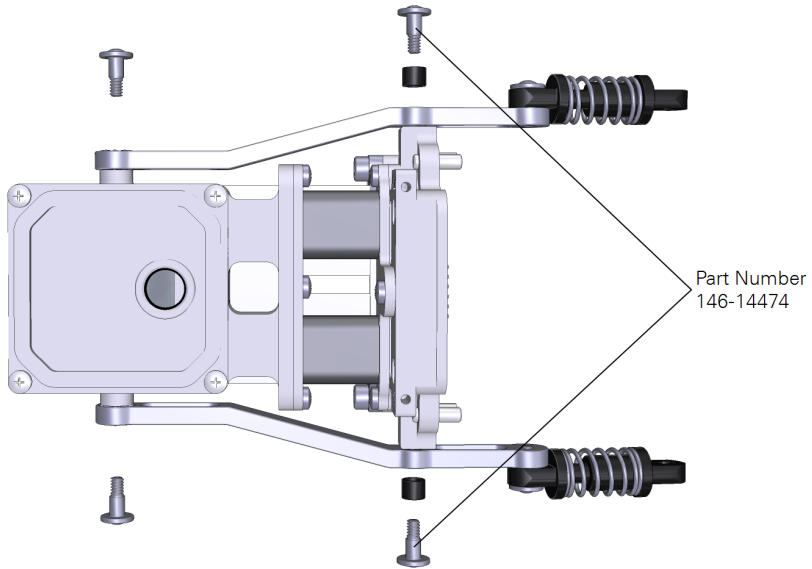


Replacing the chamber ducts

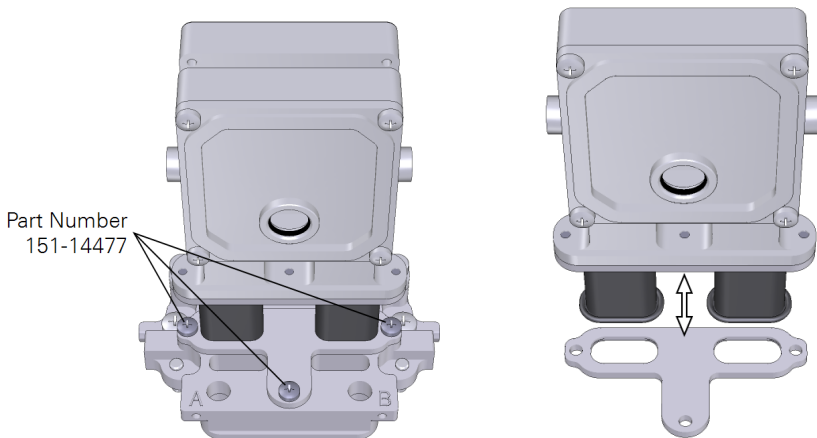
The lower chamber has two flexible ducts. The ducts may wear out over time. Follow these steps if you need to replace the ducts. This procedure describes the 3×3 cm clear top chamber, but the procedure is the same for other chambers.

- 1** Remove the chamber from the head.
See *Removing a chamber* on page 5-2. Set the chamber top-down on your workbench.
- 2** Remove the hinge arms from the chamber.

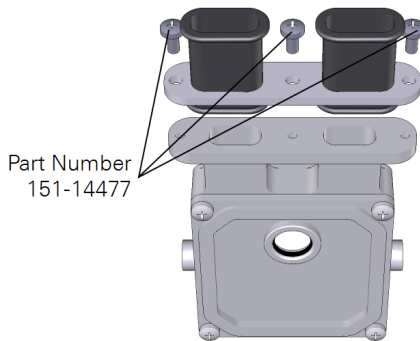
Caution: Avoid kinking the ducts. If they are kinked during removal or installation they may develop leaks.



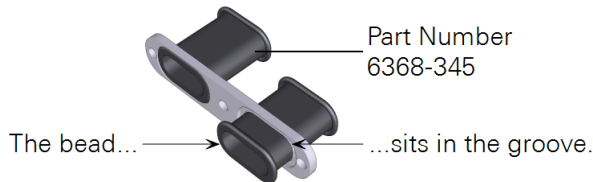
- 3** Remove the three screws that hold the ducts to the chamber.
You will have to shift the lower chamber out of the way to access the screws. After the screws are removed the lower chamber will separate from the rest of the chamber. Work the ducts out of the plate.



- 4** Remove the ducts from the lower chamber.

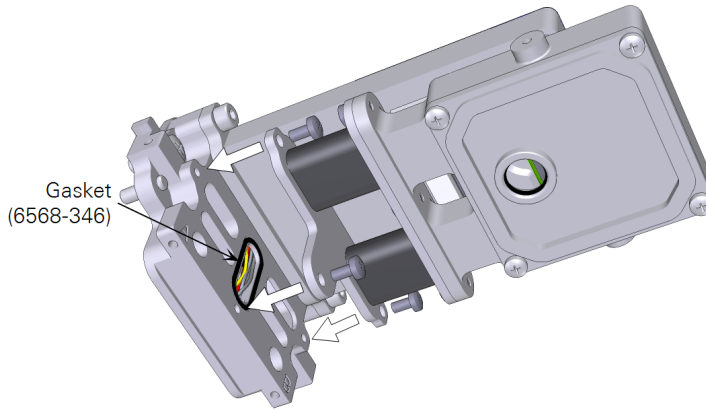


- 5** Install the new ducts on the lower chamber first (the part removed in the previous step). Work the duct through the opening, being careful to ensure that the bead seats in the groove. Use caution and be sure that the bead does not get pinched.



Tighten the screws snugly (1.1 N•m; 9.4 inch lbs if you have a torque screwdriver).

- 6** Inspect the gasket that sits behind the other plate. Replace it if it shows signs of damage (cracking, pinched spots, or breaks). There is an extra in the spares kit (part number 6568-346).
- 7** Install the lower chamber. Work the ducts through the opening in the other plate and install the three screws that hold the chamber. Tighten the screws snugly (1.1 N•m; 9.4 inch lbs if you have a torque screwdriver). Once again, use caution to be sure the bead does not get pinched.

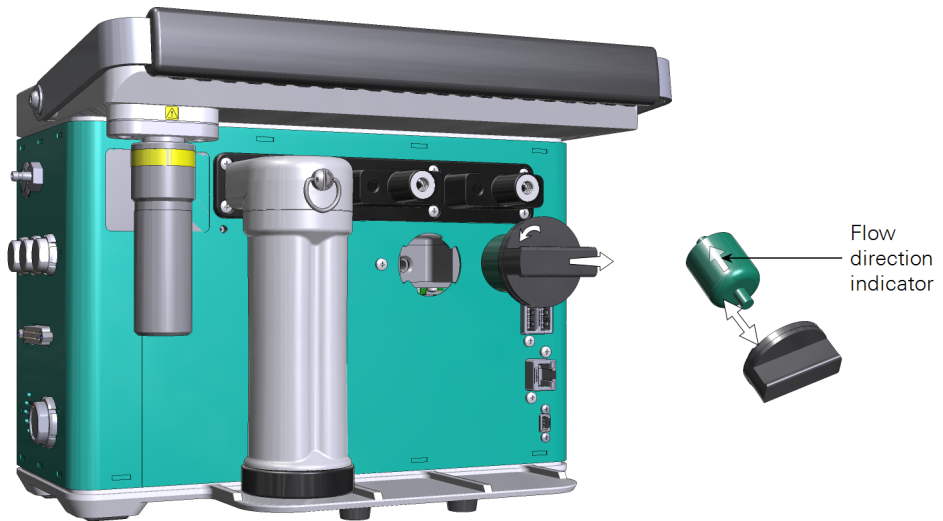


- 8** Install the hinge arms that were removed in Step 2.
Tighten the screws snugly (1.1 N•m; 9.4 inch ounces if you have a torque screwdriver).
- 9** Install the chamber on the head.
- 10** Perform the sensor tests.
They are under **Start Up > System Tests**, select **Sensor Check**.

Intake air filter

There is one intake air filter in the console. It should be replaced about once per year if the instrument has been used regularly. If the instrument is unable to achieve the set flow rate, the filter might be clogged. To replace the intake air filter:

- 1** Remove both the humidifier and soda lime columns.
- 2** Remove the filter.
Turn the intake air filter cap $\frac{1}{4}$ turn counter clockwise and pull it out. The filter and cap will come out together.



- 3** Separate the filter from the cap.
The filter is disposable. Throw it away if it is clogged.
- 4** Inspect the O-rings in the cap and the console for cracks or damage.
Replace them if you see damage. They are captive and should not come out unless you pry them out with a toothpick. To remove the console O-ring set the console on its front. Dislodge the O-ring with a toothpick. Be extremely careful so that you do not scratch the manifold or drop debris into it. Press the new O-ring into the manifold.
- 5** Install a new filter (part number 300-14311).
- 6** Do the leak test.

Chemical columns

Complete instructions for filling the chemical columns are given in *Filling the chemical columns* on page 2-5.

Column air filters

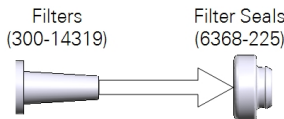
Each chemical column has two filters — one on the air-in side, and another on the air-out side. The filters may become dirty or clogged, in which case you will need to clean or replace them. Extra filters are in the spares kit. If you suspect that a filter is clogged, inspect the filter by comparing it with a new filter from the accessories kit. If you can see fouling or debris in the filter, it may need to be replaced. To replace a filter:

- 1 Remove the old filter and seal.

It will be held firmly, but it will come out if you work at it. Pry it out with your fingers or a needle-nose pliers. A pliers will probably crush the old filter, so only do this if you intend to dispose of the filter. The filter and seal come out as a unit.

- 2 Assemble a new filter.

Locate a filter (part number 300-14319) and a rubber seal (part number 6368-225) in the accessories kit. Insert the filter into the rubber seal. One side of the seal fits around the filter.



- 3 Install the filter.

Press the combined filter and seal into the housing in the column. Reinstall the column on the console. Perform the leak test to verify that everything is working as expected.



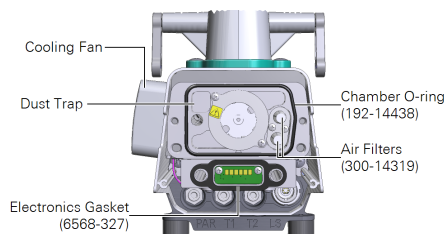
Gas analyzer maintenance

The infrared gas analyzers will need periodic care to prevent calibration drift and ensure reliable performance. The frequency of maintenance will depend somewhat upon the use conditions and the carefulness of the operator.

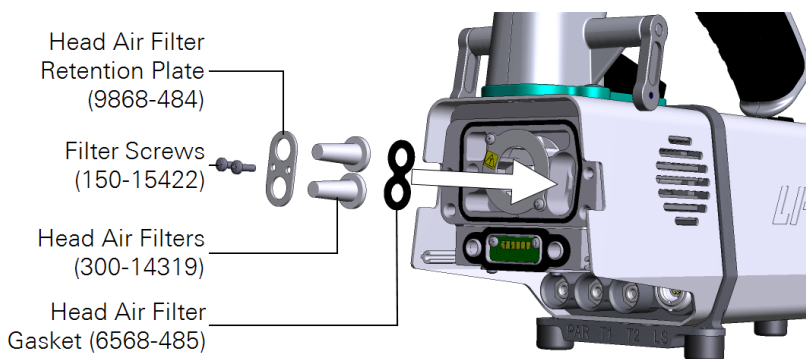
Replacing the head filters

Caution: Power off the instrument before following these procedures.

There are two small air filters in the head between the leaf chamber and the gas analyzer. These filters help prevent debris from entering the gas analyzer. You may need to clean or replace them periodically. There also is a dust trap that will collect dust.



- 1 Remove the leaf chamber as described in *Removing a chamber* on page 5-2.
- 2 The filters are held in place with two screws (M2; 6 mm long), a retention plate (part number 9868-484), and gasket (part number 6568-485).
- 3 If the filters are clogged, replace them with new filters from the spares kit (part number 300-14319).
- 4 Clean the internal surfaces around the fan, filters, ducts, and dust trap with a small swab from the spares kit.
- 5 Install a chamber and perform the leak test.



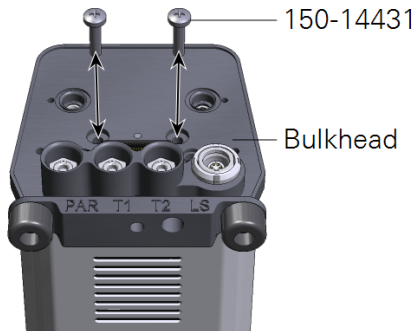
Cleaning the optical bench

The LI-6800 gas analyzers are open to the leaf chamber, but there are two filters that will prevent most debris from entering the gas analyzers. If the optical bench gets any debris in it you will need to access the gas analyzers and clean the optics. Do not open the optical bench unless you are certain that the optics are contaminated.

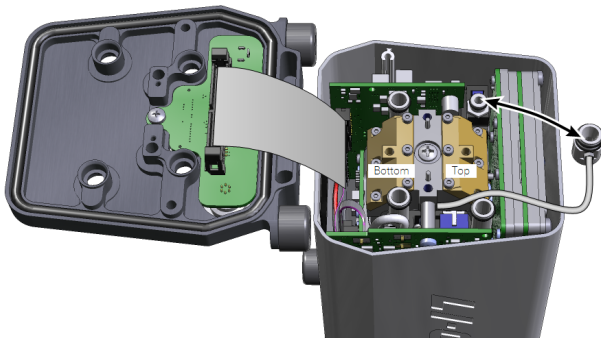
Caution: Power off the instrument before following these procedures.

Open the optical bench

- 1** Remove the leaf chamber as described in *Removing a chamber* on page 5-2.
- 2** Remove the heat exchanger assembly.
The heat exchanger is secured with two screws—one above the PAR connector, and the other above the LS connector. The screws are captive. They only need to be loosened until they move freely.
Set aside the heat exchanger and handle assembly.
- 3** Remove the bulkhead cover.
The bulkhead cover is secured with two screws (part number 150-14431). Remove them.



- 4** Gently flip over the bulkhead, exposing the gas analyzers.

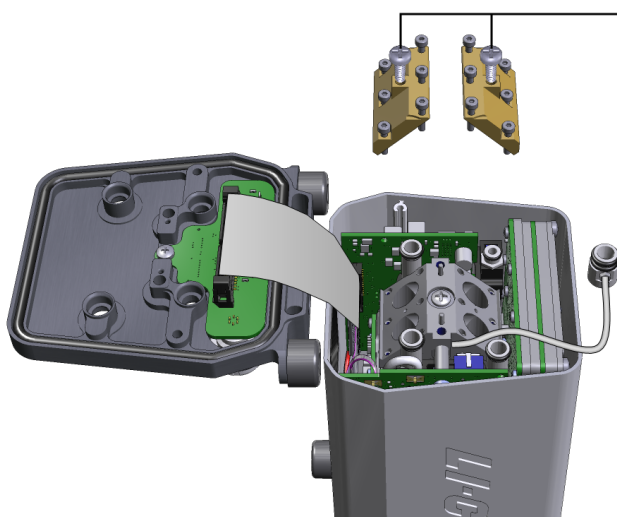


You can leave the ribbon cable in place. Lift off the pressure tube and move it to the side.

- 5** Mark the mirrors "Bottom" and "Top" with a permanent marker to help you remember which is which.

If you get the mirrors mixed up, you'll probably have a calibration shift. Remember the two screws that held the bulkhead? Insert them (or part number 150-14431 from the spares kit) into the threaded holes in the mirrors. Hold onto these screws when lifting out the mirror.

- 6** Completely loosen each of the six screws and lift out the mirror.



Insert these screws to make it easier to lift out the mirrors.

7 Inspect the IRGA volumes and sapphire windows.

If there is visible contamination, wipe them with the large swabs (part number 610-05315) from the spares kit to remove contamination. Blow out the IRGAs with canned or compressed air.

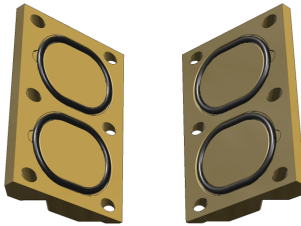
8 Wipe the mirrors with a soft lint-free cloth.

Use the soft cotton glove that is included in the spares kit.

Caution: Do not use swabs on the mirrors. Swabs can damage the delicate surface of the mirrors.

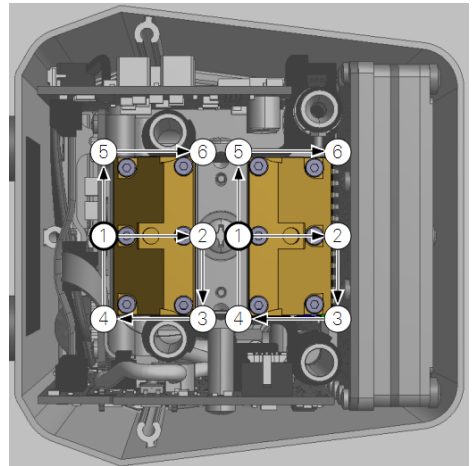
9 Inspect the mirror O-rings and replace them if any O-rings are cracked.

To remove an O-ring, dislodge it with a toothpick. Extra O-rings are in the spares kit. Press the new O-ring into the mirror.



10 Install the mirrors.

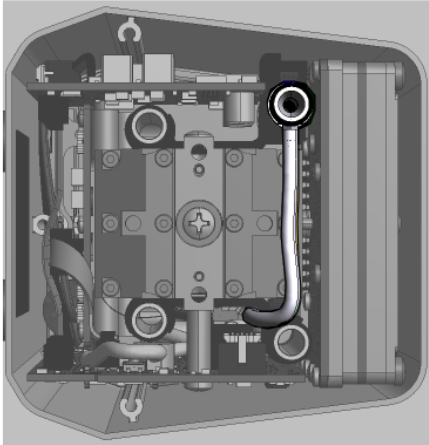
Tighten the mirror screws snugly in a spiral sequence starting with a middle screw. If you have a torque screwdriver, tighten the screws to 45 inch ounces (0.32 N•m) force. Repeat the tightening sequence.



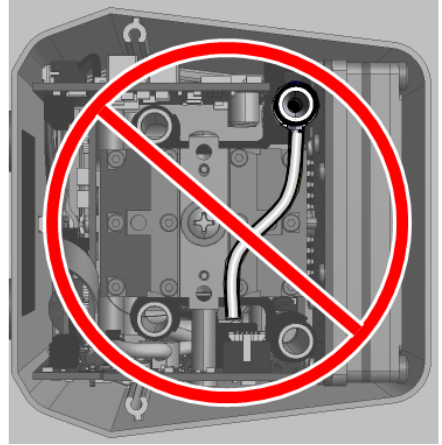
11 Install the pressure tube.

Gently press it into place. Route the tube around the mirror so it doesn't become pinched when you install the bulkhead cover.

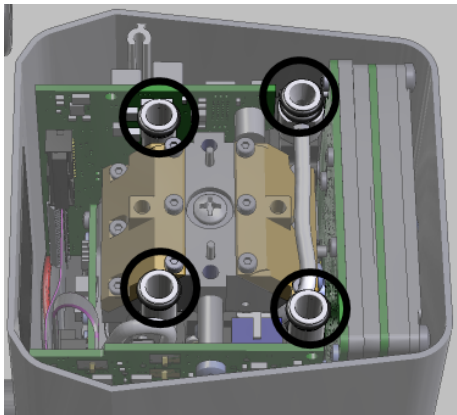
Correct routing around the mirror.



Incorrect routing will pinch the tube.

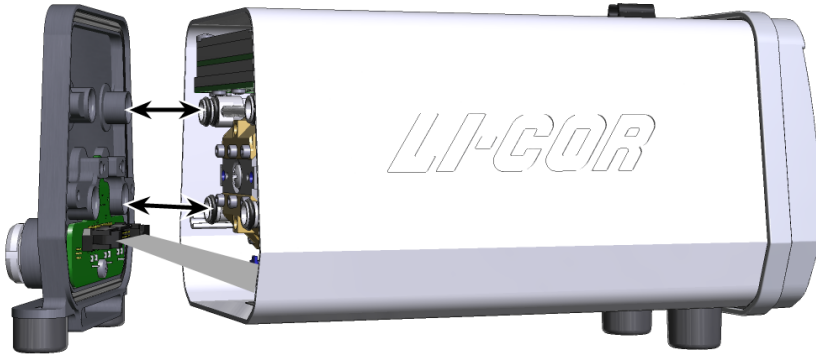
**12** Check the condition of the four bulkhead O-rings.

If they are cracked or brittle, replace them with O-rings from the spares kit (part number 192-14314).



13 Install the bulkhead.

When positioning the bulkhead, be sure that the four tubes and their O-rings are aligned correctly with the openings in the bulkhead.



Tighten the screws snugly. If you have a torque screwdriver, tighten them to 22 inch lbs (2.5 N•m).

14 Check the condition of the O-rings on the heat exchanger assembly.

If they are cracked or brittle, replace them with O-rings from the spares kit (part number 192-14314).

15 Install the heat exchanger assembly.

Tighten the screws snugly. If you have a torque screwdriver, tighten them to 22 inch lbs (2.5 N•m).

16 Install the chamber.

See *Removing a chamber* on page 5-2 for details.

17 Perform the sensor tests.

Under **Start Up > System Tests**, select **Sensor Check**. The most important one is the leak test.

18 Set the Zero and Span calibrations.

See *User calibration* on page 8-2.

Chemical bottles

Caution: Power off the instrument before following these procedures. Wear the anti-static wrist strap during this procedure. This will prevent static discharge.

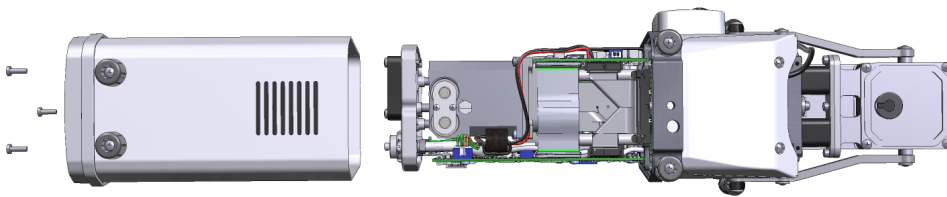
There are two small plastic bottles in the analyzer housing that keep the detectors free of CO₂ and water vapor. The bottles should last two or more years. Be sure that you have carefully considered every possible cause of an issue before concluding that the chemicals need to be replaced.

To change the sensor head soda lime and desiccant bottles:

- 1 Remove the shell from the head.

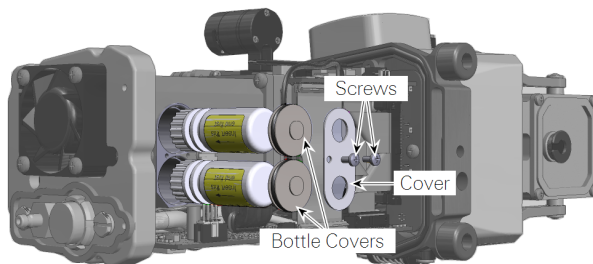
The shell is secured with three Phillips screws. Remove the screws with the #1 screwdriver. Slide the shell off of the rear of the head as shown.

Caution: Handle the head very carefully when the shell is off. Do not touch or bend the circuit boards.



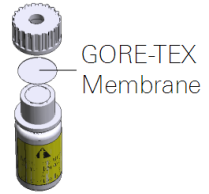
- 2 Remove the bottle cover screws, cover, and bottle covers.

Once the cover is removed, the bottles will slide out. You may need to grip them with your fingernails.



3 Prepare the new bottles.

If you have replacement bottles from LI-COR you should take the caps off of the removed bottles (the caps have a hole in the top) and put them onto the new bottles. So, the bottles that go into the head will have a hole in the cap. The GORE-TEX membrane should stay in the cap.



4 Reassemble the head.

The bottles go into the head with the caps toward the top of the sensor head. Reinstall the bottle covers, cover, and then tighten the screws snugly. Gently install the shell, and reinstall the three screws removed in step 1.

When you install the shell, be sure that the *full perimeter sits against the recessed gasket* on the front of the bulkhead, and that it is not touching the bulkhead directly. Tighten the three screws to secure the shell.

Note: After replacing the bottles, let the instrument sit for a day before checking the zeros. The bottles will need this time to scrub CO₂ and water vapor from the analyzer. You can speed this up by letting the instrument sit with the power on. If you do not wait, the zero will be incorrect.

Light source maintenance

There really is no maintenance required for the light source. The foam gasket on the light source protects the Propafilm on the chamber and the transparent plastic on the light source, and it blocks ambient light from the chamber. Inspect the foam periodically.

Clean the light source windows with a soft, lint-free cloth and rubbing alcohol if needed. Do not use other solvents on the transparent plastic.

Updating the software

Software updates, which include bug fixes and new features, will be released periodically. Software update may include packages for the head, console, Multiphase Flash Fluorometer, and the soil chamber. Update packages will be available from licor.com/env/support/LI-6800/software.html.

Caution: Do not attempt the update with a computer that is using a wireless connection. Do the update from a computer that is directly connected with an Ethernet cable to the instrument or the network. The update requires a computer running Windows 7 or newer or macOS.

After you've downloaded the software package to your computer:

- 1 Install the updater to your computer.

It is called li6800-win-updater-1.x.x.exe (Windows) or li6800-mac-updater-1.x.x.app.zip (macOS).

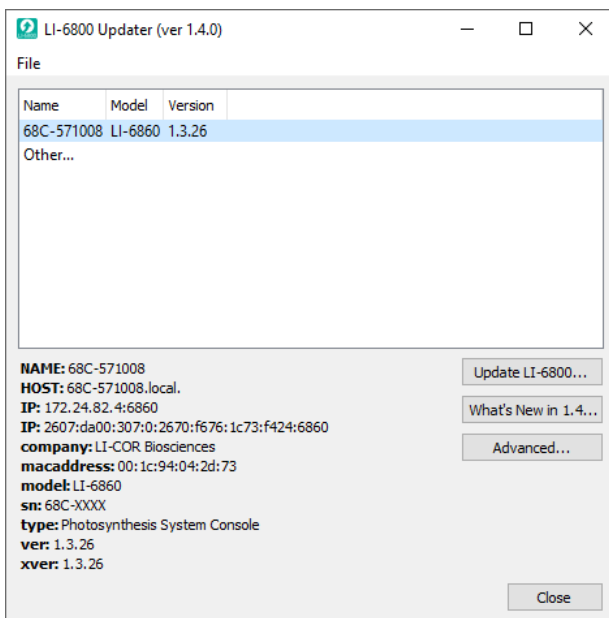
- 2 Connect the LI-6800 to your local network or your computer using the RJ-45 network cable.
- 3 Power on the LI-6800.

Be sure that the instrument has fully charged batteries or is powered from the AC power adapter so that the instrument will not power off while the update is underway.

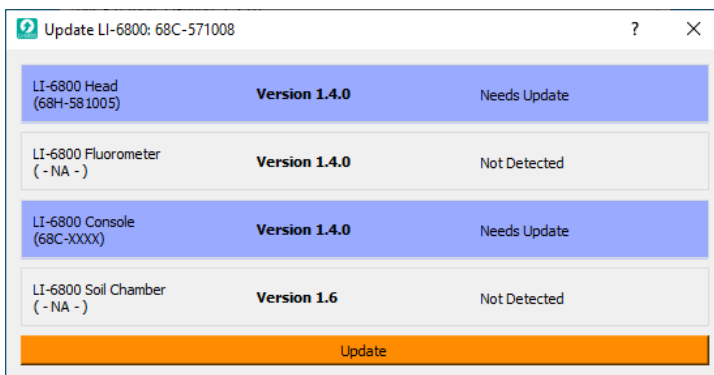
- 4 Launch the updater application.

You'll see a list of all LI-6800 consoles on your network.

- 5 Select the console (68C-xxxxx; LI-6860) and click **Update LI-6800**.



The updater will display the instrument and attached components that will be updated.



6 Click **Update** to proceed.

Be sure that your computer doesn't go to sleep while the update is underway. The updater will install the software on each component and notify you when it is done.

Note: You do not need to fully attach the soil chamber to update its firmware. Instead, you just need to connect the data cable between the chamber and the LS connector on the head.

Appendix A.

Color spec

Color Spec is a string that specifies what mix of colors to use in the light source. The string is a sequence of letter(s) (r - red, g - green, b - blue, w - white or f - farred) followed by what percentage you want that color to be. For example, "r90b10" calls for 90% red, and 10% blue. There are many shortcuts: "r90" means 90% red, and any other colors available will evenly make up the remaining 10%.

- Recognized colors are r, g, b, w, and f (f is farred, in the fluorometer)
- 6800-01/A (fluorometer) has r, b, and f, but f is never enabled unless explicitly asked for.
- 6800-02 (small light source) has r and b.
- 6800-03 (large 4 color light source) has r, g, b, and w.
- If the sum of requested percentages is <100, the difference is divided equally between any remaining unspecified colors. If there are no unspecified remaining colors, the specified colors are scaled to sum to 100.
- If the sum of requested percentages is >100, they are scaled to sum to 100.
- There is no order dependency. That is, "r30w50" is the same as "w50r30".

Table A-1. Examples of Color Spec settings and their effect on the three light sources.

Color Spec	6800-01/A			6800-02		6800-03			
	% red	% blue	% farred	% red	% blue	% red	% green	% blue	% white
r90	90	10	0	90	10	90	3.3	3.3	3.3
r90b10	90	10	0	90	10	90	0	10	0
r90b9f1	90	9	1	90.9	9.1	90	0.5	9	0.5
w100	50	50	0	50	50	0	0	0	100
w25	50	50	0	50	50	25	25	25	25
r50g30b20	71.4	28.6	0	71.4	28.6	50	30	20	0

"w25" for the 6800-01, -02: There is no white to set to 25%, so red and blue are each 50%.

"r50g30b20" for the 6800-01, -02: red is 50, blue is 20, but there is no green. That leaves 30% unaccounted for, so the 50 and 20 are scaled up to 71.4 and 28.6.

Optional Limits: Upper case color letters (R,G,B,W,F) can be added to a color spec to limit that particular color(s) to some level of intensity in ($\text{mol m}^{-2} \text{s}^{-1}$). For example, the recommended color spec for the fluorometer is "r90B40" which will keep set blue LEDs to 10% of the total, or 40 $\text{mol m}^{-2} \text{s}^{-1}$, whichever is lower. The reason for this is to have just enough blue for stomatal control and keep the actinic and the fluorescence measuring beam as spectrally close as possible.

Important side effect: Note that the 6800-02 and 6800-03 (but NOT the 6800-01) will dim the LEDs any time a color change is specified while the light is on. Thus any specification with a limit specifier (upper case color letter) has the potential of causing a color change (and momentary light intensity dip) any time a new set point target is specified. So, for example, ramping a lamp with Auto Control with the spec "r90" will cause smooth changes in light, but with "r90B40", the lamp will momentarily dim at each new setpoint above 40, since the blue has to be reduced by varying amounts to keep it at 40. Again, this is not an issue with the fluorometer, but it is with the other light sources.

Appendix B.

Control picker

When choosing controls, either in Auto Controls or Auto Programs, you select a **Group**, and from that group, a variable **Name**.

The groups and names are available for Auto Controls and Auto Programs. Several Auto Programs are capable of setting constants and settings, and those groupings are shown in *Table B-2* on the next page.

Table B-1. Environmental controls that can be selected in the AutoEnv interface.

Group	Name	Description
Env Aux Controls	AuxPwr	Auxiliary power in head (0 to 19 V)
	Dac1	DAC 1 (pin 2) -5 to +5 V
	Dac2	DAC 2 (pin 3) -5 to +5 V
	Dac3	DAC 3 (pin 4) -5 to +5 V
	Dac4	DAC 4 (pin 5) -5 to +5 V
Env CO ₂ Controls	CO ₂ _%	Injector control 0 to 100%
	CO ₂ _r	Reference CO ₂
	CO ₂ _s	Sample CO ₂
Env Fan Controls	Fan_%	Fan power (0 to 100%)
	Fan_blc	Boundary layer conductance target (mol m ⁻² s ⁻¹)
	Fan_rpm	Fan speed (rpm)
Env Flow Controls	Flow_%	Percent flow to chamber (0 to 100%)
	Flow	Flow to chamber (μmol s ⁻¹)

Table B-1. Environmental controls that can be selected in the AutoEnv interface. (...continued)

Group	Name	Description
Env H ₂ O Controls	Desiccant_%	Desiccant (only) control (0 to 100%)
	H2O_%	Full desiccant to full humidifier (-100 to 100%)
	H2O_r	Reference H ₂ O (mmol mol ⁻¹)
	H2O_s	Sample H ₂ O (mmol mol ⁻¹)
	Humidifier_%	Humidifier (only) control (0 to 100%)
	RH_air	Chamber RH (0 to 10%)
	SD_air	Saturation Deficit (kPa)
	VPD_leaf	VPD of leaf (kPa)
Env Light Controls	Q_All	All attached light sensors
	Q_Console	Console light source only
	Q_Flr	Fluorometer only
	Q_Head	Head light source only
	Qin	All light sources contributing to Leaf Q
Env Pressure Controls	Pressure	Chamber over pressure (kPa)
	Pressure_%	Overpressure valve (0 to 100%)
Env Temperature Controls	Tair	Chamber air temperature
	Tleaf	Leaf temperature
	Txchg	Heat exchanger temperature

Table B-2. Environmental constants that can be changed via the MCLoop interface.

Group	Name	Description
Env Color Controls	Color_All	All attached sources
	Color_Con	Console light source
	Color_Flr	Fluorometer
	Color_Head	Head light source
	Color_Qin	All sources contributing to Leaf Q
Flr Constants	FLR:Adark	Dark respiration rate
	FLR:Fm	Maximal fluorescence
	FLR:Fo	Minimal fluorescence
	FLR:PS2/1	Ratio PS2 to PS1

Table B-2. Environmental constants that can be changed via the MCLoop interface. (...continued)

Group	Name	Description
Flr Dark Settings	DARK:After	Far red off _ secs after actinic off
	DARK:Before	Far red on _ secs after actinic off
	DARK:Duration	Actinic off duration
	DARK:FarRed target	Far red target
Flr Induct Settings	IND:Duration	Flash duration
	IND:Red target	Red flash target
Flr MPF Settings	MPF:Phase 1	Phase 1 duration
	MPF:Phase 2	Phase 2 duration
	MPF:Phase 3	Phase 3 duration
	MPF:Ramp	Percent ramp
	MPF:Red target	Red flash target
Flr RF Settings	RF:Duration	Flash duration
	RF:Red target"	Ref flash target
Gas Exchange Constants	Const:Custom	Custom BLC
	Const:Geometry	0=broadleaf, 1=needle, 2=custom
	Const:K	Stomatal ratio
	Const:S	Leaf area (or Mass)
Leaf Temp Constants	LTConst:deltaTw	Wall - Air temp difference
	LTConst:fT1	T1 weighting
	LTConst:fT2	T2 weighting
	LTConst:fTeb	EB weighting
System Constants	SysConst:AvgTime	Average time
	SysConst:Oxygen	Oxygen percentage
User Defined Constants	??	whatever is defined

Appendix C.

Leaf-level photosynthesis and its measurement

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Leaf-level interactions with the atmosphere relevant to photosynthesis...C-4

Leaf-level photosynthetic coordination...C-5

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Leaf-level photosynthesis and its measurement

The LI-6800, when equipped with a chlorophyll fluorometer, is designed to measure two aspects of photosynthesis: 1) the light, or energy producing, reactions; and 2) the CO₂ assimilation, or energy consuming, reactions. Two different techniques are used to measure these very different reactions that occur at different points in the overall process of photosynthesis. Nonetheless, all of these reactions are biochemically connected. Consequently, unique and important information can be gleaned by assessing the combined information from the two aspects of photosynthesis. In this section, we describe how the LI-6800 probes these processes.

Leaf cross section

Leaves are bounded by epidermal cell layers that help to separate the leaf intercellular air space from the atmosphere. Between the epidermal layers are various other cell types, e.g., palisade, mesophyll, bundle sheath, etc., depending upon the many different varieties of leaf morphologies found in nature. As a starting point, we'll refer to a simplified schematic of a leaf cross-section throughout this preface (*Figure 1* on the facing page).

A typical plant cell has a ligneous wall and a plasma membrane that encapsulate the aqueous, cytosolic compartment. While there are many important cellular organelles, the cell depicted here is chloroplast-centric: the only organelle shown within the cell is a double membrane chloroplast, which encloses the aqueous stromal compartment. Localized within the stroma is a complex network of thylakoid membranes that also have been significantly simplified in the schematic. Integrated within the thylakoid membrane are all the proteins, pigments and redox cofactors that function to transduce light energy into the transfer of electrons and protons, the combined effect of which results in the storage of light energy in chemical forms of energy (i.e., NADPH and ATP) (1). These chemical intermediates can be used to power, among other processes, the enzymatic reduction of CO₂ to the level of sugar phosphates (2).

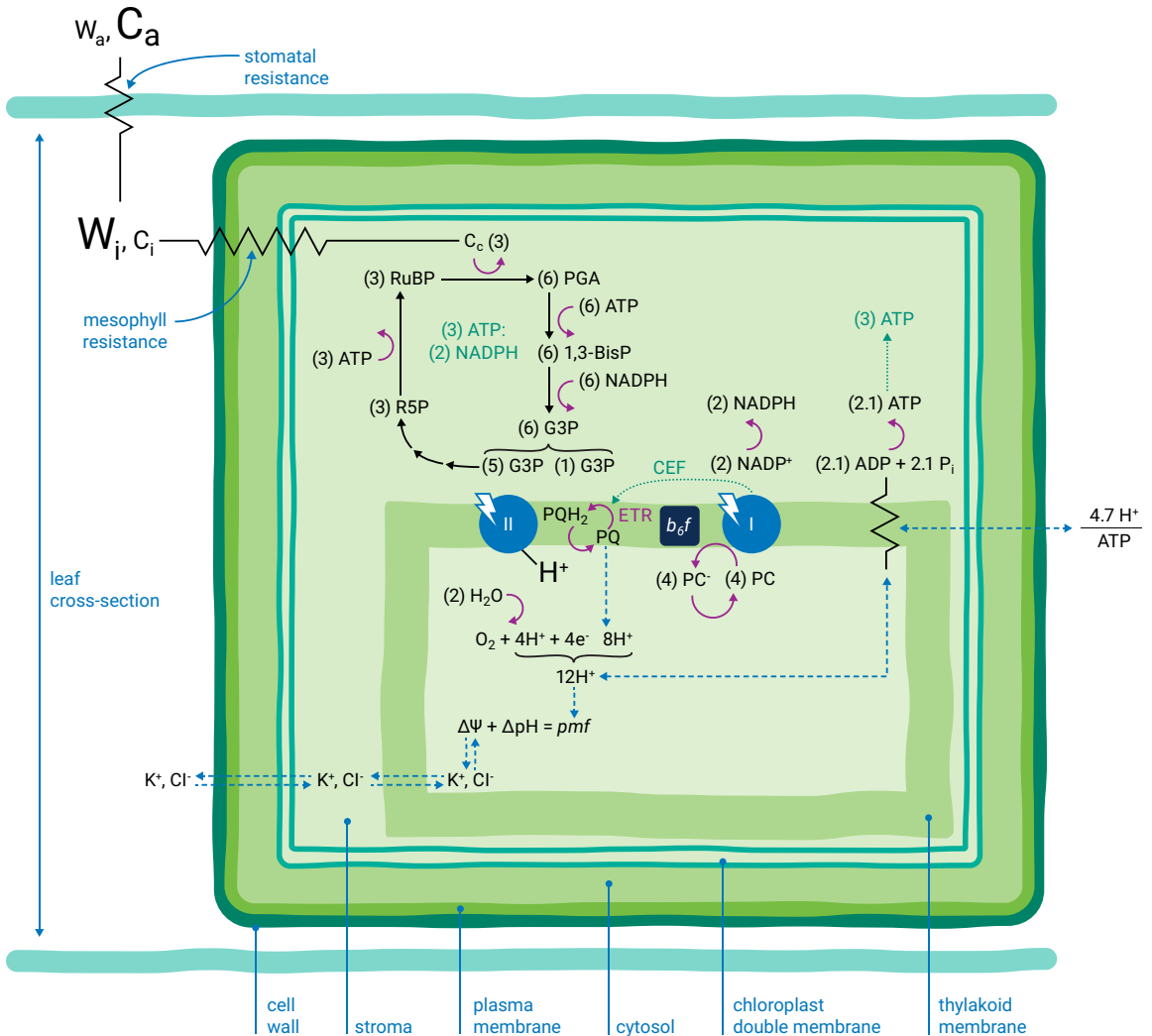


Figure 1. Cross section of a simplified leaf composed of a single cell with a single organelle: the chloroplast. This leaf schematic has many of the biophysical and biochemical photosynthetic processes represented. Resistance symbols ($\sim\sim$) indicate resistance to diffusion. The relative concentrations of gases are indicated by the size of characters.

Leaf-level interactions with the atmosphere relevant to photosynthesis

Atmospheric gases (CO_2 , O_2 , H_2O and others) diffuse into and out of the intercellular leaf air space through stomatal apertures (Figure 2). Stomatal apertures cause resistance to gas diffusion (3). The intercellular air space is assumed to be saturated for water vapor at leaf temperature (4). The relative water vapor concentration within the leaf (W_i) is typically greater than the water vapor concentration in the atmosphere (W_a) as a result of this stomatal resistance. Stomatal resistance also results in a lower CO_2 concentration in the leaf intercellular air spaces (C_i) than in the atmosphere (C_a).

Leaves principally absorb light within the visible region (mostly red and blue) of the electromagnetic spectrum (5). Light absorption, which powers photosynthesis (Figure 3), occurs within specialized pigment-binding protein complexes referred to as photosystems I and II (PSI and PSII) (6). These photosystems are localized within the chloroplast thylakoid membranes (7) and they function in series to facilitate linear electron transfer (*ETR*; $\mu\text{mol electrons m}^{-2} \text{s}^{-1}$). Linear *ETR* is a proton-coupled process (8, 9) that establishes a trans-thylakoid proton motive force (*pmf*) consisting of both electric field ($\Delta\Psi$) (10) and pH (ΔpH) gradients (11).

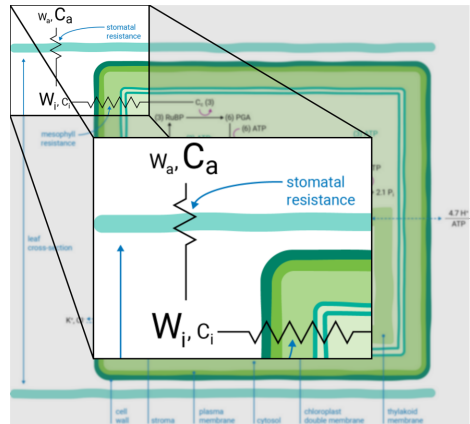


Figure 2. CO_2 and H_2O vapor move into and out of the leaf through stomatal apertures.

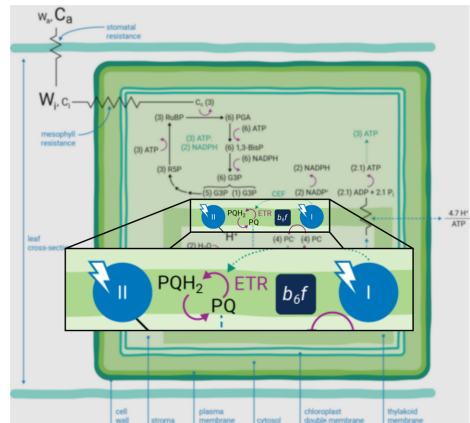


Figure 3. Photosystems I (right) and II (left), are pigment binding protein complexes where the leaf absorbs light.

Photosynthesis-driven transpiration begins with evaporation of H_2O from cell wall orifices and into leaf intercellular air spaces (12). Transpiration moves H_2O from below-ground soil sources to the atmosphere, transporting ions (e.g., K^+ and Cl^- , etc.) (13) throughout the plant. These ions serve many cellular purposes including regulation of the relative partitioning of the pmf into $\Delta\Psi$ and ΔpH components (11, 14).

Leaf-level photosynthetic coordination

Energy producing and consuming reactions

The energy producing (i.e., NADPH and ATP formation) and consuming (CO_2 assimilation, etc.) reactions of photosynthesis are tightly coordinated (15). The tight coordination is a function of the rigid stoichiometric relationships of key steps within the series of photosynthetic reactions. Assuming 100% quantum conversion at PSI and PSII (16), absorption of four photons at each photosystem results in the net oxidation of two H_2O molecules, thereby releasing four protons (H^+) into the thylakoid lumen and the generation one molecule of O_2 (Figure 1 on page C-3). The electrons freed during these thermodynamically favorable reactions can be subsequently used to form two molecules of NADPH. This transfer of electrons is a somewhat more explicit description of linear *ETR*.

If the Q-cycle at the cytochrome *b₆f* complex operates constitutively (9, 17 - 19), oxidation of two H_2O molecules would result in the net accumulation of twelve protons in the thylakoid lumen, thereby contributing to the pmf (8). The resulting amount of ATP generated as these twelve protons move down their electrochemical gradient through the chloroplast ATP synthase is determined by the enzyme's H^+/ATP ratio (20). The ATP synthase imposes a resistance to proton efflux from the lumen to the stroma of the chloroplast (Figure 4) (1). The H^+/ATP ratio of the ATP synthase is contested (21), but if the ratio is four, movement of twelve

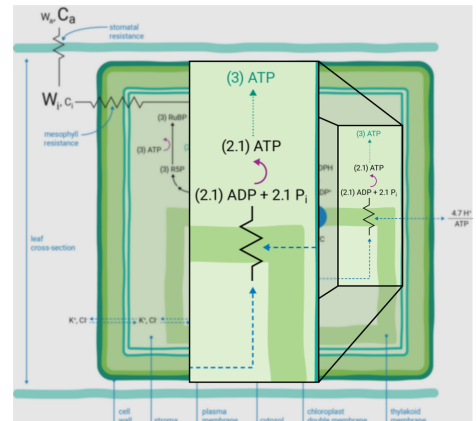


Figure 4. Protons encounter resistance as they move from the lumen to the stroma of the chloroplast.

protons through the ATP synthase would result in the formation of three ATP molecules. Thus, oxidation of two H_2O molecules would result in two NADPH and three ATP molecules, precisely the chemical stoichiometry needed to assimilate a single CO_2 molecule (2).

In contrast, given the putative c-subunit composition of the chloroplast ATP synthase (22) (Figure 5 above), an H^+/ATP ratio of 4.7 has been suggested. The net result is a deficit in the amount of ATP produced by linear *ETR* alone in comparison to that needed to balance the ATP:NADPH ratio for CO_2 assimilation alone. Cyclic electron transfer (*ETR*) around PSI has been suggested as a mechanism for augmenting the *pmf*, thereby accounting for the deficit of ATP produced by linear *ETR* (23, 24). The rate of cyclic *ETR* around PSI needed to accommodate for the short fall in ATP formation has been suggested to be about 12% of linear *ETR* (21).

In either case, the relationship between linear *ETR* and the rate of CO_2 assimilation is predicted to have a ratio of slightly more than four under conditions when CO_2 assimilation is the predominant sink for ATP and NADPH (25). Such conditions occur when photorespiration is inhibited by performing experiments on C_4 plants or, under low O_2 , on C_3 plants.

Photosynthetic light reactions must be highly adaptive to a dynamic environment to coordinate the energetic demands for ATP and NADPH while simultaneously regulating light capture (26). Processes other than CO_2 assimilation (such as photorespiration, nitrogen and sulfur metabolism) have different ATP and NADPH requirements. Non-photochemical quenching (*NPQ*) of excess absorbed light energy serves as a photo-protective mechanism (27, 28, 29), preventing formation of highly reactive forms of oxygen: singlet oxygen ($^1\text{O}_2$) and the superoxide radical anion (O_2^-).

Energy-dependent quenching (q_E) (30), the predominant component of *NPQ*, is a dual function of the ΔpH component of the *pmf* (27). The ΔpH both 1) activates a

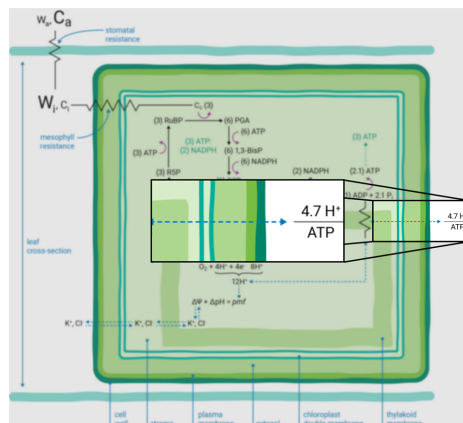


Figure 5. The ratio of H^+ to ATP is typically 4.7.

thylakoid lumen-localized enzyme violaxanthin de-epoxidase, which converts violaxanthin (*V*) to zeaxanthin (*Z*) in the PSII antenna (31); and 2) regulates protonation of thylakoid lumen-exposed, amino acid residues associated with PSII proteins (32, 33).

Dynamic coordination of the light reactions is required to meet the energetic and photoprotective demands during fluctuating biochemistries that occur during environmental stress, such as drought, for example (1, 26). The coordination is mediated by changes in cyclic *ETR* around PSI and changes in the proton conductance of the chloroplast ATP synthase (g_{H^+}) (14, 34). The former is thought to be predominantly involved in regulating the output ratio of ATP:NADPH, while the latter is thought to be primarily responsible for modulating q_E sensitivity (1, 26).

Energy production and photoprotection

Under fluctuating biochemical demands, q_E sensitivity refers to a relative increase in the amount of q_E at a given flux of linear *ETR* (14, 35). Linear *ETR* and q_E increase in hyperbolic and sigmoid manners, respectively, with increasing light intensity under ambient CO_2 and 21% O_2 (Figure 6 above, black squares), as well as under 50 ppm CO_2 and 21% O_2 (Figure 6 above, red circles). The lower CO_2 concentration artificially imposes a change in biochemical demand that mimics various environmental stresses. At the highest light intensity, lowering the CO_2 concentration decreased linear *ETR* by 42% in comparison to values under ambient CO_2 , whereas q_E increased by approximately 2-fold under the same low CO_2 concentration treatment. In essence, q_E became more sensitive to linear *ETR* in response to the diminished biochemical demand. Combining analyses of electron and proton transfer reactions of photosynthesis has previously shown that changes in q_E sensitivity are predominantly attributable to proportional changes in g_{H^+} (14, 35).

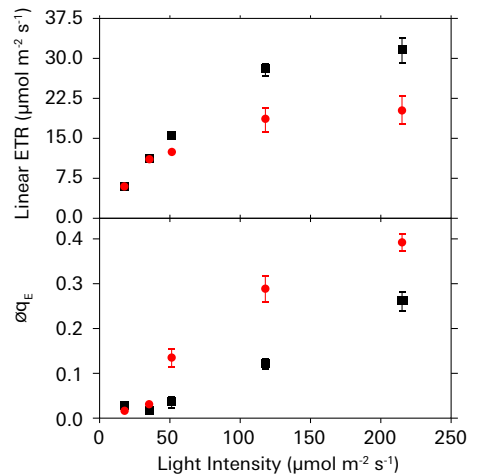


Figure 6. Linear *ETR* and q_E both increase with increasing light intensity under ambient CO_2 and 21% oxygen (black squares), and under 50 ppm CO_2 and 21% oxygen (red circles).

Coordination of assimilation and transpiration

Optimal assimilation of CO_2 from the atmosphere by leaf-level photosynthesis is directly coupled with H_2O loss. Energy use from light capture, carbon assimilation, and H_2O loss is coordinated through regulation of stomatal conductance (g_s) (3) and mesophyll conductance (g_m) (36 - 40). Research into understanding the interdependencies of these regulatory mechanisms has led to interest in selecting for plants to increase the $g_m:g_s$ ratio, the result of which could lead to more water use efficient plants (41).

Assessing the gas exchange parameters

The gas exchange model described below assumes that the system has no leaks and that air is perfectly mixed in the chamber. In practice, this does not always happen. We present a detailed description of the computations when these assumptions are not met in *Mass balance derivation using a two-compartment model* on page C-38.

An open gas exchange system

The LI-6800 is an open gas exchange system (4) that measures many parameters needed to estimate photosynthetic gas exchange. The gas exchange system measures the flow rate ($\mu\text{mol air s}^{-1}$) entering the leaf chamber (μ_0) and the CO_2 and H_2O concentrations entering (c_0 and w_0 , respectively) and exiting (c_a and w_a , respectively) the leaf chamber (*Figure 7* below).

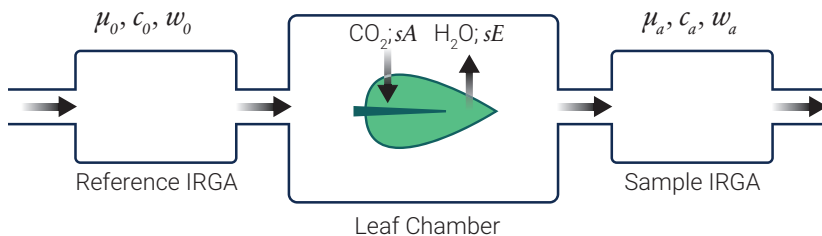


Figure 7. Schematic of an open gas exchange system for leaf-level photosynthesis measurements.

The LI-6800 computes the mass flow per time (i.e., $\mu\text{mol CO}_2 \text{ s}^{-1}$ and $\text{mmol H}_2\text{O s}^{-1}$) of these gases into and out of the chamber. The differences between the CO_2 and H_2O concentrations into and out of the leaf chamber are due to leaf-level CO_2 assimilation and transpiration on a leaf area (s) basis ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ and $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$). During photosynthetic carbon assimilation (A), the leaf takes up CO_2

from the air entering the chamber while simultaneously releasing H₂O via transpiration (E) into the chamber air. While the flow rate of air coming out of the chamber (μ_a) is not directly measured, it is assumed to be:

$$\mu_a = \mu_0 + sE \quad \text{C-1}$$

where s represents leaf area. Transpiration from the leaf is adding water vapor to the chamber, which changes the air density and therefore is added to the total flow into the leaf chamber.

The open-system measurements can subsequently be used to estimate important gas exchange parameters (4, 42). Transpiration, on a leaf area (s) basis, is calculated as:

$$sE = \mu_a w_a - \mu_0 w_0 \quad \text{C-2}$$

We can rearrange equations C-1 and C-2 to give:

$$E = \frac{\mu_0(w_a - w_0)}{s(1 - w_a)} \quad \text{C-3}$$

Similarly, we compute net CO₂ assimilation (A_{Net}) (42, 43) as:

$$sA_{Net} = \mu_0 c_0 - \mu_a c_a \quad \text{C-4}$$

Using equation C-1, equation C-4 rearranges to:

$$A_{Net} = \frac{\mu_0 \left[c_0 - c_a \left(\frac{1 - w_0}{1 - w_a} \right) \right]}{s} \quad \text{C-5}$$

The cascade of derived parameters

In accordance with reasonable assumptions (4), we use E to derive an expression that represents the total conductance of a leaf to water vapor flux (g_{Tw}):

$$g_{Tw} = \frac{E \left(1 - \frac{w_i + w_a}{2} \right)}{w_i - w_a} \quad \text{C-6}$$

In general, transpiration comes from stomata located on both sides of the leaf. On each side, water vapor diffuses through the stomata and then through the boundary layer to the mixed atmosphere. For each side of the leaf, total resistance to water vapor diffusion (r_w) is the sum of stomatal resistance (r_{sw}) plus boundary layer resistance (r_{bw}). So,

$$r_w^l = r_{sw}^l + r_{bw}^l \quad \text{C-7}$$

and

$$r_w^u = r_{sw}^u + r_{bw}^u \quad \text{C-8}$$

for lower (l) and upper (u) leaf surfaces, respectively. We assume boundary layer resistance is the same on both sides of the leaf, so $r_{bw}^l = r_{bw}^u = r_{bw}^u$. We define a stomatal ratio:

$$K = \frac{r_{sw}^u}{r_{sw}^l} \quad \text{C-9}$$

Total stomatal resistance is a parallel combination of stomatal resistances from the upper and lower surfaces, so

$$\frac{1}{r_{sw}} = \frac{1}{r_{sw}^u} + \frac{1}{r_{sw}^l} \quad \text{C-10}$$

Total conductance measured by equation C-6 represents a parallel combination of series resistances from the upper leaf surface and lower leaf surface, so

$$g_{Tw} = \frac{1}{r_{Tw}} = \frac{1}{r_{sw}^u + r_{bw}^u} + \frac{1}{r_{sw}^l + r_{bw}^l}, \quad \text{C-11}$$

since $g_i = 1/r_i$. From these relationships it can be shown that

$$g_{Tw} = \frac{1}{(1+K)r_{sw} + r_{bw}} + \frac{K}{(1+K)r_{sw} + Kr_{bw}}. \quad \text{C-12}$$

Or, in terms of conductances,

$$g_{Tw} = \frac{1}{(1+K)\frac{1}{g_{sw}} + \frac{1}{g_{bw}}} + \frac{K}{(1+K)\frac{1}{g_{sw}} + \frac{K}{g_{bw}}}. \quad \text{C-13}$$

Solving for g_{sw} yields:

$$g_{sw} = \frac{2}{\left(\frac{1}{g_{Tw}} - \frac{1}{g_{bw}}\right) + \sqrt{\left(\frac{1}{g_{Tw}} - \frac{1}{g_{bw}}\right)^2 + \frac{4K}{(K+1)^2} \left(2\frac{1}{g_{Tw}} - \frac{1}{g_{bw}}\right) \frac{1}{g_{bw}}}}. \quad \text{C-14}$$

Similarly, for conductance to CO_2 ,

$$g_{Tc} = \frac{1}{(1+K)\frac{1.6}{g_{sw}} + \frac{1.37}{g_{bw}}} + \frac{K}{(1+K)\frac{1.6}{g_{sw}} + K\frac{1.37}{g_{bw}}} \quad \text{C-15}$$

where 1.6 is the ratio of the diffusivities of CO₂ to water vapor in air and 1.37 is the same ratio in the boundary layer.

Lastly, we give an expression for C_i (4):

$$C_i = \frac{(g_{TC} - \frac{E}{2})c_a - A_{Net}}{(g_{TC} + \frac{E}{2})} \quad \text{C-16}$$

It should be noted that equation C-16 is derived from the compilation of many fundamental parameters measured by the LI-6800 gas exchange system.

The Farquhar-von Caemmerer-Berry model

Development of the Farquhar-von Caemmerer-Berry (FvCB) model of photosynthesis (42) was made possible with the types of above-mentioned measured and derived parameters. The general form of the FvCB model is (43, 46):

$$A_{Net} = \min(w_c, w_j, w_p) \left(1 - \frac{\Gamma^*}{C_i}\right) - R_d \quad \text{C-17}$$

Equation C-17 expresses A_{Net} as a function of the minimum rate of photosynthesis that can be supported by: 1) RuBisCO activity (w_c); 2) energetic chemical-intermediate (i.e., ATP and NADPH) production for ribulose 1,5-bisphosphate (RuBP) regeneration (w_j); and 3) triose phosphate export and utilization (w_p). The CO₂ compensation point in the absence of day respiration (Γ^*) and day respiration (R_d) are routinely estimated from literature sources but can also be measured (47).

While expressions for all three potentially limiting processes can be more fully defined (46), we will focus on the RUBP-regeneration limited expression because it directly links measurements of linear ETR and A . The full expression for RUBP-regeneration limited photosynthesis is (43, 46):

$$A_{Net} = \frac{JC_i}{4C_i + 8\Gamma^*} \left(1 - \frac{\Gamma^*}{C_i}\right) - R_d \quad \text{C-18}$$

J is the electron transport rate needed to supply energy for carboxylation and oxygenation reactions catalyzed by RuBisCO. We'll say more about J below. The coefficients 4 and 8 can be altered depending upon the assumptions about the relative energy demands for ATP and NADPH (46).

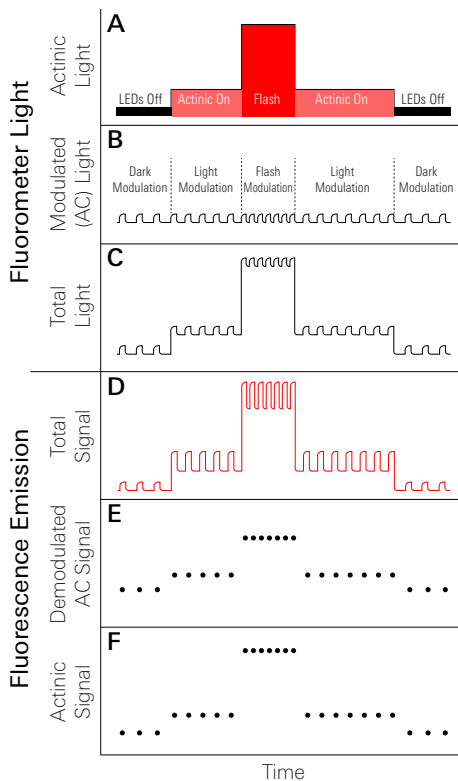
Quantifying the light reaction parameters

Light sources and functions

The LI-6800 employs two techniques to evaluate chlorophyll *a* fluorescence: 1) measurements of fluorescence emission with a pulse-amplitude modulation (PAM) fluorometer, and 2) measurements of maximum fluorescence yield upon application of a saturating flash of light (48). Each technique uses different LEDs, which have distinct experimental functions. PAM chlorophyll *a* fluorometers provide actinic light (continuous; sometimes called DC) and modulated light (sometimes called AC) shown in *Figure 8* above, **A**, **B**, and **C**.

The modulated light consists of short light pulses (*Figure 8* above, **B**) with rise and fall times within tens of nanoseconds. In the LI-6800 fluorometer, each pulse is approximately 1 microsecond and the frequencies of the pulses range from 50 Hz to 250 KHz, depending upon the type of measurements being performed. The amplitude of each pulse is fixed at a constant value of $100 \mu\text{mol m}^{-2} \text{s}^{-1}$. (There will be some instrument-to-instrument variability.) The product of the pulse width, frequency, and amplitude provide a measure of the integrated intensity (i.e., quanta in units of $\mu\text{mol m}^{-2} \text{s}^{-1}$) of the modulated light.

The combined PAM chlorophyll *a* fluorescence and saturation flash techniques are predicated on the assumption that the integrated, modulation light intensity is dim enough so as to NOT induce photosynthesis, but simply induce a low level of



*Figure 8. Depiction of actinic (A), modulated (B), and total (C) light; and the resulting total chlorophyll *a* fluorescence (D), demodulated AC fluorescence (E), and actinic (continuous) fluorescence (F) signals.*

steady-state fluorescence that probes various aspects of the prevailing state of the photosynthetic light reactions (48).

The continuous, or actinic light (*Figure 8* on the previous page, **A**) is used to 1) activate photosynthesis over a range of intensities and for extended periods of time, thereby altering the steady-state fluorescence, and 2) measure the maximum fluorescence during rapid (i.e., 1 second) saturation flashes at irradiances much greater than full solar intensity (48). The modulated LEDs of the LI-6800 fluorometer are distinct from the red and blue LEDs used for continuous light. The intensities of all three sets of LEDs can be controlled independently. While modulated and continuous light sources originate from independent LEDs, the combined light incident on a leaf is the sum of both types of light (*Figure 8* on the previous page, **C**).

Modes of fluorescence

Two modes of fluorescence are measured with the LI-6800 fluorometer (*Figure 8* on the previous page, **D**). During leaf measurements, the amplitude and pulse width of the modulation light remains constant, whereas the modulation frequencies can vary according to user-prescribed values that depend upon the experimental protocol (*Figure 8* on the previous page, **B**). With a dark-adapted leaf in the chamber, the modulated light source elicits pulses of fluorescence (*Figure 8* on the previous page, **D**), termed modulated fluorescence. Since the amplitudes of the modulated light pulses remain constant, the resulting modulated fluorescence is referred to as the quantum yield of fluorescence (Φ_F) (49, 50).

The actinic, or continuous, light is initially depicted as being off (*Figure 8* on the previous page, **A**), therefore no continuous fluorescence is stimulated and the modulated Φ_F can be seen to transiently rise above zero (*Figure 8* on the previous page, **D**) in a manner that solely reflects the relative elicitation of fluorescence from the leaf due to the modulated light. Simultaneous illumination of a leaf with both modulated and actinic light sources elicits continuous fluorescence, on top of which are pulses of modulated Φ_F . Application of a short, intense saturating flash of continuous light results in a significant increase in the amplitude of the continuous fluorescence. Once again, modulated Φ_F is elicited, albeit at a greater amplitude, on top of the flash-induced, continuous fluorescence. The continuous light, whether it be of the actinic or the saturation flash variety, alters the redox photophysics of the PSII reaction center (51) in such a manner that changes in the amplitudes of the modulated Φ_F 's reflect, rather than changes in the amplitudes of the modulated

light pulses, alteration of the rate constants of various, photosynthesis-related processes.

Demodulation of fluorescence signals

The fluorometer measures both modulating and continuous fluorescence signals (Figure 8 on page C-12, E and F). Demodulation is the electronic process of extracting and separating the modulating and continuous fluorescence components from the combined fluorescence signal (Figure 8 on page C-12, D). The continuous signal is derived from the measurements of fluorescence while the modulating LEDs are off, whereas the modulating signal is derived by subtracting each on-reading from the average of the two adjacent off-readings.

Chlorophyll photophysics

Various photophysical processes control the energy state of chlorophyll during photosynthesis. Using a simple, hypothetical model of an isolated chlorophyll molecule (Figure 9 above), excitation of a ground-state chlorophyll (*Chl*) molecule by a photon results in the very rapid formation, either directly or indirectly, of the singlet-excited state of chlorophyll ($^1Chl^*$). It is thermodynamically favorable for $^1Chl^*$ to return to *Chl* and it can do so via a number of processes.

The rate constants

When considering a hypothetical population of isolated chlorophyll molecules, the proportion of $^1Chl^*$ decaying to *Chl* is determined by the parallel, first order rate constants (k) of three processes: fluorescence (k_F), internal conversion (k_{IC}), and intersystem crossing (k_{ISC}) into the triplet state of chlorophyll ($^3Chl^*$) (49, 52).

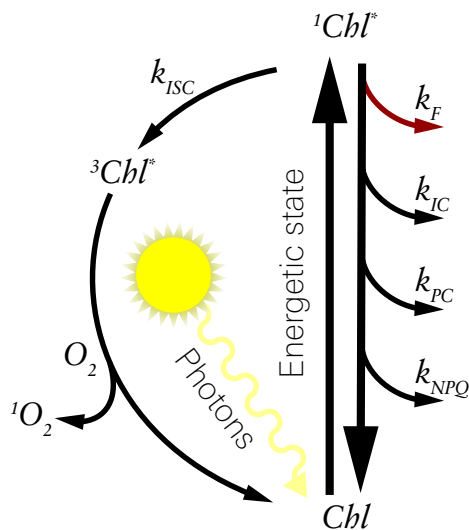


Figure 9. When a chlorophyll pigment is in its ground state (*Chl*), the absorption of a photon elevates it to a higher energy state ($^1Chl^*$). It returns to the ground state, dissipating the energy through one or more pathways.

$^3Chl^*$ can decay to the Chl through being quenched by specialized pigments (carotenoids) or by reacting with O_2 to produce 1O_2 .

A quantitative understanding of how Φ_F is used to calculate various photosynthetic parameters can be clarified by elucidation of the concept of a rate constant. In order to do so, we will make use of a simple

chemical model (Figure 10 above) involving first order decay of a population of $^1Chl^*$ to Chl via fluorescence. A differential expression to describe the rate of change over time in the concentration of the $^1Chl^*$ ($[^1Chl^*]$) is:

$$\frac{d[^1Chl^*]}{dt} = -k_F [^1Chl^*] \quad C-19$$

By convention, the negative sign depicts the fact that the $^1Chl^*$ population is decreasing. Multiplying both sides of the expression by $\frac{dt}{[^1Chl^*]}$ results in a useful

expression, both sides of which can be integrated:

$$\frac{d[^1Chl^*]}{[^1Chl^*]} = -k_F dt \quad C-20$$

Integrating both sides of the equation C-20, results in the following expression:

$$e^{\ln[^1Chl^*](t)} = e^{\ln[^1Chl^*](0)} e^{-kt} \quad C-21$$

$[^1Chl^*](t)$ and $[^1Chl^*](0)$ correspond to concentrations of the $^1Chl^*$ excited states at some arbitrary time t and time zero (the initial concentration), respectively. With a little algebraic rearrangement, equation C-21 becomes:

$$[^1Chl^*](t) = [^1Chl^*](0)e^{-kt} \quad C-22$$

Rather than relative to some arbitrary time t , equation C-22 can be defined with respect to what is referred to as the time constant, τ (45):

$$[^1Chl^*](\tau) = [^1Chl^*](0)e^{-k\tau} \quad C-23$$

By definition $\tau = 1/k$, so equation C-23 becomes:

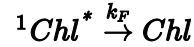


Figure 10. First order decay of $^1Chl^*$ to Chl .

$$\left[{}^1Chl^* \right] (\tau) = \left[{}^1Chl^* \right] (0)e^{-1} \quad C-24$$

Equation C-24 further reduces to:

$$\left[{}^1Chl^* \right] (\tau) = \left[{}^1Chl^* \right] (0) * 0.37 \quad C-25$$

So, equation C-25 indicates that τ represents the time it takes for $[{}^1Chl^*](0)$ to decrease by 63%. If τ is expressed in seconds, the corresponding rate constant is expressed in reciprocal seconds, or s^{-1} .

Quantum yield of fluorescence parameters

The mathematics of leaf-level Φ_F can now be more explicitly developed by a consideration of the assortment of rate constants for the above-mentioned photophysical processes (k_F , k_{IC} , and k_{ISC}), as well as two other apparent rate constants for processes that occur when chlorophyll molecules are integrated into the leaf photosynthetic machinery. Photochemistry (PC and K_{PC}) denotes the coupled electron transfer processes that comprise linear ETR . NPQ is a composite of reactions that serve to dissipate excess absorbed light energy (27 - 29, 53, 54). It should be noted that these five reactions predominate under most conditions in leaves to competitively dissipate populations of ${}^1Chl^*$, but there are a number of auxiliary reactions that can also be engaged under certain, unique conditions (30). Fluorescence flux (F_F) can be defined as (50):

$$F_F = Q * \frac{k_F}{k_S} \quad C-26$$

Q represents light intensity ($\mu\text{mol photons m}^{-2} \text{s}^{-1}$) and k_S is the sum of the above-mentioned rate constants:

$$k_S = \Sigma(k_F + k_{IC} + k_{ISC} + K_{PC} + K_{NPQ}) \quad C-27$$

The fluorescence yield Φ_F can then be defined as (50):

$$\Phi_F = \frac{F_F}{Q} \quad C-28$$

Combining equations C-26 and C-28 results in an expression that represents the convention for mathematically defining Φ_F as it is commonly formulated (49, 50):

$$\Phi_F = \frac{k_F}{\Sigma(k_F + k_{IC} + k_{ISC} + K_{PC} + K_{NPQ})} \quad C-29$$

The rate constants for *PC* and *NPQ* are considered apparent because they are in reality much more complex than a simple first order rate constant (55). K_{PC} is multiplied by Q_A (49, 56), the proportion of the oxidized primary electron acceptor within the PSII reaction center, and is defined as:

$$Q_A = \frac{[Q_A]}{[Q_A] + [Q_{A-}]} \quad \text{C-30}$$

Q_A and Q_{A-} represent the oxidized (open) and reduced (closed) chemical forms. Brackets indicate concentrations.

The PAM chlorophyll *a* fluorescence method, in conjunction with the saturation flash technique (48, 50), provides measurements of useful, well-defined Φ_F parameters (57, 58). From the darkened state of leaves, an estimate of the minimum Φ_F (F_o) can be measured:

$$F_o = \Phi_F = \frac{k_F}{k_F + k_{IC} + k_{ISC} + K_{PC}(Q_A=1)} \quad \text{C-31}$$

Two assumptions of equation C-31 are: 1) The proportion of oxidized Q_A is equal to 1; and 2) the apparent rate constant for *NPQ*, a process induced by actinic light, is zero; thus K_{NPQ} is omitted from the denominator. The darkened maximum Φ_F (F_m) is defined as:

$$F_m = \Phi_F = \frac{k_F}{k_F + k_{IC} + k_{ISC}} \quad \text{C-32}$$

F_m is measured during a short (approximately 1 second) (48), intense (4,000 to 25,000 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$) flash of light that is intended to cause $K_{PC}Q_A \rightarrow 0$ (the population of oxidized Q_A is assumed to approach zero and the apparent rate constant is removed from the denominator). The minimum Φ_F measured in a leaf under steady-state actinic illumination (F_s) is defined as:

$$F_s = \Phi_F = \frac{k_F}{k_F + k_{IC} + k_{ISC} + K_{PC}(0 < Q_A < 1) + K_{NPQ}} \quad \text{C-33}$$

The proportion of Q_A is assumed to attain a value somewhere between 0 and 1 depending upon light, stress level, etc. Actinic light induces *NPQ* formation and thus K_{NPQ} is included in the denominator of equation C-33. The maximum Φ_F under steady-state illumination (F_m') is defined as:

$$F_m' = \Phi_F = \frac{k_F}{k_F + k_{IC} + k_{ISC} + K_{NPQ}} \quad \text{C-34}$$

As with F_m , F_m' is determined during an intense, short flash of light that is intended to cause $K_{PC}Q_A \rightarrow 0$ and so this apparent rate constant does not appear in the denominator of equation C-34.

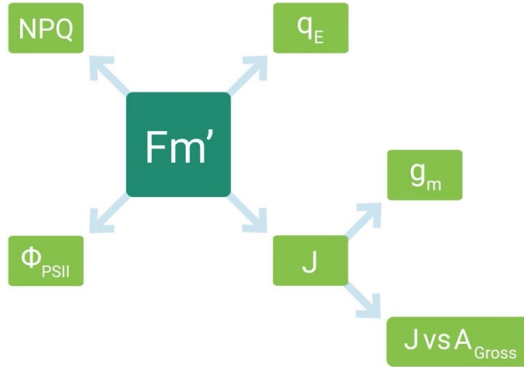


Figure 11. F_m' is useful to compute a variety of other parameters.

The leaf-level Φ_F 's (F_m , F_m' , etc.) can subsequently be used to determine many different aspects of the light reactions. The maximum quantum yield of PSII electron transfer (F_v/F_m), a sensitive indicator of various leaf-level, environmental stresses (59, 60), is defined as:

$$\frac{F_v}{F_m} = \frac{[F_m - F_o]}{F_m} = \frac{K_{PC}(Q_A = 1)}{k_F + k_{IC} + k_{ISC} + K_{PC}(Q_A = 1)} \quad \text{C-35}$$

F_m' is unique insofar as it can be used to determine important photosynthetic parameters under actinic illumination (Figure 11 above). The light acclimated, maximum quantum yield of PSII electron transfer (Φ_{PSII}) is calculated as:

$$\Phi_{PSII} = \frac{[F_m' - F_s]}{F_m'} = \frac{K_{PC}(0 < Q_A < 1)}{k_F + k_{IC} + k_{ISC} + K_{PC}(0 < Q_A < 1) + K_{NPQ}} \quad \text{C-36}$$

Φ_{PSII} can itself be used to quantify the actual flux of electrons ($\mu\text{mol e}^- \text{m}^{-2} \text{s}^{-1}$) (J) through PSII (that is a proxy for linear ETR) (25, 56, 61):

$$J = \Phi_{PSII} * i * \alpha * f_{II} \quad \text{C-37}$$

α , i , and f_{II} are leaf absorbance, incident actinic light intensity, and the fraction of absorbed light partitioned to PSII versus PSI (62). The quantum yield of NPQ (Φ_{NPQ}) can be estimated as (49, 63):

$$\Phi_{NPQ} = \frac{[F_m - F_m']}{F_m} * \frac{F_s}{F_m'} = \frac{k_{NPQ}}{k_F + k_{IO} + k_{ISC} + K_{PC}(0 < Q_A < 1) + K_{NPQ}} \quad \text{C-38}$$

NPQ is comprised of three different processes: 1) q_E ; 2) state transitions (q_T); and 3) and PSII inhibition (q_I) (27). The apparent rate constant K_{NPQ} is the sum of the apparent rate constants for these three processes ($K_{NPQ} = \Sigma(K_{qE} + K_{qT} + K_{qI})$). The quantum yield of q_E (Φ_{qE}) can be estimated as (49, 63):

$$\Phi_{qE} = \frac{(F_m'' - F_m')}{F_m''} * \frac{F_s}{F_m'} = \frac{K_{qE}}{k_F + k_{IO} + k_{ISC} + K_{PC}(0 < Q_A < 1) + K_{NPQ}} \quad \text{C-39}$$

F_m'' is the maximum Φ_F on the seconds-to-minutes timescale after cessation of actinic illumination. Comparison of (Φ_{qE}) and J over a range of environmental stress conditions can provide quantitative information about how light capture is regulated (14, 35). Regulation of light capture is essential for plant survival (64).

It deserves pausing at this juncture to quickly point out that simple, algebraic manipulation of expressions for Φ_F 's (F_m , F_m' , etc.) can be used to derive an assortment of altogether-different quantum yield expressions that represent useful photosynthetic parameters (49, 63).

Integrating gas exchange and light reaction parameters

Information from combining gas exchange and PAM chlorophyll a fluorescence parameters can be used to further elucidate physiological processes that are difficult to explore with one technique only. Comparison of PAM chlorophyll a fluorometry-derived estimates of J as a function of gross CO_2 assimilation ($A_{Gross} = A_{Net} - R_d$), as measured by gas analysis, can provide a measure of the electron requirement of CO_2 assimilation (16, 25, 65). The electron requirement of CO_2 assimilation is a quantitatively important parameter that can provide information about environmental stress, C_3 or C_4 photosynthesis, and the extent of photorespiration.

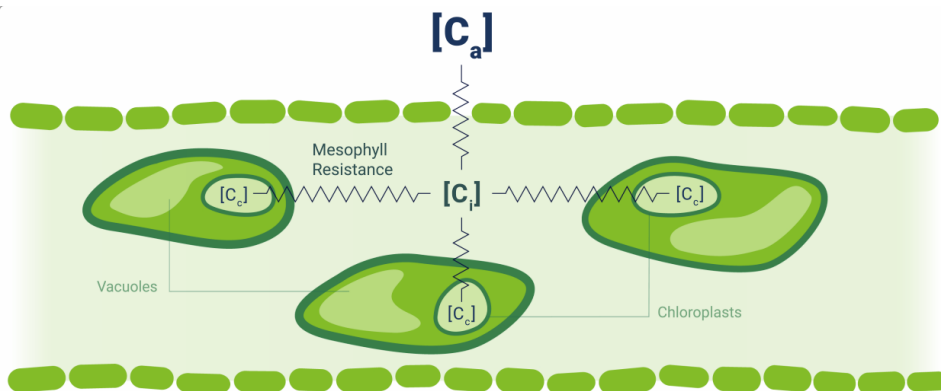


Figure 12.

Fluorescence-derived estimates of J can also be used, in conjunction with a rearranged version of the RUBP-regeneration-limited version of A_{Net} (equation C-18), to estimate g_m , the tortuous pathway of CO_2 diffusion from the leaf intercellular air space (C_i) to the site of carboxylation in the chloroplast, (C_c) (40, 66, 67):

$$g_m = \frac{A_{Net}}{C_i - \frac{\Gamma^* [J + 8(A + R_d)]}{J - 4(A + R_d)}} \quad \text{C-40}$$

Estimation of g_m also requires knowledge of A_{Net} , Γ^* , R_d , C_i , all of which can be measured via gas exchange.

Additional calculations

In addition to those calculations already described, the LI-6800 measures and computes the following parameters.

Light

The photosynthetic photon flux density (PPFD) readings Q_{in} and Q_{out} are computed from

$$Q_{in} = \frac{S_{qin} \times (2.5 \times 10^6) \times I_{qin}}{2 \times 16 \times 65536 \times 200} \quad \text{C-41}$$

$$Q_{out} = \frac{S_{qout} \times (2.5 \times 10^6) \times I_{qout}}{2 \times 32 \times 65536 \times 1470} \quad \text{C-42}$$

where S_{qin} and S_{qout} are the calibration factors, and I_{qin} and I_{qout} are the raw counts from the A/D converter.

Leaf temperature

Leaf temperatures T_{l1} , T_{l2} (°C) for the two thermocouples are computed from signals V_{l1} and V_{l2} (mV) by

$$T_{l1} = T_{j1} + \frac{V_{l1}}{(0.05845 + 0.0008V_{l1} + 0.0001T_{j1})} \quad \text{C-43}$$

$$T_{l2} = T_{j2} + \frac{V_{l2}}{(0.05845 + 0.0008V_{l2} + 0.0001T_{j2})} \quad \text{C-44}$$

where T_{j1} and T_{j2} are junction temperatures (°C).

Pressure

Atmospheric pressure P_a (kPa) is reported directly from the on-board sensor.

Chamber over-pressure ΔP_c (kPa) is computed from

$$\Delta P_c = V_p - V_{po} - (p_{ca}G^2 + p_{cb}F) \quad \text{C-45}$$

where V_p is the signal from the differential pressure sensor (in V, but taken to be kPa) V_{po} is the calibration zero for the sensor, G is fan speed (rpm), F is flow rate, and p_{ca} and p_{cb} are empirical coefficients that depend on chamber type (*Table C-1* below).

Table C-1. Values for pressure correction coefficients for each chamber type.

Chamber	pca	pcb
6800-01 Fluorometer	0	0
6800-01A Fluorometer	4.3321E-10	1.0215E-5
6800-12 3x3	0	0
6800-12A 3x3	2.6520E-10	1.3064E-5
6800-13 6x6	4.3998E-11	1.4792E-5
6800-17 Small Plant	0	0
6800-19 Custom	0	0

Flow

The flow F ($\mu\text{mol s}^{-1}$) to the leaf chamber is computed from

$$\begin{aligned} F &= (a_{f1} + (a_{f2} + (a_{f3} + a_{f4}x)x)x) \quad \text{C-46} \\ x &= (V_f - V_{fo})(1 - (T_k - a_{f5})(a_{f6} + (V_f - V_{fo})a_{f7})) \end{aligned}$$

where $af1\dots af7$ are factory calibration constants, V_f is flow meter signal (V), V_{fo} is calibration zero, and T_k is IRGA block temperature.

Reference and sample IRGA cell flows F_r and F_s ($\mu\text{mol s}^{-1}$) are computed from

$$\begin{aligned} F_r &= (a_{r1} + (a_{r2}T_r + (a_{r3}T_r + a_{r4}v)v)v) \quad \text{C-47} \\ v &= V_{fr} - V_{fro} \end{aligned}$$

$$\begin{aligned} F_s &= (a_{s1} + (a_{s2}T_s + (a_{s3}T_s + a_{s4}v)v)v) \quad \text{C-48} \\ v &= V_{fs} - V_{fso} \end{aligned}$$

where V_{fr} and V_{fs} are cell exit flow sensor voltage, V_{fro} and V_{fso} are the offset value of that voltage, and $ar1\dots ar4$ and $as1\dots as4$ are factory calibration coefficients.

CO₂

Reference and sample CO₂ concentrations C_r and C_s are given by

$$\begin{aligned} C_r &= C_b \quad \text{C-49} \\ C_s &= C_a + M_c(C_a) \end{aligned}$$

where $M_c()$ is the CO₂ match correction function:

$$M_c(x) = m_{c0} + m_{c1}x + m_{c2}x^2 + m_{c3}x^3 \quad \text{C-50}$$

$$C_a = f_c \left(\frac{\alpha_{cs}(S_{cso} + \alpha_{cs}S_{cs1})}{P_a \Psi_c(W_a, X_o)} \right) (T_s + 273.15) \Psi_c(W_a, X_o) \quad \text{C-51}$$

$$C_b = f_c \left(\frac{\alpha_{cr}(S_{cro} + \alpha_{cr}S_{cr1})}{P_a \Psi_c(W_b, X_o)} \right) (T_r + 273.15) \Psi_c(W_b, X_o) \quad \text{C-52}$$

where α_{cr} and α_{cs} are absorptances, P_a is atmospheric pressure, T_r and T_s are the cell inlet temperatures, f_c is a 5th order polynomial with coefficients ($br1\dots br5$ or

bs1...bs5) determined at the factory, S_{cro} ; S_{cso} and S_{cr1} ; S_{cs1} are the span1 and span2 user calibration settings, W_a and W_b are water concentrations, X_o is the oxygen concentration in percent. Ψ_c is the band broadening function for the effect of water and oxygen on CO_2 , given by

$$\Psi_c(W, X_o) = 1 + (B_{wc} - 1) \frac{W}{1000} + (B_{oc} - 1) \frac{X_o}{100} \quad \text{C-53}$$

where B_{wc} and B_{oc} are the band broadening coefficients for H_2O and O_2 respectively on CO_2 .

Reference and sample absorptances α_{cr} and α_{cs} are corrected for zero drift with temperature, span drift with temperature, and cross sensitivity with water.

$$\alpha_{cr} = \alpha' (1 + (T_k - \sigma_{cr3})(\sigma_{cr1} + \alpha' \sigma_{cr2})) \quad \text{C-54}$$

$$\alpha' = 1 - \left(\frac{I_{cr}}{I_{cro}} + X_{wcr} \left(1 - \frac{I_{wr}}{I_{wro}} \right) \right) (Z_{cro} + Z_{cr} * T_k)$$

$$\alpha_{cs} = \alpha' (1 + (T_k - \sigma_{cs3})(\sigma_{cs1} + \alpha' \sigma_{cs2})) \quad \text{C-55}$$

$$\alpha' = 1 - \left(\frac{I_{cs}}{I_{cso}} + X_{wcs} \left(1 - \frac{I_{ws}}{I_{wso}} \right) \right) (Z_{cso} + Z_{cs} * T_k)$$

where *cr1...cr3* and *cs1...cs3* are empirical coefficients for CO_2 absorptance span drift determined during calibration, I_{cr} , I_{cro} , I_{cs} , and I_{cso} are the raw IRGA detector absorbing and non-absorbing readings for CO_2 , and I_{wr} , I_{wro} , I_{ws} and I_{wso} are those for water. X_{wcr} and X_{wcs} are empirical cross sensitivity coefficients for water on CO_2 determined during calibration, Z_{cro} and Z_{cso} are the current user CO_2 zero factors, and Z_{cr} and Z_{cs} are the factors for CO_2 zero drift with temperature, determined during calibration.

Dry mole fractions C_{rd} and C_{sd} are computed from

$$C_{rd} = \frac{C_r}{1000 - W_r} \quad \text{C-56}$$

$$C_{sd} = \frac{C_s}{1000 - W_s}$$

H₂O

Reference and sample H₂O concentrations H_r and H_s are given by

$$\begin{aligned} W_r &= W_b \\ W_s &= W_a + M_w(W_a) \end{aligned} \quad \text{C-57}$$

where $M_w()$ is the H₂O match correction function:

$$M_w(x) = m_{wo} + m_{w1}x + m_{w2}x^2 + m_{w3}x^3 \quad \text{C-58}$$

$$W_a = f_w \left(\frac{\alpha_{ws}(S_{wso} + \alpha_{ws}S_{ws1})}{P_a} \right) (T_s + 273.15) \Psi_o(X_o) \quad \text{C-59}$$

$$W_b = f_w \left(\frac{\alpha_{wr}(S_{wro} + \alpha_{wr}S_{wr1})}{P_a} \right) (T_r + 273.15) \Psi_o(X_o) \quad \text{C-60}$$

where α_{wr} and α_{ws} are the reference and sample absorptances, f_w is a 3rd order polynomial with coefficients ($dr1...dr3$ and $ds1...ds3$) determined at the factory, S_{wro} , S_{wr1} and S_{wso} , S_{ws1} are the H₂O span1 and span2 user calibration settings for reference and sample, Ψ_o is the band broadening function for the effect of oxygen on H₂O, given by

$$\Psi_o(X_o) = B_{ow1} + B_{ow2}X_o \quad \text{C-61}$$

where B_{ow1} and B_{ow2} are empirical coefficients.

Absorptances α_{wr} and α_{ws} are corrected for zero drift with temperature, span drift with temperature, and cross sensitivity with water.

$$\alpha_{wr} = \alpha' (1 + (T_k - \sigma_{wr3})(\sigma_{wr1} + \alpha' \sigma_{wr2})) \quad \text{C-62}$$

$$\alpha' = 1 - \left(\frac{I_{cr}}{I_{cro}} + X_{cwr} \left(1 - \frac{I_{wr}}{I_{wro}} \right) \right) (Z_{cro} + Z_{cr}T_k)$$

$$\alpha_{ws} = \alpha' (1 + (T_k - \sigma_{ws3})(\sigma_{ws1} + \alpha' \sigma_{ws2})) \quad \text{C-63}$$

$$\alpha' = 1 - \left(\frac{I_{cs}}{I_{cso}} + X_{cws} \left(1 - \frac{I_{ws}}{I_{wso}} \right) \right) (Z_{cso} + Z_{cs}T_k)$$

where $w_{r1}...w_{r3}$ and $w_{s1}...w_{s3}$ are empirical coefficients for H₂O absorptance span drift determined during calibration. X_{cwr} , X_{cws} are empirical cross sensitivity coefficients for CO₂ on H₂O determined during calibration, Z_{wro} and Z_{wso} are the current user H₂O zero factors, and Z_{wr} and Z_{ws} are the factors for H₂O zero drift with temperature, determined during calibration.

The vapor pressure (kPa) of the air in the reference and sample cells e_r and e_s is given by

$$e_r = \frac{W_r P_a}{1000.0} \quad \text{C-64}$$

$$e_s = \frac{W_s P_a}{1000.0} \quad \text{C-65}$$

Reference and sample cell dew point temperatures T_{dr} and T_{ds} are given by

$$T_{dr} = \frac{240.97x}{17.502 - x} \quad \text{C-66}$$

$$x = \ln\left(\frac{e_r}{0.61365}\right)$$

$$T_{ds} = \frac{240.97x}{17.502 - x} \quad \text{C-67}$$

$$x = \ln\left(\frac{e_s}{0.61365}\right)$$

Gas exchange

Humidity indicators

The leaf chamber vapor pressure e_c (kPa) is given by

$$e_c = \frac{W_r (P_a + \Delta P_c)}{1000.0} \quad \text{C-68}$$

The saturation vapor pressure e_{sc} in the leaf chamber is a function of chamber air temperature T_a :

$$e_{sc} = e_s(T_a) \quad \text{C-69}$$

where $e_s(T)$ is the saturation vapor pressure function:

$$e_s(T) = 0.6135e^{\left(\frac{17.502T}{240.97 + T}\right)} \quad \text{C-70}$$

The relative humidity h_c (%) in the leaf chamber is given by

$$h_c = \frac{e_c}{e_{cs}} 100 \quad \text{C-71}$$

The vapor pressure deficit of the leaf $e_{\Delta l}$ is computed from

$$e_{\Delta l} = e_s(T_l) - e_c \quad \text{C-72}$$

where T_l is leaf temperature (°C).

Transpiration

The mass balance of water vapor in an open system is given by

$$sE = u_o w_o - u_e w_e \quad \text{C-73}$$

where s is leaf area (m^2), E is transpiration rate ($\text{mol H}_2\text{O m}^{-2} \text{s}^{-1}$), u_e and u_o are incoming and outgoing flow rates (mol s^{-1}) from the chamber, and w_e and w_o are incoming and outgoing water mole fractions ($\text{mol H}_2\text{O (mol air)}^{-1}$). Since

$$u_o = u_e + sE \quad \text{C-74}$$

we can write

$$sE = (u_e + sE)w_o - u_e w_e \quad \text{C-75}$$

which rearranges to

$$E = \frac{u_e(w_o - w_e)}{s(1 - w_o)} \quad \text{C-76}$$

The relationships between the terms in equations C-73 through C-76 and what the LI-6800 measures are

$$\begin{aligned} u_e &= F/10^6 & \text{C-77} \\ w_e &= W_r/10^3 \\ w_o &= W_s/10^3 \\ s &= S/10^4 \end{aligned}$$

where F is air flow rate ($\mu\text{mol s}^{-1}$), W_s and W_r are sample and reference water mole fractions ($\text{mmol H}_2\text{O (mol air)}^{-1}$), and S is leaf area (cm^2). The equation that the LI-6850 uses for transpiration is thus

$$E = \frac{F(W_s - W_r)}{100S(1000 - W_s)} \quad \text{C-78}$$

Stomatal conductance

The total conductance g_{tw} of the leaf to water vapor is

$$g_{tw} = \frac{E \left(1000 - \frac{W_l + W_s}{2} \right)}{W_l - W_s} \quad \text{C-79}$$

where W_l is the molar concentration of the water vapor within the leaf ($\text{mmol H}_2\text{O (mol air)}^{-1}$), which is computed from the leaf temperature T_l ($^{\circ}\text{C}$) and the total atmospheric pressure in the leaf chamber

$$W_l = \frac{e(T_l)}{P_a + \Delta P_c} 1000 \quad \text{C-80}$$

We assume that the total resistance for the upper r^u or lower r^l surface of a leaf is the sum of the stomatal resistance r_s and boundary layer resistance r_b of that surface

$$r^l = r_s^l + r_b^l \quad \text{C-81}$$

$$r^u = r_s^u + r_b^u \quad \text{C-82}$$

and that the upper and lower boundary layer resistances are the same

$$r_b = r_b^u = r_b^l \quad \text{C-83}$$

and we define K to be the ratio of stomatal resistances of the two sides

$$K = \frac{r_s^u}{r_s^l} \quad \text{C-84}$$

Leaf stomatal resistance r_s is given by

$$\frac{1}{r_s} = \frac{1}{r_s^u} + \frac{1}{r_s^l} \quad \text{C-85}$$

$$= \frac{1}{K r_s^l} + \frac{1}{r_s^l} \quad \text{C-86}$$

$$= \frac{1}{K r_s^l} + \frac{K}{K r_s^l} \quad \text{C-87}$$

$$= \frac{1 + K}{K r_s^l} \quad \text{C-88}$$

Total conductance g (the inverse of the total resistance r) can thus be written

$$g = \frac{1}{r} = \frac{1}{r_s^u + r_b} + \frac{1}{r_s^l + r_b} \quad \text{C-89}$$

$$= \frac{1}{K \left(\frac{1+K}{K} \right) r_s + r_b} + \frac{1}{\left(\frac{1+K}{K} \right) r_s + r_b} \quad \text{C-90}$$

$$= \frac{1}{(1+K)r_s + r_b} + \frac{K}{(1+K)r_s + K r_b} \quad \text{C-91}$$

For water vapor, the total conductance g_{tw} is related to stomatal conductance g_s and one sided boundary layer conductance g_{bw} by

$$g_{tw} = \frac{1}{(1+K)\frac{1}{g_{sw}} + \frac{1}{g_{bw}}} + \frac{K}{(1+K)\frac{1}{g_{sw}} + K\frac{1}{g_{bw}}} \quad \text{C-92}$$

Solving equation C-90 for g_{sw} yields

$$g_{sw} = \frac{2}{\left(\frac{1}{g_{tw}} - \frac{1}{g_{bw}}\right) + \sqrt{\left(\frac{1}{g_{tw}} - \frac{1}{g_{bw}}\right)^2 + \frac{4K}{(K+1)^2} \left(2\frac{1}{g_{tw}} - \frac{1}{g_{bw}}\right) \frac{1}{g_{bw}}}} \quad \text{C-93}$$

Note that although we defined K to be a particular ratio of upper to lower stomata resistances (equation C-84), since K appears as $\frac{K}{(K+1)^2}$, we get the same math-

ematical result if we had defined it the other way. In other words, K is equivalent to $1/K$. Note also that the LI-6400 does not use equation C-93, but rather a simplified approximation:

$$g_{sw}^{6400} = \frac{1}{\frac{1}{g_{tw}} - \frac{K^2 + 1}{(K+1)^2 g_{bw}}} \quad \text{C-94}$$

Boundary layer

The one sided boundary layer conductance to water vapor g_{bw} for a broadleaf is a function of fan speed G (rpm) and leaf area S (cm²).

$$g_{bw}(f, s) = c_o + c_1 f + c_2 f s^2 + c_3 s f + c_4 f^2 \quad \text{C-95}$$

where f is

$$f = \frac{GP_a}{1000P_o} \quad \text{C-96}$$

and s is forced to be

$$S_{min} \geq s \leq S_{max} \quad \text{C-97}$$

The empirical coefficients $c_0 \dots c_4$, reference pressure P_o , and leaf area limits S_{min} and S_{max} depend on chamber type.

Table C-2. Values for boundary layer conductance for chamber types.

Chamber	c_0	c_1	c_2	c_3	c_4	P_o	S_{min}	S_{max}
6800-01 Flr	0.250	0.35860	-4.01816E-3	0.00451074	-0.0044762	96.9	1	6
6800-01A 6cm ²	0.578	0.5229739	3.740252E-3	-6.197961E-2	-5.608586E-3	96.9	1	6
6800-01A 2cm ²	0.572	0.3872742	-1.870584E-2	0.0	-7.37389E-3	96.9	1	2
6800-12 3x3	0.500	0.44869569	1.9000035E-3	-4.26088781E-2	-3.456516E-3	96.7	2	9
6800-12A 9cm ²	0.579	0.3210639	-1.109987E-3	5.106816E-3	-3.283688E-3	96.7	2	9
6800-12A 6cm ² FB	0.345	0.552336	-4.7985e-3	0.0	-7.3557e-3	96.7	1	6
6800-12A 6cm ² SS	0.418	0.5145466	-2.5106E-3	0.0	-8.1206E-3	96.7	1	6
6800-12A 3cm ² FB	0.188	0.5795409	-1.15295E-2	0.0	-9.7259E-3	96.7	1	3
6800-12A 3cm ² SS	0.141	0.5263354	-1.27376E-2	0.0	-1.10157E-2	96.7	1	3
6800-13 6x6	0.430	0.267827	-1.164018E-4	2.248202E-3	-5.109462E-3	96.8	6	36

Net assimilation

The mass balance of CO₂ in an open system is given by

$$sa = u_e c_e - u_o c_o \quad \text{C-98}$$

where a is assimilation rate (mol CO₂ m⁻¹ s⁻¹), c_e and c_o are entering and outgoing mole fractions (mol CO₂ (mol air)⁻¹). Using equation C-74, we can write

$$sa = u_e c_e - (u_e + sE)c_o \quad \text{C-99}$$

which rearranges to

$$a = \frac{u_e(c_e - c_o)}{s} - Ec_o \quad \text{C-100}$$

To write equation C-100 in terms of what the LI-6800 measures, we use equations C-77 and

$$c_e = C_r / 10^6 \quad \text{C-101}$$

$$c_o = C_s / 10^6$$

$$a = A / 10^6$$

where C_r and C_s are sample and reference CO_2 concentrations ($\mu\text{mol mol}^{-1}$), and A is the net assimilation by the leaf ($\mu\text{mol m}^{-2} \text{s}^{-1}$). Substitution yields

$$A = \frac{F(C_r - C_s)}{100S} - C_s E \quad \text{C-102}$$

$$\begin{aligned} & F \left(C_r - C_s - C_s \left(\frac{W_s - W_r}{1000 - W_s} \right) \right) \\ = & \frac{\hspace{1.5cm}}{100S} \end{aligned} \quad \text{C-103}$$

$$\begin{aligned} & F \left(C_r - C_s \left(\frac{1000 - W_r}{1000 - W_s} \right) \right) \\ = & \frac{\hspace{1.5cm}}{100S} \end{aligned} \quad \text{C-104}$$

Intercellular CO_2

The intercellular CO_2 concentration C_i ($\mu\text{mol CO}_2 (\text{mol air})^{-1}$) is given by

$$C_i = \frac{\left(g_{tc} - \frac{E}{2} \right) C_s - A}{g_{tc} + \frac{E}{2}} \quad \text{C-105}$$

where g_{tc} is the total conductance to CO_2 . From equation C-91, we can write

$$g_{tc} = \frac{1}{(1 + K) \frac{1.6}{g_{sw}} + \frac{1.37}{g_{bw}}} + \frac{K}{(1 + K) \frac{1.6}{g_{sw}} + K \frac{1.37}{g_{bw}}} \quad \text{C-106}$$

where 1.6 is the ratio of the diffusivities of CO_2 and water in air, and 1.37 is the same ratio for the boundary layer. This is another departure from the LI-6400, which does not use equation C-106, but a simplified approximation

$$g_{tc}^{6400} = \frac{1}{\frac{1.6}{g_{sw}} - \frac{1.37(K^2 + 1)}{(K + 1)^2 g_{bw}}} \quad \text{C-107}$$

Energy balance

The LI-6800 provides several potential sources for leaf temperature: it can be directly measured with some combination of the two leaf thermocouples (T_{l1} and T_{l2}), or computed indirectly from a leaf energy balance (T_{eb}). The user can specify what combination to use via three weighting factors f_{T1} , f_{T2} , and f_{Teb} :

$$T_l = f_{T1}T_{l1} + f_{T2}T_{l2} + f_{Teb}T_{eb} \quad \text{C-108}$$

The energy balance temperature T_{eb} assumes that the energy balance of a leaf in the chamber has three components: net radiation R (W m^{-2}), sensible heat flux H (W m^{-2}), and latent heat flux L (W m^{-2}), and that they all sum to zero:

$$0 = H + L + R \quad \text{C-109}$$

We consider two components of net radiation: short wave (visible and near IR) and thermal.

$$R = R_{abs} + R_{nt} \quad \text{C-110}$$

where R_{abs} is absorbed short wave, and R_{nt} is net thermal. The absorbed short wave radiation is computed by

$$R_{abs} = Q_{abs}k \quad \text{C-111}$$

where Q_{abs} is the absorbed irradiance by the leaf ($\mu\text{mol m}^{-2} \text{s}^{-1}$), and k is the conversion factor for transforming ($\mu\text{mol m}^{-2} \text{s}^{-1}$) to (W m^{-2}), based on the spectral characteristics of the light source.

The net thermal balance is based on the leaf temperature T_l and the surrounding chamber wall temperature T_w , so the total radiation balance R can be written as

$$R_{nt} = 2\epsilon\sigma ((T_w + 273)^4 - (T_l + 273)^4) \quad \text{C-112}$$

where ϵ is the thermal emissivity of the leaf (usually assumed to be 0.95), and σ is the Stefan-Boltzmann constant ($5.67 \text{ W m}^{-2} \text{ K}^{-1}$). The 2's in equation C-110

account for both sides of the leaf. Wall temperature T_w is not measured, but depends on a user-specified offset ΔT_w from chamber air temperature.

$$T_w = T_a + \Delta T_w \quad \text{C-113}$$

The latent heat flux L is the transpiration rate E converted to W m^{-2} .

$$L = 44100E \quad \text{C-114}$$

The sensible heat flux H is a function of the leaf - chamber air temperature difference $T_l - T_a$, the specific heat capacity of the air c_p ($29.3 \text{ J mol}^{-1} \text{ K}^{-1}$) and the one sided boundary layer conductance for heat transfer of the leaf g_{bh} , which is 0.92 times the boundary layer conductance for water vapor g_{bw} .

$$H = 2c_p g_{bh} (T_l - T_a) \quad \text{C-115}$$

$$= 1.84c_p g_{bw} (T_l - T_a) \quad \text{C-116}$$

Equation C-109 becomes:

$$0 = 1.84c_p g_{bw} (T_l - T_a) + 44100E + R_{abs} + 2\epsilon\sigma ((T_w + 273)^4 - (T_l + 273)^4) \quad \text{C-117}$$

If we let $\Delta T = T_l - T_a$, and note for small ΔT

$$(T_l + 273)^4 \approx (T_a + 273)^4 + 4(T_a + 273)^3 \Delta T \quad \text{C-118}$$

Substituting equation C-118 into C-117 and solving for ΔT yields

$$\Delta T = \frac{R_{abs} + 2\epsilon\sigma ((T_w + 273)^4 - (T_a + 273)^4) - 44100E}{1.84c_p g_{bw} + 8\epsilon\sigma (T_a + 273)^3} \quad \text{C-119}$$

The energy balance leaf temperature T_{eb} is then

$$T_{eb} = T_a + \Delta T \quad \text{C-120}$$

Sensor head calibration coefficients

Table C-3. Sensor head calibration coefficients, with XML location.

Symbol	Description	XML Locator (/licor/li6850/...)
$a_{f1}...a_{f7}$	Main flow sensor	../factory/flowmeter/a...g
$a_{r1}...a_{r4}$	Ref flow sensor	../factory/irga_a/flow/a1...a4
$a_{s1}...a_{s4}$	Sample flow sensor	../factory/irga_b/flow/a1...a4
B_{wc}	Band broadening coefficient for water on CO ₂	../factory/bb/ch
B_{oc}	Band broadening coefficient for oxygen on CO ₂	../factory/bb/cx
B_{ow1} , B_{ow2}	Band broadening correction for oxygen on water	../factory/bb/hx0, hx1
$b_{r1}...b_{r5}$	Reference CO ₂ calibration coefficients	../factory/irga_b/co2/a1...a5
$b_{s1}...b_{s5}$	Sample CO ₂ calibration coefficients	../factory/irga_a/co2/a1...a5
$d_{r1}...d_{r3}$	Reference H ₂ O calibration coefficients	../factory/irga_b/h2o/a1...a3
$d_{s1}...d_{s3}$	Sample H ₂ O calibration coefficients	../factory/irga_b/h2o/a1...a3
$m_{co}...m_{c3}$	CO ₂ match coefficients	../cfg/match/co2_adj ... co2 adj 3
$m_{wo}...m_{w3}$	H ₂ O match coefficients	../cfg/match/h2o_adj ... h2o adj 3
p_{ca}	Pressure correction or fan speed and flow rate	../factory/chamber/pca
p_{cb}	Pressure correction or fan speed and flow rate	../factory/chamber/pcb
$\sigma_{cr1}... \sigma_{cr3}$	CO ₂ reference absorptance span drift with temp	../factory/irga_b/co2/s1...s3
$\sigma_{cs1}... \sigma_{cs3}$	CO ₂ sample absorptance span drift with temp	../factory/irga_a/co2/s1...s3
$\sigma_{wr1}... \sigma_{wr3}$	H ₂ O reference absorptance span drift with temp	../factory/irga_b/h2o/s1...s3
$\sigma_{ws1}... \sigma_{ws3}$	H ₂ O sample absorptance span drift with temp	../factory/irga_a/h2o/s1...s3
S_{cro}	Span1 for reference CO ₂	../cal/co2bspan1
S_{cr1}	Span2 for reference CO ₂	../cal/co2bspan2
S_{cso}	Span1 for sample CO ₂	../cal/co2aspan1
S_{cs1}	Span2 for sample CO ₂	../cal/co2aspan2
S_{qin}	In-chamber light sensor cal	../cfg/ppfdin/mult
S_{qout}	External quantum sensor cal	../cfg/ppfdout/mult
S_{wro}	Span1 for reference H ₂ O	../cal/h2obspan1
S_{wr1}	Span2 for reference H ₂ O	../cal/h2obspan2
S_{wso}	Span1 for sample H ₂ O	../cal/h2oaspan1
S_{ws1}	Span2 for sample H ₂ O	../cal/h2oaspan2
V_{fo}	Zero offset for main flow meter	../cal/flowmeterzero
V_{fro}	Zero offset for reference flow meter	../cal/flowbzero
V_{fso}	Zero offset for sample flow meter	../cal/flowazero
V_{po}	Zero parameter for differential pressure sensor	../cal/chamberpressurezero
X_o	Oxygen concentration, percent	../factory/cal/oxygen

Table C-3. Sensor head calibration coefficients, with XML location. (...continued)

Symbol	Description	XML Locator (/licor/li6850/...)
X_{cwr}	Cross sensitivity, CO ₂ on H ₂ O, reference cell	../factory/irga_b/xhc
X_{cws}	Cross sensitivity, CO ₂ on H ₂ O, sample cell	../factory/irga_b/xch
X_{wcr}	Cross sensitivity, H ₂ O on CO ₂ , reference cell	../factory/irga_a/xhc
X_{wcs}	Cross sensitivity, H ₂ O on CO ₂ , sample cell	../factory/irga_a/xch
Z_{cr}	Zero drift with temperature for reference CO ₂	../factory/irga_b/z
Z_{cro}	Zero offset for reference CO ₂	../cal/co2bzero
Z_{cs}	Zero drift with temperature for sample CO ₂	../factory/irga_a/z
Z_{cso}	Zero offset for sample CO ₂	../cal/co2azero
Z_{wr}	Zero drift with temperature for reference H ₂ O	../factory/irga_b/z
Z_{wro}	Zero offset for reference H ₂ O	../cal/h2obzero
Z_{ws}	Zero drift with temperature for sample H ₂ O	../factory/irga_a/z
Z_{wso}	Zero offset for sample H ₂ O	../cal/h2obzero

Sensor measurements and computations

Table C-4. Sensor measurements and computations, and where to find them in the Data Dictionary.

Symbol	Description	Name (Label)	Group
α_{cr}	Reference cell CO ₂ absorptance	abs_c_b	Raw
α_{cs}	Sample cell CO ₂ absorptance	abs_c_a	Raw
α_{wr}	Reference cell H ₂ O absorptance	abs_h_b	Raw
α_{ws}	Sample cell H ₂ O absorptance	abs_h_a	Raw
$c_{0...c4}$	Boundary layer function coeffs	b1c_a...b1c_e	ChambConst
C_a	Sample cell CO ₂ , not adjusted for match	CO2_a	Meas
C_b	Sample cell CO ₂ ($\mu\text{mol mol}^{-1}$)	CO2_b	Meas2
C_r	Reference cell CO ₂ ($\mu\text{mol mol}^{-1}$)	CO2_r	Meas
C_{rd}	Reference cell CO ₂ , dry mole fraction	CO2_r_d	Meas
C_s	Sample cell CO ₂ ($\mu\text{mol mol}^{-1}$)	CO2_s	Meas
C_{sd}	Sample cell CO ₂ , dry mole fraction	CO2_s_d	Meas
ΔP_c	Chamber over pressure (kPa)	Pchamber (ΔP_{cham})	Meas
e_r	Reference cell vapor pressure (kPa)	e_r	Meas2
e_s	Sample cell vapor pressure (kPa)	e_s	Meas2
I_{cr}	Reference CO ₂ raw detector count	Wc_r	Raw
I_{cro}	Reference CO ₂ raw detector reference count	Wco_r	Raw
I_{cs}	Sample CO ₂ , raw detector count	Wc_s	Raw
I_{cso}	Sample CO ₂ raw detector reference count	Wco_s	Raw
I_{qin}	In-chamber PPF sensor raw counts		
I_{qout}	External quantum sensor raw counts		

Table C-4. Sensor measurements and computations, and where to find them in the Data Dictionary. (...continued)

Symbol	Description	Name (Label)	Group
I_{wr}	Reference H ₂ O raw detector count	Ww_r	Raw
I_{wro}	Reference H ₂ O raw detector reference count	Wwo_r	Raw
I_{ws}	Sample H ₂ O raw detector count	Ww_s	Raw
I_{wso}	Sample H ₂ O raw detector reference count	Wwo_s	Raw
P_a	Atmospheric pressure (kPa)	Press (Pa)	Meas
F	Flow to chamber ($\mu\text{mol s}^{-1}$)	Flow	Meas
F_r	Flow from reference cell ($\mu\text{mol s}^{-1}$)	Flow_r	Status
F_s	Flow from sample cell ($\mu\text{mol s}^{-1}$)	Flow_s	Status
G	Chamber fan rotation rate (rpm)	Fan speed	Status
Q_{in}	In-chamber PPFD	PPFD_in (Q_amb_in)	Meas
Q_{out}	External PPFD	PPFD_out (Q_amb_out)	Meas
T_a	Leaf chamber air temperature (C)	Tchamber (Tair)	Meas
T_{dr}	Dewpoint temperature reference cell (C)	Td_r	Meas2
T_{ds}	Dewpoint temperature sample cell (C)	Td_s	Meas2
T_{j1}	Leaf T/C 1 junction temperature (C)	Tleafjunction (Tleaf_j)	Status2
T_{j2}	Leaf T/C 2 junction temperature (C)	Tleafjunction2 (Tleaf2_j)	Status2
T_k	IRGA block temperature (C)	Tirga_block (Tirga)	Status
T_{l1}	Leaf temperature 1 (C)	Tleaf	Meas
T_{l2}	Leaf temperature 2 (C)	Tleaf2	Meas
T_r	Reference cell inlet temperature (C)	Tb (Tr)	Status
T_s	Sample cell inlet temperature (C)	Ta (Ts)	Status
V_f	Signal (V) from main flow sensor	Flow	Raw
V_{fr}	Signal (V) from reference flow sensor	Flow_b_v (Flow_r_v)	Raw
V_{fs}	Signal (V) from sample flow sensor	Flow_a_v (Flow_s_v)	Raw
V_{l1}	Leaf temperature 1 signal (mV)	leaf_t_mv (Tleaf_mv)	Raw
V_{l2}	Leaf temperature 2 signal (mV)	leaf2_t_mv (Tleaf2_mv)	Raw
V_p	Differential pressure signal (V or kPa)	VPchamber	Raw
W_a	Sample cell H ₂ O not corrected for match	H2O_a	Meas
W_b	Reference cell H ₂ O	H2O_a	Meas2
W_r	Reference cell H ₂ O (mmol mol^{-1})	H2O_r	Meas
W_s	Sample cell H ₂ O (mmol mol^{-1})	H2O_s	Meas

Gas exchange variables

Table C-5. Gas exchange variables, and where to find them in the Data Dictionary.

Symbol	Description	Name (Label)	Group
A	Net assimilation rate ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	A	GasEx
$c_0 \dots c_4$	Boundary layer function coeffs	blc_a...blc_e	ChambConst
C_i	Intercellular CO_2 ($\mu\text{mol mol}^{-1}$)	Ci	GasEx
ΔT_w	Difference between wall and chamber air temperature (C)	deltaTw	LTConst
e_c	Leaf chamber vapor pressure (kPa)	VPcham	GasEx
e_{cs}	Leaf chamber saturation vapor pressure (kPa)	SVPcham	GasEx
$e_{\Delta l}$	Leaf vapor pressure deficit (kPa)	VPDleaf	GasEx
E	Transpiration rate ($\text{mol m}^{-2} \text{s}^{-1}$)	E	GasEx
f_{T1}	Leaf temp weighting factor for T_{I1}	fT1	LTConst
f_{T2}	Leaf temp weighting factor for T_{I2}	fT2	LTConst
f_{Teb}	Leaf temp weighting factor for T_{leb}	fTeb	LTConst
g_{tc}	Total leaf conductance to CO_2 ($\text{mol m}^{-2} \text{s}^{-1}$)	gtc	GasEx
g_{tw}	Total leaf conductance to H_2O ($\text{mol m}^{-2} \text{s}^{-1}$)	gtw	GasEx
g_{bw}	1-sided boundary layer conductance to H_2O ($\text{mol m}^{-2} \text{s}^{-1}$)	gbw	GasEx
g_{sw}	Stomatal conductance to H_2O ($\text{mol m}^{-2} \text{s}^{-1}$)	gsw	GasEx
H	Energy balance Sensible heat flux (W m^{-2})	SenHFlux	GasEx
h_c	Leaf chamber relative humidity (%)	RHcham	GasEx
K	Ratio of stomatal density of leaf sides	K	GasEx
k	Converts $\mu\text{mol m}^{-2} \text{s}^{-1}$ to W m^{-2}	convert	LeafQ
L	Energy balance latent heat flux (W m^{-2})	LatHFlux	GasEx
Q_{abs}	Absorbed radiation ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	Qabs	LeafQ
Q_{in}	Incident radiation ($\mu\text{mol m}^{-2} \text{s}^{-1}$)	Qin	LeafQ
R_{abs}	Absorbed short wave radiation (W m^{-2})	Rabs	GasEx
R_{nt}	Net thermal radiation (W m^{-2})	NetTherm	GasEx
S	Leaf area (cm^{-2})	S	GasEx
T_{eb}	Energy balance leaf temperature (C)	TleafEB	GasEx
T_l	Effective leaf temperature (C)	TleafCnd	GasEx
$R_{abs} + R_{nt} + H + L$	Sum of energy balance components	EBSum	GasEx

Mass balance derivation using a two-compartment model

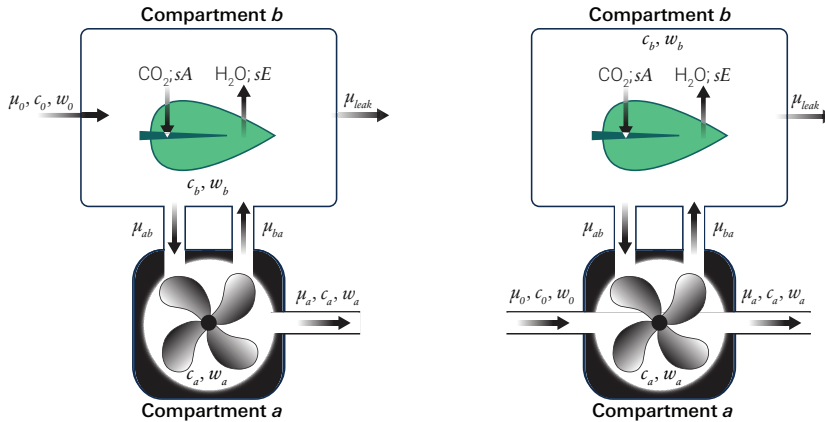


Figure C-13. The two-compartment model for this analysis for chambers (left) and original 6800-01 and 6800-12 chambers (right).

A basic assumption of the LI-6800's gas exchange equations is that the chamber volume is a single, well-mixed volume. A quick look at the instrument, however, tells you that we have a chamber volume that is attached to the rest of the system (and mixing fan) by four out-and-return channels, so there really are two volumes: one for the chamber and one for the mixing fan and temperature controller, that are connected by four air passages.

A more careful analysis leads to this conclusion: when fan speed is high (10,000 rpm or greater) and there is little leakage ($< 100 \mu\text{mol s}^{-1}$) in the leaf chamber, the single volume analysis works just fine. At higher leak rates, especially combined with lower fan speeds (below 10,000 rpm), then the single volume analysis starts to break down.

In the original 6800-01 and 6800-12 chambers, the incoming and outgoing flow rates were both from **compartment a**. Deriving the mass balance for that configuration leads to a small influence of leaks on the concentrations measured by the sample gas analyzer, leading to a correction implemented in *Bluestem v1.1*. The 6800-01 and 6800-12 chambers have been re-designed (and renamed to 6800-01A

and 6800-12A) to route the incoming flow directly to the chamber, which eliminates the need for a leak correction.

The mass balance equations provided here are assuming a two-compartment system where **compartment a** is the mixing fan volume and **compartment b** is the leaf volume (*Figure C-13* on the previous page). In the diagram and subsequent equation set, c represents CO₂ concentration, w represents H₂O concentration, and μ represents flow rate. Subscripts on flow rates denote the 'to' location followed by the 'from' location. For example, μ_{ab} is the flow to **compartment a** (the subscript a) from **compartment b** (the subscript b).

Derivation of mass-balance equations for chambers

This section describes the computations used for the 6800-01A, 6800-12A, 6800-13, 6800-17, 6800-24 and custom chamber interface.

The mass balance equations describing compartment a

$$\mu_{ab} = \mu_{ba} + \mu_a \quad \text{C-121}$$

and compartment b

$$\mu_0 + \mu_{ba} + sE = \mu_{ab} + \mu_{leak} \quad \text{C-122}$$

can be combined to solve for the sample IRGA flow rate μ_a

$$\mu_a = \mu_0 + sE - \mu_{leak} \quad \text{C-123}$$

H₂O analysis

Behavior of water vapor with respect to time in compartment a

$$V_a \rho \frac{dw_a}{dt} = \mu_{ab} w_b - \mu_{ba} w_a - \mu_a w_a \quad \text{C-124}$$

And compartment b

$$V_b \rho \frac{dw_b}{dt} = \mu_0 w_0 + \mu_{ba} w_a - \mu_{leak} w_b - \mu_{ab} w_b + sE \quad \text{C-125}$$

Assuming steady-state conditions ($dW/dt = 0$), equation C-125 can be solved for W_b :

$$w_b = \frac{sE + \mu_{ba} w_a + \mu_0 w_0}{\mu_{ab} + \mu_{leak}} \quad \text{C-126}$$

The solution for W_b can be substituted into equation C-124 (again, assuming steady-state):

$$\mu_0 w_0 - \mu_{ab} \frac{sE + \mu_{ba} w_a + \mu_{b0} w_0}{\mu_{ab} + \mu_{leak}} - \mu_{ba} w_a = 0 \quad \text{C-127}$$

Since $\mu_{ab} = \mu_{ba} + \mu_{oa}$ (equation C-121), substitute into equation C-127:

$$\mu_0 w_0 - (\mu_{ba} + \mu_a) \frac{sE + \mu_{ba} w_a + \mu_0 w_0}{\mu_{ba} + \mu_a + \mu_{leak}} - \mu_{ba} w_a = 0 \quad \text{C-128}$$

Re-arranging and simplifying equation C-128:

$$w_a = \frac{\mu_0 w_0 + \mu_{ba} w_a + sE}{\mu_{leak} + \mu_{ba} - \mu_a} \quad \text{C-129}$$

Equation C-123 can be used to substituted for μ_a and the equation solved for sE :

$$sE = \frac{\mu_{b0}(w_a - w_0)}{(1 - w_a)} \quad \text{C-130}$$

It can be shown that the concentrations of compartments a and b are equivalent by substituting equation C-121 into equation C-124:

$$(\mu_{ba} + \mu_a) w_b - \mu_{ba} w_a - \mu_a w_a = 0 \quad \text{C-131}$$

Re-arranging and canceling yields:

$$w_a = w_b \quad \text{C-132}$$

CO₂ analysis

Behavior of CO₂ with respect to time in compartment a

$$V_a \rho \frac{dC_a}{dt} = \mu_{ab} C_b - \mu_{ba} C_a - \mu_a C_a \quad \text{C-133}$$

And compartment b

$$V_b \rho \frac{dC_b}{dt} = \mu_0 C_0 + \mu_{ba} C_a - \mu_{leak} C_b - \mu_{ab} C_b - sA \quad \text{C-134}$$

First solve for C_b in equation C-134 assuming steady-state conditions ($dC/dt = 0$)

$$C_b = \frac{\mu_0 C_0 + \mu_{ba} C_a - sA}{\mu_{ab} + \mu_{leak}} \quad \text{C-135}$$

Substitute C_b into equation C-133

$$\mu_{ab} \frac{\mu_0 C_0 + \mu_{ba} C_a - sA}{\mu_{ab} + \mu_{leak}} - \mu_{ba} C_a - \mu_a C_a = 0 \quad \text{C-136}$$

From equation C-121, $\mu_{ab} = \mu_{ba} + \mu_a$, substitute into C-136

$$\left(\mu_{ba} + \mu_a \right) \frac{\mu_0 C_0 + \mu_{ba} C_a - sA}{\mu_{ba} + \mu_a + \mu_{leak}} - \mu_{ba} C_a - \mu_a C_a = 0 \quad \text{C-137}$$

Which simplifies to

$$C_a = \frac{\mu_0 C_0 + \mu_{ba} C_a - sA}{\mu_{ba} + \mu_a + \mu_{leak}} \quad \text{C-138}$$

From equation C-123, $\mu_a = \mu_{a0} + sE - \mu_{leak}$ substitute into C-138

$$C_a = \frac{\mu_0 C_0 + \mu_{ba} C_a - sA}{\mu_{ba} + \mu_{a0} + sE - \mu_{leak} + \mu_{leak}} \quad \text{C-139}$$

Which can be re-arranged to solve for sA

$$sA = \mu_0 (C_0 - C_a) - sEC_a \quad \text{C-140}$$

Combining equation C-133 with equation C-121 yields:

$$(\mu_{ba} + \mu_a) C_b - \mu_{ba} C_a - \mu_a C_a = 0 \quad \text{C-141}$$

which simplifies to:

$$C_a = C_b \quad \text{C-142}$$

Derivation of mass-balance equations for 6800-01 and 6800-12 chambers

This section describes the computations used for the original 6800-01 Fluorometer and 6800-12 clear-top 3×3 cm chamber.

The mass balance equations describing compartment a

$$\mu_0 + \mu_{ab} = \mu_{ba} + \mu_a \quad \text{C-143}$$

and compartment b

$$\mu_{ba} + sE = \mu_{ab} + \mu_{leak} \quad \text{C-144}$$

can be combined to solve for the sample IRGA flow rate μ_a

$$\mu_a = \mu_0 + sE - \mu_{leak} . \quad \text{C-145}$$

H₂O analysis

Behavior of water vapor with respect to time in **compartment a** is

$$V_a \rho \frac{dw_a}{dt} = \mu_0 w_0 + \mu_{ab} w_b - \mu_{ba} w_a - \mu_a w_a \quad \text{C-146}$$

and **compartment b**,

$$V_b \rho \frac{dw_b}{dt} = \mu_{ba} w_a - \mu_{leak} w_b - \mu_{ab} w_b + sE . \quad \text{C-147}$$

Assuming steady-state conditions ($dw/dt = 0$), equation C-147 can be solved for w_b :

$$w_b = \frac{sE + w_a \mu_{ba}}{\mu_{ab} + \mu_{leak}} \quad \text{C-148}$$

The solution for w_b can be substituted into equation C-146 (again assuming steady-state):

$$\mu_0 w_0 + \mu_{ab} \frac{sE + w_a \mu_{ba}}{\mu_{ab} + \mu_{leak}} - \mu_{ba} w_a - \mu_a w_a = 0 \quad \text{C-149}$$

Since $\mu_{ab} = \mu_{ba} + \mu_a - \mu_0$ (equation C-143), substitute into equation C-149

$$\mu_0 w_0 + \left(\mu_{ba} + \mu_a - \mu_0 \right) \frac{sE + w_a \mu_{ba}}{\mu_{ba} + \mu_a - \mu_0 + \mu_{leak}} = \mu_{ba} w_a + \mu_a w_a \quad \text{C-150}$$

$$sE + w_a \mu_{ba} = \left(\mu_{ba} w_a + \mu_a w_a - \mu_0 w_0 \right) \frac{\mu_{ba} + \mu_a - \mu_0 + \mu_{leak}}{\mu_{ba} + \mu_a - \mu_0} \quad \text{C-151}$$

Equation C-145 can be substituted for μ_a :

$$sE + w_a \mu_{ba} = \left(\mu_{ba} w_a + (\mu_0 + sE - \mu_{leak}) w_a - \mu_0 w_0 \right) \frac{\mu_{ba} + (\mu_0 + sE - \mu_{leak}) - \mu_0 + \mu_{leak}}{\mu_{ba} + (\mu_0 + sE - \mu_{leak}) - \mu_0} \quad \text{C-152}$$

$$sE + w_a \mu_{ba} = \left(\mu_{ba} w_a + (\mu_0 + sE - \mu_{leak}) w_a - \mu_0 w_0 \right) \frac{\mu_{ba} + sE}{\mu_{ba} + sE - \mu_{leak}} \quad \text{C-153}$$

K can be defined as

$$K = \frac{\mu_{ba} + sE}{\mu_{ba} + sE - \mu_{leak}} \quad \text{C-154}$$

Since $sE \ll \mu_{ba}, \mu_{leak}$, K can be approximated as

$$K \cong 1 + \frac{\mu_{leak}}{\mu_{ba} - \mu_{leak}} \quad \text{C-155}$$

Substituting K into equation C-153:

$$sE + w_a \mu_{ba} = (\mu_{ba} w_a + (\mu_0 + sE - \mu_{leak}) w_a - \mu_0 w_0) K \quad \text{C-156}$$

$$sE + w_a \mu_{ba} = K \mu_{ba} w_a + K \mu_0 w_a + K sE w_a - K \mu_{leak} w_a - K \mu_0 w_0 \quad \text{C-157}$$

$$sE - K sE w_a = K \mu_{ba} w_a + K \mu_0 w_a - K \mu_{leak} w_a - K \mu_0 w_0 - w_a \mu_{ba} \quad \text{C-158}$$

Re-arranging and solving for sE :

$$sE (1 - K w_a) = K \mu_0 (w_a - w_0) + K w_a (\mu_{ba} - \mu_{leak}) - w_a \mu_{ba}$$

$$sE (1 - K w_a) = K \mu_0 (w_a - w_0) + w_a (K \mu_{ba} - K \mu_{leak} - \mu_{ba}) \quad \text{C-159}$$

$K \mu_{ba} - K \mu_{leak} - \mu_{ba}$ can be eliminated

$$K \mu_{ba} - K \mu_{leak} - \mu_{ba} = \frac{\mu_{ba}}{\mu_{ba} - \mu_{leak}} \mu_{ba} - \frac{\mu_{ba}}{\mu_{ba} - \mu_{leak}} \mu_{leak} - \mu_{ba}$$

$$= \frac{\mu_{ba}}{\mu_{ba} - \mu_{leak}} \mu_{ba} - \frac{\mu_{ba}}{\mu_{ba} - \mu_{leak}} \mu_{leak} - \mu_{ba} \frac{\mu_{ba} - \mu_{leak}}{\mu_{ba} - \mu_{leak}}$$

$$= 0 \quad \text{C-160}$$

Leaving:

$$sE = K \frac{\mu_0 (w_a - w_0)}{(1 - K w_a)} \quad \text{C-161}$$

CO₂ analysis

Behavior of CO₂ with respect to time in compartment a

$$V_a \rho \frac{dc_a}{dt} = \mu_0 c_0 + \mu_{ab} c_b - \mu_{ba} c_a - \mu_a c_a \quad \text{C-162}$$

And compartment b

$$V_b \rho \frac{dc_b}{dt} = \mu_{ba} c_a - \mu_{leak} c_b - \mu_{ab} c_b - sA \quad \text{C-163}$$

First solve for C_b in equation C-163 assuming steady-state conditions ($dC/dt = 0$)

$$C_b = \frac{c_a \mu_{ba} - sA}{\mu_{ab} + \mu_{leak}} \quad \text{C-164}$$

Substitute C_b into equation C-162

$$\mu_0 c_0 + \mu_{ab} \frac{c_a \mu_{ba} - sA}{\mu_{ab} + \mu_{leak}} - \mu_{ba} c_a - \mu_a c_a = 0 \quad \text{C-165}$$

From equation C-143, $\mu_{ab} = \mu_{ba} + \mu_a - \mu_0$, substitute into C-165

$$\mu_0 c_0 + (\mu_{ba} + \mu_a - \mu_0) \frac{c_a \mu_{ba} - sA}{(\mu_{ba} + \mu_a - \mu_0) + \mu_{leak}} - \mu_{ba} c_a - \mu_a c_a = 0 \quad \text{C-166}$$

From equation C-145, $\mu_a = \mu_0 + sE - \mu_{leak}$ substitute into C-166

$$\mu_0 c_0 + (\mu_{ba} + sE - \mu_{leak}) \frac{c_a \mu_{ba} - sA}{\mu_{ba} + sE} - \mu_{ba} c_a - c_a (\mu_0 + sE - \mu_{leak}) = 0 \quad \text{C-167}$$

From equation C-154, $K = \mu_{ba} + sE / \mu_{ba} + sE - \mu_{leak}$ substitute into C-167

$$\mu_0 c_0 + \frac{c_a \mu_{ba} - sA}{K} - \mu_{ba} c_a - c_a (\mu_0 + sE - \mu_{leak}) = 0 \quad \text{C-168}$$

Solving for sA yields

$$sA = K\mu_0 c_0 + c_a \mu_{ba} - K\mu_{ba} c_a - Kc_a (\mu_0 + sE - \mu_{leak})$$

$$sA = K\mu_0 c_0 + c_a \mu_{ba} - K\mu_{ba} c_a - Kc_a \mu_0 - Kc_a sE + Kc_a \mu_{leak}$$

$$sA = K\mu_0 (c_0 - c_a) + c_a \mu_{ba} - K\mu_{ba} c_a - Kc_a sE + Kc_a \mu_{leak}$$

$$sA = K\mu_0 (c_0 - c_a) + c_a (\mu_{ba} - K\mu_{ba} - KsE + K\mu_{leak}) \quad \text{C-169}$$

From equation C-160, $K\mu_{ba} - K\mu_{leak} - \mu_{ba}$ can be eliminated, leaving

$$sA = K\mu_0 (c_0 - c_a) - c_a KsE \quad \text{C-170}$$

$$sA = K(\mu_0 (c_0 - c_a) - c_a sE) \quad \text{C-171}$$

$$= K\mu_0 \left[(c_0 - c_a) - c_a K \frac{w_a - w_0}{1 - Kw_a} \right] \quad \text{C-172}$$

Calculating C_b

The LI-6800 IRGA is measuring the concentration of **compartment a**. However, the concentration in **a** will not be equal to concentration in **compartment b**, where the leaf is located. To compute intercellular CO₂ (C_i), we need to know C_b which can be calculated from combining equations C-143 and C-164

$$C_b = \frac{c_a \mu_{ba} - sA}{\mu_{ba} + \mu_a - \mu_0 + \mu_{leak}} \quad \text{C-173}$$

Then substituting equation C-145 solved for sE

$$C_b = \frac{c_a \mu_{ba} - sA}{\mu_{ba} + sE} \quad \text{C-174}$$

Eliminating the small impact of sE , the resulting equation for C_b is

$$C_b = c_a - \frac{sA}{\mu_{ba}} \quad \text{C-175}$$

Discussion

This section only applies to the early 6800-01 and 6800-12 chambers. All other chambers do not require a leak correction. In a two compartment model, the gas exchange equations become:

$$sE = K \frac{\mu_0(w_a - w_0)}{(1 - Kw_a)} \quad \text{C-176}$$

$$sA = K(\mu_0(c_0 - c_a) - c_a sE) \quad \text{C-177}$$

The difference between the flux equations between the one-compartment derivation and this model is the correction factor K which is $\frac{\mu_{ba}}{\mu_{ba} - \mu_{leak}}$, or the mixing fan flow rate divided by the difference between mixing fan flow rate and leak rate. As $\mu_{ba} \gg \mu_{leak}$, the equation can be simplified to

$$K = 1 + \frac{\mu_{leak} * M}{\mu_{ba} - (\mu_{leak} * M)} \quad \text{C-178}$$

The multiplier M is a chamber-specific multiplier value. The necessity for the multiplier M arises from the different chamber geometry and port sizes of the tubes

between the leaf cuvette and the mixing fan volume. In current chambers, M is 2 and 1.5 for the fluorometer and 3×3 cm chambers, respectively.

The parameter μ_{ba} is also not measured directly by the instrument but must be calculated from the measured parameter fan speed (in RPM, rotations per minute). The amount of flow created by the mixing fan is not linear across the range of rotations per minute applied. Consequently, an empirical fit best describing fan speed vs. air flow was generated.

$$\mu_{ba} = \left(\frac{a+b*Fan_RPM}{1+c*Fan_RPM} \right) \left(\frac{P}{(T+273)} \frac{299}{96} \right) \quad C-179$$

where μ_{ba} is the flow rate created by the fan in $\mu\text{mol s}^{-1}$, Fan_RPM is the measured fan rotation rate, a (-6280), b (6.60), and c (1.71E-05) are empirically determined parameters, T is temperature ($^{\circ}\text{C}$) and P is pressure (kPa).

To test the impact of the correction, experimental data were collected on both fluorometer and 3×3 cm chambers. In the experiment, a chamber was sealed containing a mechanical structure that creates a constant flux. Fluxes were recorded first with no chamber leaks and again with measured controlled leak rates. The % error reported is the difference in measured flux between the leak and no-leak situation. Data is shown prior to any correction (*Figure C-14* on the facing page), after having the correction applied to them (*Figure C-15* on the facing page, B).

Equations C-176 and C-177 can also be used to determine where the impacts from leaks on calculated fluxes can become significant. A quick analysis shows that the correction factor K only becomes significant in circumstances of high leak rates AND low mixing fan speeds. For example, at a relatively fast fan speed of 10,000 RPM, a leak of 350 $\mu\text{mol s}^{-1}$ in the 3×3 cm chamber and 250 $\mu\text{mol s}^{-1}$ in the fluorometer generates a 1% bias. In contrast, a fan speed of 3000 RPM obtains a 1% bias with smaller leaks, 80 $\mu\text{mol s}^{-1}$ in the 3×3 cm chamber and a 60 $\mu\text{mol s}^{-1}$ in the fluorometer.

In summary, the susceptibility of the LI-6800 to chamber leaks will be negligible in most operating conditions. Best measurement practice includes operating at fast fan speeds and preventing significant chamber leaks which can introduce bias into your measurements. Software release 1.1 includes information regarding current leak status as well as the correction factor being applied to assist with avoiding problem areas.

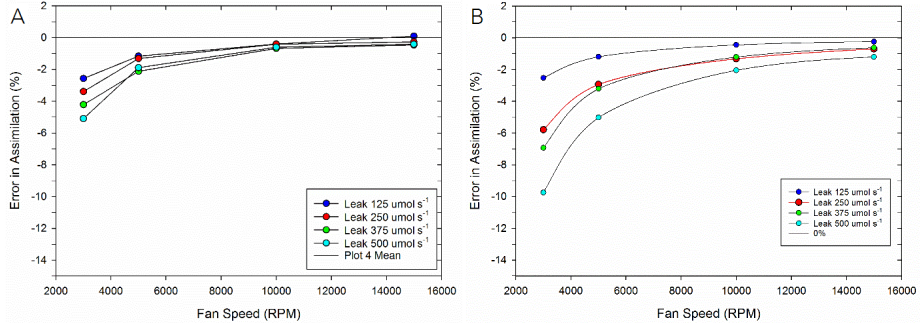


Figure C-14. Uncorrected Data collected from A) the 3 cm \times 3 cm Chamber and B) the Fluorometer.

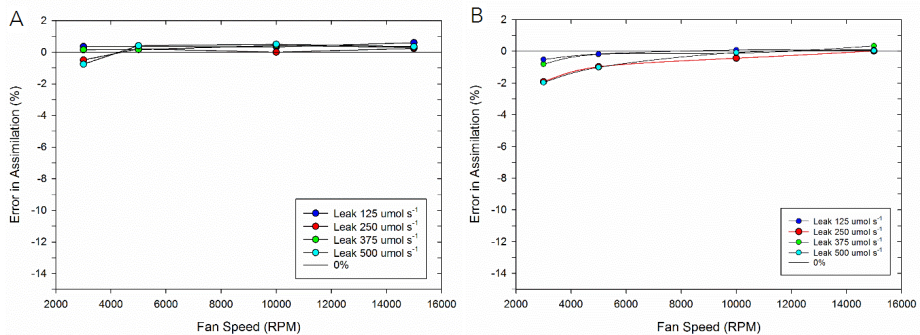


Figure C-15. Corrected Data collected from A) the 3 cm \times 3 cm Chamber and B) the Fluorometer.

References

- 1 Cruz JA, et al. (2005) *Plasticity in light reactions of photosynthesis for energy production and photoprotection*. *J Exp Bot* **56**:395-406.
- 2 Blankenship RE (2002) Carbon metabolism. *Molecular mechanisms of photosynthesis*, ed Press ISU (Blackwell science Ltd), pp 171-203.
- 3 Farquhar GD & Sharkey TD (1982) *Stomatal conductance and photosynthesis*. *Annu Rev Plant Physiol* **33**:317-345.
- 4 von Caemmerer S & Farquhar GD (1981) *Some relationships between the biochemistry of photosynthesis and the gas exchange of leaves*. *Planta* **153**:376-387.

- 5 Moss RA & Loomis WE (1952) *Absorption spectra of leaves. I. The visible spectrum*. *Plant Physiol* **27**:370-391.
- 6 Merchant S & Sawaya MR (2009) *The light reactions: a guide to recent acquisitions for the picture gallery*. *The Plant Cell* **17**:648-663.
- 7 Gomez SM, Nishio JN, Faull KF, & Whitelegge JP (2002) *The chloroplast grana proteome defined by intact mass measurements from liquid chromatography mass spectrometry*. *Mol Cell Proteomics* **1**:46-59.
- 8 Kramer DM, Cruz JA, & Kanazawa A (2003) *Balancing the central roles of the thylakoid proton gradient*. *Trends Plant Sci* **8**:27-32.
- 9 Sacksteder CA, Kanazawa A, Jacoby ME, & Kramer DM (2000) *The proton to electron stoichiometry of steady-state photosynthesis in living plants: A proton-pumping Q cycle is continuously engaged*. *Proc Natl Acad Sci U S A* **97**:14283-14288.
- 10 Witt HT & Zickler A (1978) *Electrical evidence for the field indicating absorbance change in bioenergetic membranes*. *FEBS Letters* **37**:307-310.
- 11 Cruz JA, Sacksteder CA, Kanazawa A, & Kramer DM (2001) *Contribution of electric field to steady-state transthylakoid proton motive force (pmf) in vitro and in vivo. Control of pmf parsing into electric field and pH gradients by ionic strength* *Biochemistry* **40**:1226-1237.
- 12 Jackson P, et al. (2016) *Genetic variation in transpiration efficiency and relationships between whole plant and leaf gas exchange measurements in Saccharum spp. and related germplasm*. *J Exp Bot* **67**:861-871.
- 13 Taiz L & Zeiger E (2006) *Transport and translocation of water and solutes*. *Plant Physiology*, (Sinauer Associates, Inc.), pp 37-124.
- 14 Avenson TJ, Cruz JA, & Kramer DM (2004) *Modulation of energy-dependent quenching of excitons in antennae of higher plants*. *Proc Natl Acad Sci U S A* **101**:5530-5535.
- 15 Kiirats O, Kramer DM, & Edwards GE (2010) *Co-regulation of dark and light reactions in three biochemical subtypes of C(4) species*. *Photosynth Res* **105**:89-99.
- 16 Edwards GE & Baker NR (1993) *Can CO₂ assimilation in maize leaves be predicted accurately from chlorophyll fluorescence analysis?* *Photosynth Res* **37**:89-102.
- 17 Cramer WA, Hasan SS, & Yamashita E (2011) *The Q cycle of cytochrome bc complexes: a structure perspective*. *Biochim Biophys Acta* **1807**:788-802.
- 18 Cramer WA & Zhang H (2006) *Consequences of the structure of the cytochrome b_Lf complex for its charge transfer pathways*. *Biochim Biophys Acta* **1757**:339-345.
- 19 Smirnov AY & Nori F (2012) *Modeling the Q-cycle mechanism of transmembrane energy conversion*. *Phys Biol* **9**:016011.
- 20 Blankenship RE (2002) Chemiosmotic coupling and ATP synthesis. *Molecular mechanisms of photosynthesis*, ed Press ISU), pp 157-170.
- 21 Avenson TJ, Cruz JA, Kanazawa A, & Kramer DM (2005) *Regulating the proton budget of higher plant photosynthesis*. *Proc Natl Acad Sci U S A* **102**:9709-9713.

- 22 Vollmar M, Schlieper D, Winn M, Buchner C, & Groth G (2009) ***Structure of the c14 rotor ring of the proton translocating chloroplast ATP synthase.*** *J Biol Chem* **284**:18228-18235.
- 23 Allen JF (2002) ***Photosynthesis of ATP-electrons, proton pumps, rotors, and poise.*** *Cell* **110**:273-276.
- 24 Livingston AK, Kanazawa A, Cruz JA, & Kramer DM (2010) ***Regulation of cyclic electron flow in C(3) plants: differential effects of limiting photosynthesis at ribulose-1,5-bisphosphate carboxylase/oxygenase and glyceraldehyde-3-phosphate dehydrogenase.*** *Plant, Cell & Env* **33**:1779-1788.
- 25 Earl HJ & Ennahli S (2004) ***Estimating photosynthetic electron transport via chlorophyll fluorometry without Photosystem II light saturation.*** *Photosynth Res* **82**(2):177-186.
- 26 Avenson TJ, et al. (2005) ***Integrating the proton circuit into photosynthesis: progress and challenges.*** *Plant, Cell & Env* **28**:97-109.
- 27 Muller P, Li XP, & Niyogi KK (2001) ***Non-photochemical quenching. A response to excess light energy.*** *Plant Physiol* **125**:1558-1566.
- 28 Niyogi KK (1999) ***PHOTOPROTECTION REVISITED: Genetic and Molecular Approaches.*** *Annu Rev Plant Physiol Plant Mol Biol* **50**:333-359.
- 29 Niyogi KK (2000) ***Safety valves for photosynthesis.*** *Curr Opin Plant Biol* **3**:455-460.
- 30 Kramer DM & Crofts AR (1996) ***Control and measurement of photosynthetic electron transport in vivo.*** *Photosynthesis and the Environment*, ed Baker NR (Kluwer Academic Publishers, The Netherlands), pp 25-66.
- 31 Bjorkman O & Demmig-Adams B (1994) ***Ecophysiology of photosynthesis.*** eds Schulze ED & Caldwell MM (Springer, Berlin), pp 17-47.
- 32 Li XP, et al. (2000) ***A pigment-binding protein essential for regulation of photosynthetic light harvesting.*** *Nature* **403**:391-395.
- 33 Niyogi KK, Li XP, Rosenberg V, & Jung HS (2005) ***Is PsbS the site of non-photochemical quenching in photosynthesis?*** *J Exp Bot* **56**:375-382.
- 34 Walker BJ, Strand DD, Kramer DM, & Cousins AB (2014) ***The response of cyclic electron flow around photosystem I to changes in photorespiration and nitrate assimilation.*** *Plant Physiol* **165**:453-462.
- 35 Kanazawa A & Kramer DM (2002) ***In vivo modulation of nonphotochemical exciton quenching (NPQ) by regulation of the chloroplast ATP synthase.*** *Proc Natl Acad Sci U S A* **99**:12789-12794.
- 36 Flexas J, et al. (2012) ***Mesophyll diffusion conductance to CO₂: an unappreciated central player in photosynthesis.*** *Plant Sci* **193-194**:70-84.
- 37 Galle A, et al. (2009) ***The role of mesophyll conductance during water stress and recovery in tobacco (Nicotiana glauca): acclimation or limitation?*** *J Exp Bot* **60**:2379-2390.

- 38 Niinemets U, Diaz-Espejo A, Flexas J, Galmes J, & Warren CR (2009) *Role of mesophyll diffusion conductance in constraining potential photosynthetic productivity in the field.* *J Exp Bot* **60**:2249-2270.
- 39 Perez-Martin A, et al. (2014) *Regulation of photosynthesis and stomatal and mesophyll conductance under water stress and recovery in olive trees: correlation with gene expression of carbonic anhydrase and aquaporins.* *J Exp Bot* **65**:3143-3156.
- 40 Pons TL, et al. (2009) *Estimating mesophyll conductance to CO₂: Methodology, potential errors, and recommendations.* *J Exp Bot* **60**:2217-2234.
- 41 Zhu X-G, Long SP, & Ort DR (2010) *Improving photosynthetic efficiency for greater yield.* *Annu Rev Plant Biol* **61**:235-261.
- 42 Farquhar GD, von Caemmerer S, & Berry J (1980) *A biochemical model of photosynthetic CO₂ assimilation in leaves of C3 plants.* *Planta* **149**:78-90.
- 43 Long SP & Bernacchi CJ (2003) *Gas exchange measurements, what can they tell us about the underlying limitations to photosynthesis? Procedures and sources of error.* *J Exp Bot* **54**:2393-2401.
- 44 LI-COR, Inc (2008) *LI-6400XT Portable Photosynthesis System Instruction Manual.* LI-COR, Inc, Lincoln, Nebraska, USA.
- 45 Nobel PS (2009) *Physicochemical and environmental plant physiology* (Academic Press Publication, Linacre House, Oxford).
- 46 von Caemmerer S (2000) *Biochemical models of leaf photosynthesis* (CSIRO Publishing, Collingwood, Australia).
- 47 Brooks A & Farquhar GD (1985) *Effect of temperature on the CO₂/O₂ specificity of ribulose-1,5-bisphosphate carboxylase/oxygenase and the rate of respiration in the light.* *Planta* **165**:397-406.
- 48 Schreiber U (2004) *Pulse-amplitude-modulation (PAM) fluorometry and the saturation pulse method: An overview.* *Chlorophyll a fluorescence: a signature of photosynthesis*, eds Papageorgiou GC & Govindjee (The Netherlands), pp 279-319.
- 49 Kramer DM, Johnson GN, Kiirats O, & Edwards GE (2004) *New fluorescence parameters for determination of QA redox state and excitation energy fluxes.* *Photosynth Res* **79**:209-218.
- 50 Krause GH (1991) *Chlorophyll fluorescence and photosynthesis: The basics.* *Annu Rev Plant Physiol Plant Mol Biol* **42**:313-349.
- 51 Schatz GH, Brock H, & Holzwarth AR (1988) *Kinetic and energetic model for the primary processes in photosystem II.* *Biophys J* **54**:397-405.
- 52 Butler WL (1978) *Energy distribution in the photochemical apparatus of photosynthesis.* *Annu Rev Plant Physiol* **29**:345-378.
- 53 Havaux M, Bonfils JP, Lutz C, & Niyogi KK (2000) *Photodamage of the photosynthetic apparatus and its dependence on the leaf developmental stage in the npq1 Arabidopsis mutant deficient in the xanthophyll cycle enzyme violaxanthin de-epoxidase.* *Plant Physiol* **124**:273-284.

- 54 Havaux M & Niyogi KK (1999) *The violaxanthin cycle protects plants from photooxidative damage by more than one mechanism*. *Proc Natl Acad Sci U S A* **96**:8762-8767.
- 55 Lavergne J & Trissl H-W (1995) *Theory of fluorescence induction in photosystem II: derivation of analytical expressions in a model including exciton-radical-pair equilibrium and energy transfer between units*. *Biophys J* **68**:2474-2492.
- 56 Loriaux SD, et al. (2013) *Closing in on maximum yield of chlorophyll fluorescence using a single multiphase flash of sub-saturating intensity*. *Plant Cell Environ* **36**:1755-1770.
- 57 Maxwell K & Johnson GN (2000) *Chlorophyll fluorescence: a practical guide*. *J Exp Bot* **51**:659-668.
- 58 van Kooten O & Snell JFH (1990) *The use of chlorophyll fluorescence nomenclature in plant stress physiology*. *Photosynth Res* **25**:147-150.
- 59 Tyystjarvi E & Aro E-M (1996) *The rate constant of photoinhibition, measured in lincromycin-treated leaves, is directly proportional to light intensity*. *Proc Natl Acad Sci U S A* **93**:2213-2218.
- 60 Valladares F & Pearcy RW (2005) *Drought can be more critical in the shade than the sun: a field study of carbon gain and photoinhibition in a California shrub during a dry El nino year*. *Plant, Cell & Env* **25**:1-10.
- 61 Markgraf T & Berry J (1990) *Measurement of photochemical and non-photochemical quenching: correction for turnover of PS2 during steady-state photosynthesis*. *Current Research in Photosynthesis*, ed Baltscheffsky M (Kluwer Academic Press, The Netherlands), Vol IV, pp 279-282.
- 62 Depege N, Bellafiore S, & Rochaix J-D (2003) *Role of chloroplast protein kinase Stt7 in LHClI phosphorylation and state transition in Chlamydomonas*. *Science* **299**:1572-1575.
- 63 Ahn TK, et al. (2009) *Investigating energy partitioning during photosynthesis using an expanded quantum yield convention*. *Chem Phys* **357**:151-158.
- 64 Kulheim C, Agren J, & Jansson S (2002) *Rapid regulation of light harvesting and plant fitness in the field*. *Science* **297**:91-93.
- 65 Genty BE, Briantas JM, & Baker NR (1989) *The relationship between the quantum yield of photosynthetic electron transport and photochemical quenching of chlorophyll fluorescence*. *Biochim Biophys Acta* **990**:87-92.
- 66 Flexas J, Ribas-Carbo M, Diaz-Espejo A, Galmes J, & Medrano H (2008) *Mesophyll conductance to CO₂: current knowledge and future prospects*. *Plant, Cell & Env* **31**:602-621.
- 67 Harley PC, Loreto F, DiMarco G, & Sharkey TD (1992) *Theoretical considerations when estimating mesophyll conductance to CO₂ flux by analyses of the response of photosynthesis to CO₂*. *Plant Physiol* **98**:1429-1436.

Appendix D.

Specifications

The following list of specifications¹ describes the performance characteristics of the LI-6800 and its accessories.

CO₂ gas analyzer

Type: Absolute non-dispersive infrared gas analyzer

Measurement range: 0 – 3100 $\mu\text{mol mol}^{-1}$

Precision (signal noise) at 400 $\mu\text{mol mol}^{-1}$

RMS with 4-second signal averaging: $\leq 0.1 \mu\text{mol mol}^{-1}$

Accuracy: Within 1% of reading at 200 $\mu\text{mol mol}^{-1}$ or above, $\pm 2 \mu\text{mol mol}^{-1}$ at $< 200 \mu\text{mol mol}^{-1}$

Orientation sensitivity: $\leq \pm 1 \mu\text{mol mol}^{-1}$ variation at 400 $\mu\text{mol mol}^{-1}$ from any orientation

H₂O gas analyzer

Type: Absolute non-dispersive infrared gas analyzer

Measurement range: 0 – 75 mmol mol^{-1}

Precision (signal noise) at 10 mmol mol^{-1}

RMS with 4-second signal averaging: $\leq 0.01 \text{mmol mol}^{-1}$

Accuracy: within 1.5% of reading at $> 5 \text{mmol mol}^{-1}$; $\pm 0.08 \text{mmol mol}^{-1}$ at $< 5 \text{mmol mol}^{-1}$

Temperatures

Operating temperature range: 0 – 50 °C

Storage temperature range: -20 °C – 60 °C

Temperature control range

Leaf temperature: ± 10 °C from ambient with 3 cm \times 3 cm chamber

¹Specifications subject to change without notice.

Setpoint resolution: 0.1 °C

Chamber exhaust air temperature and temperature control block

Type: Thermistor

Range: -10 – 60 °C

Accuracy: ±0.15 °C

Leaf temperature sensor

Type: Type E fine-wire thermocouple

Sensitivity range: -10 – 60 °C

Accuracy: <±0.5 °C total; ±0.2 °C cold junction reference; ±0.3 °C thermocouple when within ±10 °C of cold junction temperature

Air flow rates

Bulk flow rate range: 680 – 1700 $\mu\text{mol s}^{-1}$ at SATP¹

Leaf chamber flow rate range: 0 – 1400 $\mu\text{mol s}^{-1}$ at SATP

Pressure

Console pressure sensor

Operating range: 50 – 110 kPa

Accuracy: ±0.4 kPa

Resolution: 1.5 Pa typical

Signal noise: ≤ 0.004 kPa peak-to-peak with 4-second signal averaging

Chamber pressure sensor

Range: -2 – 2 kPa

Resolution: < 1 Pa typical

Signal noise: 1 Pa peak-to-peak with 4-second signal averaging

Setpoint resolution: 1.0 Pa

Control range: 0 – 200 Pa (dependent on flow rate through the chamber)

Communication

RJ-45 Ethernet; IP/TCP for networks and computers: 1

Head connections: 2

Accessory connections: 2

¹Standard Ambient Temperature (25 °C) and Pressure (100 kPa).

Batteries

Weight: 0.435 kg

Capacity: 6800 mAh

Type: Lithium ion

Storage: -20 – 60 °C; ≤ 80% RH

CO₂ control

CO₂ control range: 0 – > 2000 μmol mol⁻¹ (with pump set to low; dependent on bulk flow rate)

Sources:

8 gram CO₂ cartridge

External tank with optional adapter kit

CO₂ cartridge lifetime: > 8 hours after puncture (dependent on setpoint)

CO₂ scrubber: Soda lime

H₂O control

H₂O control range: 0 – 90% RH (noncondensing)

Humidifier substrate: Pall Stuttgarter Masse ceramic substrate (Pall Corporation)

Desiccant: Sorbead[®] Orange CHAMELEON[®] silica gel beads

Light measurement

Chamber and light source PAR sensors:

Detector: Gallium arsenide

Sensitivity range: 0 – 3000 μmol m⁻² s⁻¹

Resolution: < 1 μmol mol⁻¹

Calibration accuracy: ±5% of reading; traceable to the U.S. National Institute of Technology (NIST)

External LI-190R PAR Sensor:

Detector: Silicon photodiode

Sensitivity: 5 – 10 μA per 1000 μmol s⁻¹ m⁻²

Calibration accuracy: ±5% of reading; traceable to NIST

Console

Processor: 800 MHz ARM[®] Cortex[™] A8

Memory: 512 MB RAM; 8 GB Flash memory

Display: Sunlight-readable TFT LCD with capacitive touch screen

Resolution: 1024 × 600 pixels

Dimensions: 26 cm diagonally

Size: 18.5 cm × 27.5 cm × 21 cm; (D × W × H)

Weight: 6.1 kg

Power requirements: 12 to 18 VDC or 24 VDC

Head

Size with 3×3 cm chamber: 37 cm × 11.5 cm × 21.6 cm (L × W × H)

Weight: 2.15 kg without chamber

Display resolution: 128 × 128 pixels

Display dimensions: 3.15 cm diagonally

Sensor head inputs:

Leaf temperature thermocouple: 2

LI-190R light sensor: 1

Sensor head light source connections: 1

Chambers, Accessories, and Light Sources

The following components may be ordered separately or as a package with the LI-6800.

Clear-top chamber (3 cm × 3 cm)

Aperture areas: 9 cm² (3 cm × 3 cm), 6 cm² (2 cm × 3 cm), 3 cm² (1 cm × 3 cm)

Size: 15.4 cm × 11.5 cm × 5.9 cm (L × W × H)

Weight: 0.3 kg

Large leaf and needle chamber (6 cm × 6 cm)

Maximum leaf area: 36 cm²

External dimensions: 11.5 cm wide (spring to spring) × 16.8 cm long (front of chamber to back of chamber link) × 5.9 cm tall (bottom of chamber manifold to top of chamber)

Weight: 0.35 kg

Large leaf and needle chamber with conifer sprig blocks

Chamber Volume: 420.8 cm³ (internal)

Maximum planar leaf area: 36 cm²

Internal chamber height: 6.7 cm

External dimensions: 11.5 cm wide (spring to spring) × 16.8 cm long (front of chamber to back of chamber link) × 7.2 cm tall (bottom to top of chamber)

Small plant chamber

Chamber volume: 193.2 cm³ (internal)

Internal dimensions: 7 cm diameter; 4.46 cm deep

External dimensions: 8.4 cm wide (chamber block) × 12.7 cm long (chamber manifold interface to tip of chamber) × 6.47 cm (bottom of chamber manifold to top of chamber)

Weight: 0.60 kg

Bryophyte chamber

Chamber volume: 193.2 cm³ (internal)

Internal dimensions: 7 cm diameter; 4.45 cm deep

External dimensions: 8.4 cm wide (chamber block) × 12.7 cm long (chamber manifold interface to tip of chamber) × 6.47 cm (bottom of chamber manifold to top of chamber)

Weight: 0.60 kg

Insect respiration chamber

Chamber volume: 49.9 cm³ (not including tubing)

Exterior dimensions: 11.25 cm length; 3 cm diameter

Weight: 0.07 kg

Soil CO₂ flux chamber

Chamber volume: 4244 cm³

IRGA volume: 57 cm³

Soil area: 317.8 cm²

Air temperature thermistor:

Operating range: -20 to 45 °C

Accuracy: ±0.5 °C over 0 °C to 70 °C

Custom chamber interface

Chamber volume: 34.2 cm³ (internal volume)

External Dimensions: 1.25 cm × 7.67 cm × 5.85 cm (L × W × H)

3 cm × 3 cm light source

Total output range: 0 – >2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Blue output range: 0 – > 400 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Red output range: 0 – > 1600 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Red peak wavelength: 660 nm

Blue peak wavelength: 453 nm

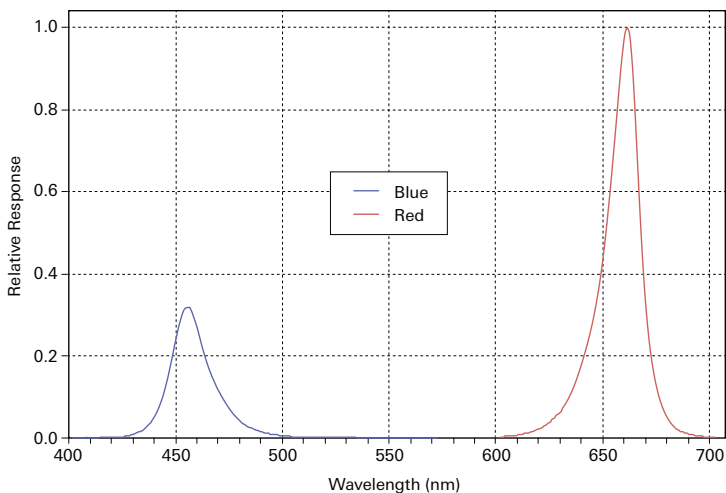


Figure D-1. Typical spectral output of the LEDs in the 3 cm × 3 cm light source.

Uniformity:

±10% over 90% of the aperture with white top gasket, typically

±10% over 77% of the aperture with black gasket, typically

Power consumption at 2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$: < 5 W

Operating temperature range: 0 – 50 °C

Size: 6.6 cm × 5.9 cm × 5.8 cm (L × W × H)

Weight: 0.21 kg

Multiphase Flash™ Fluorometer

Modulated light: Software controlled, selectable frequencies of 1 Hz – 250 kHz

Measuring light peak wavelength: 625 nm

Blue actinic peak wavelength: 475 nm

Red actinic and saturating flask peak wavelength: 625 nm

Far-red peak wavelength: 735 nm

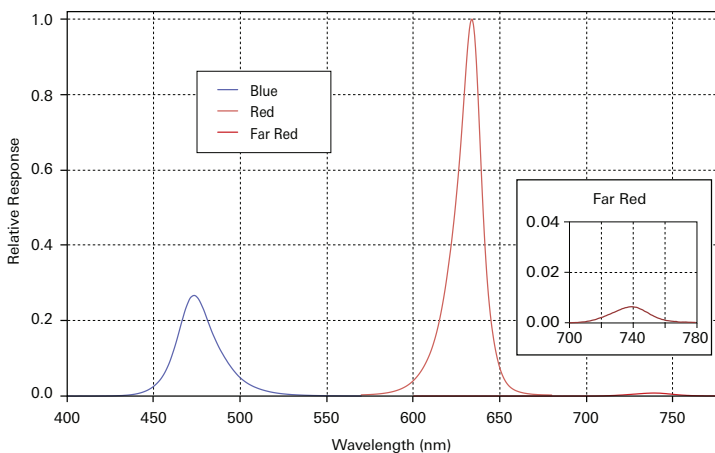


Figure D-2. Typical spectral output of the blue, red, and far red LEDs in the fluorometer.

Actinic light output:

0 – 3000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ total at 25 °C

0 – 1000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ blue at 25 °C

0 – 2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ red at 25 °C

Saturation light: Software controlled intensity; 0 – 16,000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Far-red light: Software controlled intensity; 0 – 20 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Fluorescence signal temperature dependence: -0.25% per °C

Uniformity:

<±10% over 92% of the aperture with white top gasket

<±10% over 90% of the aperture with black top gasket

Power consumption:

< 18 W at 25 °C with 3000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ actinic light

< 60 W at 25 °C with 16,000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ saturating flash

Leaf area: 6 cm^2 or 2 cm^2 ; Round apertures

Dimensions: 16.6 cm × 11.5 cm × 13.6 cm (L × W × H)

Weight: 0.86 kg

Large light source

Total output range: 0 – >2500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Blue output range: >2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Green output range: >1000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Red output range: >2400 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

White output range: >1500 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at 25 °C

Blue peak wavelength: 453 nm

Green peak wavelength: 523 nm

Red peak wavelength: 660 nm

White color temperature: 4000 K

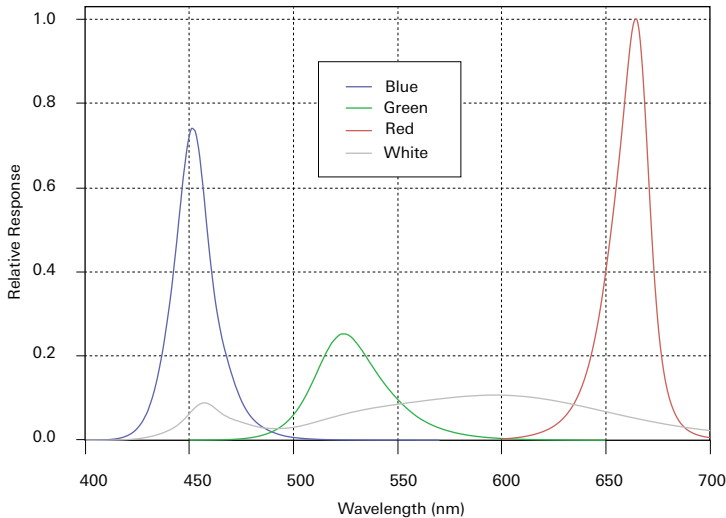


Figure D-3. Typical spectral output of the blue, green, red, and white LEDs in the 6 cm × 6 cm large light source.

Uniformity: ±10% over 90% of the aperture

Power consumption: 15 W at 2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ equal parts red, green, blue, and white

Operating temperature range: 0 – 50 °C

Operating relative humidity range: 0 – 85%

Size: 11.7 cm × 11 cm × 13 cm (L × W × H)

Weight: 0.54 kg

Appendix E.

Warranty

Each LI-COR, Inc. instrument is warranted by LI-COR, Inc. to be free from defects in material and workmanship; however, LI-COR, Inc.'s sole obligation under this warranty shall be to repair or replace any part of the instrument which LI-COR, Inc.'s examination discloses to have been defective in material or workmanship without charge and only under the following conditions, which are:

- 1** The defects are called to the attention of LI-COR, Inc. in Lincoln, Nebraska, in writing within one year after the shipping date of the instrument.
- 2** The instrument has not been maintained, repaired or altered by anyone who was not approved by LI-COR, Inc.
- 3** The instrument was used in the normal, proper and ordinary manner and has not been abused, altered, misused, neglected, involved in an accident or damaged by act of God or other casualty.
- 4** The purchaser, whether it is a DISTRIBUTOR or direct customer of LI-COR or a DISTRIBUTOR'S customer, packs and ships or delivers the instrument to LI-COR, Inc. at LI-COR Inc.'s factory in Lincoln, Nebraska, U.S.A. within 30 days after LI-COR, Inc. has received written notice of the defect. Unless other arrangements have been made in writing, transportation to LI-COR, Inc. (by air unless otherwise authorized by LI-COR, Inc.) is at customer expense.
- 5** No-charge repair parts may be sent at LI-COR, Inc.'s sole discretion to the purchaser for installation by purchaser.
- 6** LI-COR, Inc.'s liability is limited to repair or replace any part of the instrument without charge if LI-COR, Inc.'s examination disclosed that part to have been defective in material or workmanship.

There are no warranties, express or implied, including but not limited to any implied warranty of merchantability of fitness for a particular purpose on underwater cables or on expendables such as batteries, lamps, thermocouples, and calibrations.

Other than the obligation of LI-COR, Inc. expressly set forth herein, LI-COR, Inc. disclaims all warranties of merchantability or fitness for a particular purpose. The foregoing constitutes LI-COR, Inc.'s sole obligation and liability with respect to damages resulting from the use or performance of the instrument and in no event shall LI-COR, Inc. or its representatives be liable for damages beyond the price paid for the instrument, or for direct, incidental or consequential damages.

The laws of some locations may not allow the exclusion or limitation on implied warranties or on incidental or consequential damaged, so the limitations herein may not apply directly. This warranty gives you specific legal rights, and you may already have other rights which vary from state to state. All warranties that apply, whether included by this contract or by law, are limited to the time period of this warranty which is a twelve-month period commencing from the date the instrument is shipped to a user who is a customer or eighteen months from the date of shipment to LI-COR, Inc.'s authorized distributor, whichever is earlier.

This warranty supersedes all warranties for products purchased prior to June 1, 1984, unless this warranty is later superseded. To the extent not superseded by the terms of any extended warranty, the terms and conditions of LI-COR's Warranty still apply.

DISTRIBUTOR or the DISTRIBUTOR's customers may ship the instruments directly to LI-COR if they are unable to repair the instrument themselves even though the DISTRIBUTOR has been approved for making such repairs and has agreed with the customer to make such repairs as covered by this limited warranty.

Further information concerning this warranty may be obtained by writing or telephoning Warranty manager at LI-COR, Inc.

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