Gas exchange measurements

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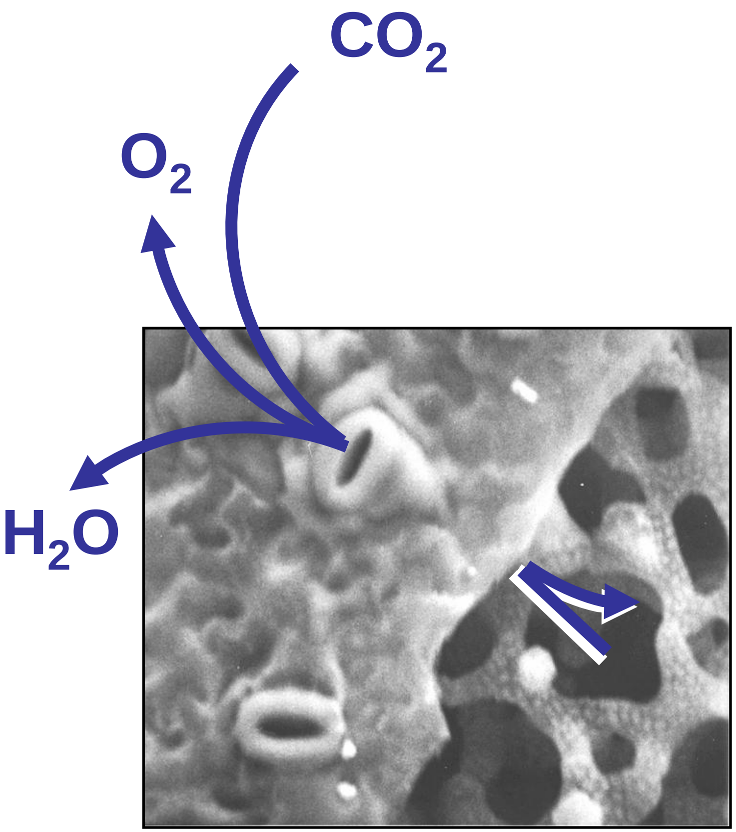
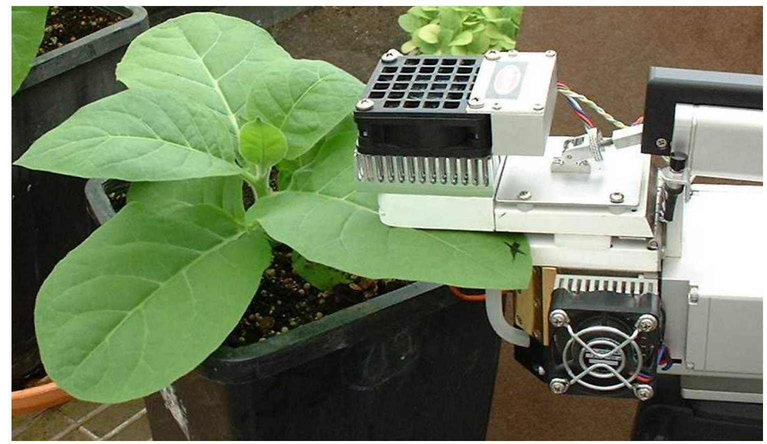
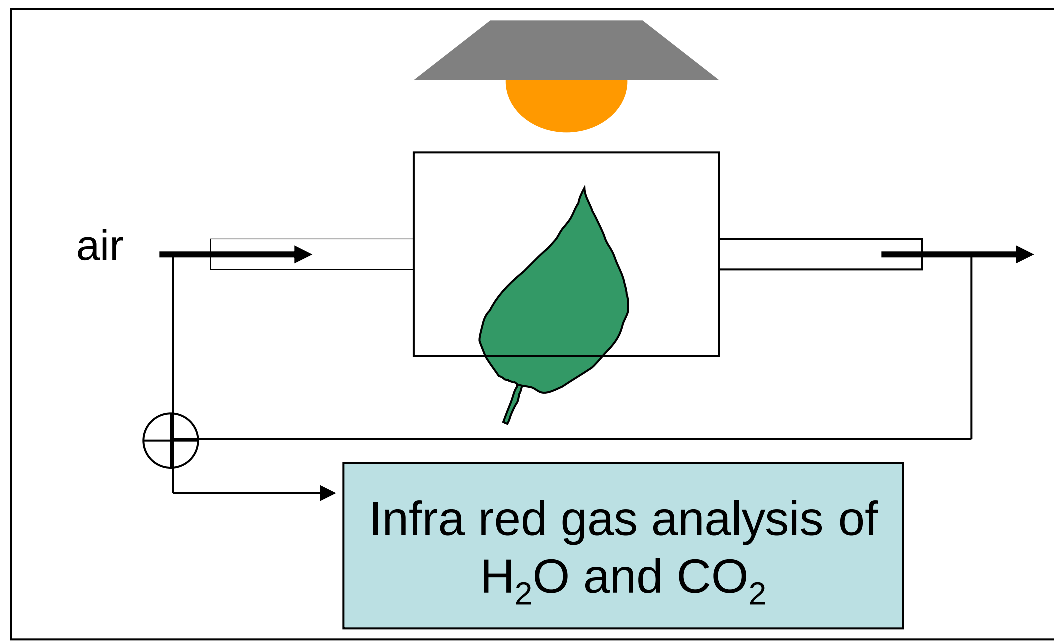






10.1071/FP10900_AC

Supplementary Material: *Functional Plant Biology*, 2014, 41(3), 223-226.

http://www.licor.com/env/Products/li6400/6400_manuals.jsp

Transpiration

The mass balance of water vapor in an open system (Figure 1-4) is given by

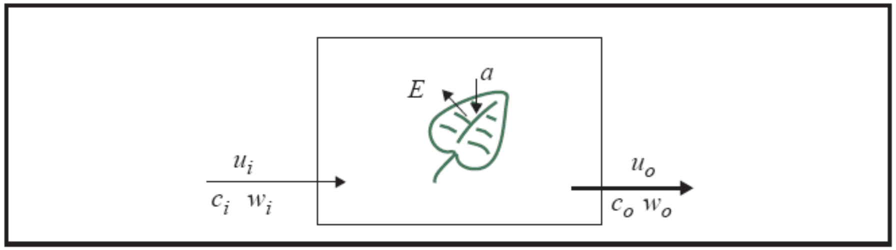


Figure 1-4. Measuring fluxes in an open system. Transpiration rate (E) and photosynthetic rate (a) change the water and CO_2 concentrations of air as it passes through the chamber. Transpiration also causes the exit flow u_o to be greater than the incoming flow rate (u_i).

$$sE = u_o w_o - u_i w_i \tag{1-1}$$

where s is leaf area (m^{-2}), E is transpiration rate (mol m^{-2} s^{-1}), u_i and u_o are incoming and outgoing flow rates (mol s^{-1}) from the chamber, and w_i and w_o are incoming and outgoing water mole fractions (mol H_2O mol air^{-1}). Since

$$u_o = u_i + sE \tag{1-2}$$

we can write

$$sE = (u_i + sE)w_o - u_i w_i \tag{1-3}$$

which rearranges to

$$E = \frac{u_i(w_o - w_i)}{s(1 - w_o)} \tag{1-4}$$

Sections of this handout are copied from 'Using the LI-6400' which can be downloaded from the link above. The manuals contain detailed information and helpful pictures. This handout is aimed at providing a quick guide to some of the basic information to get you started.

Page 1-7

[1] S.von Caemmerer and G.D.Farquhar (1981) Some relationships between the biochemistry of photosynthesis and the gas exchange of leaves, *Planta* 153:376-

Knowing E, we can calculate conductance, g

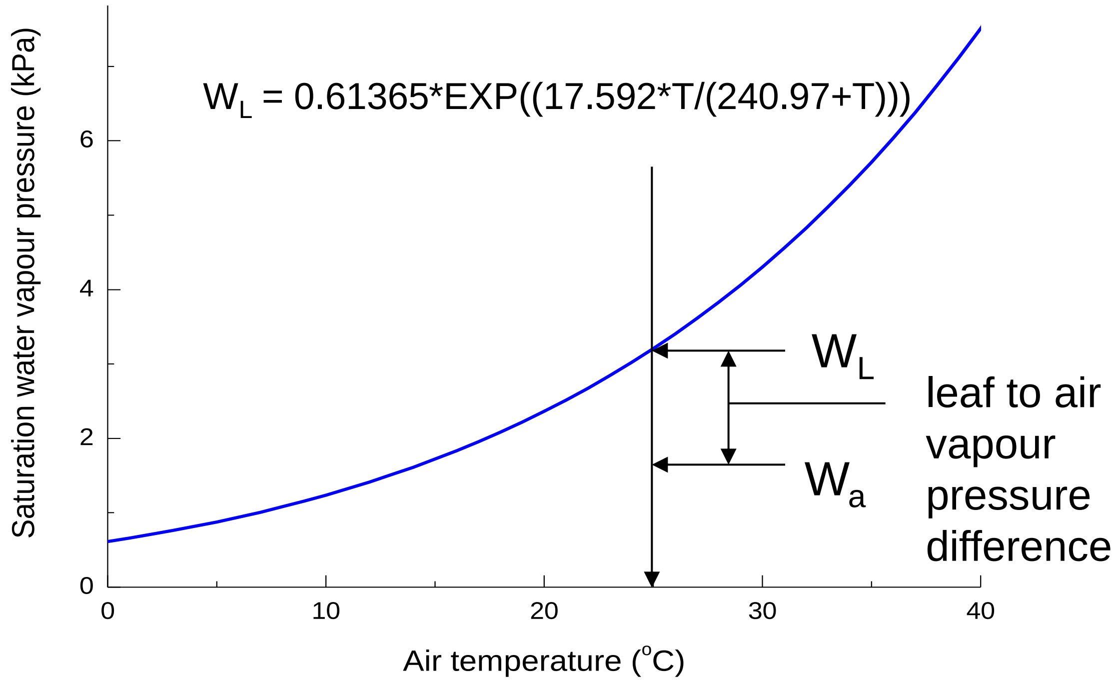
$$E = g\,(W_L - W_a)$$

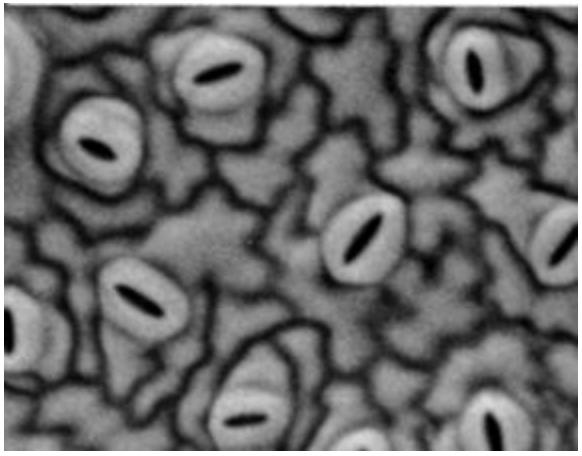
$$g = E / (W_L - W_a)$$

E	Transpiration rate
g	conductance to water
W_l	saturation vapour pressure inside leaf
W_a	vapour pressure in air

Conductance is the ease with which gases can exchange between two places. Exchange occurs through stomatal pores in the epidermis. Plants control water loss by varying stomatal apertures.

Function relating saturation vapour pressure to temperature





Net Photosynthesis

The mass balance of CO_2 in an open system is given by

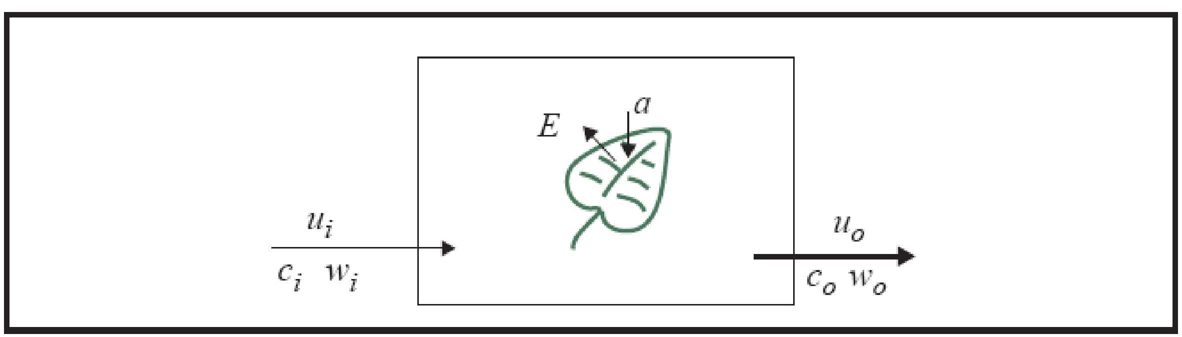
$$sa = u_i c_i - u_o c_o \qquad (1\text{-}11)$$

where a is assimilation rate (mol CO_2 m^{-2} s^{-1}), c_i and c_o are incoming and outgoing mole fractions (mol CO_2 mol air^{-1}) of carbon dioxide. Using (1-2), we can write

$$sa = u_i c_i - (u_i + sE)c_o \qquad (1\text{-}12)$$

which rearranges to

$$a = \frac{u_i(c_i - c_o)}{s} - Ec_o \qquad (1\text{-}13)$$



Using Fick's Law, we can relate the rate of CO_2 assimilation to conductance and calculate the intercellular CO_2 mole fraction

$$A = g/1.6\ (C_a - C_i)$$

$$C_i = C_a - 1.6\ A/g$$

A	rate of CO_2 assimilation
g	conductance to water
C_a	ambient CO_2 mole fraction
C_i	intercellular CO_2 mole fraction

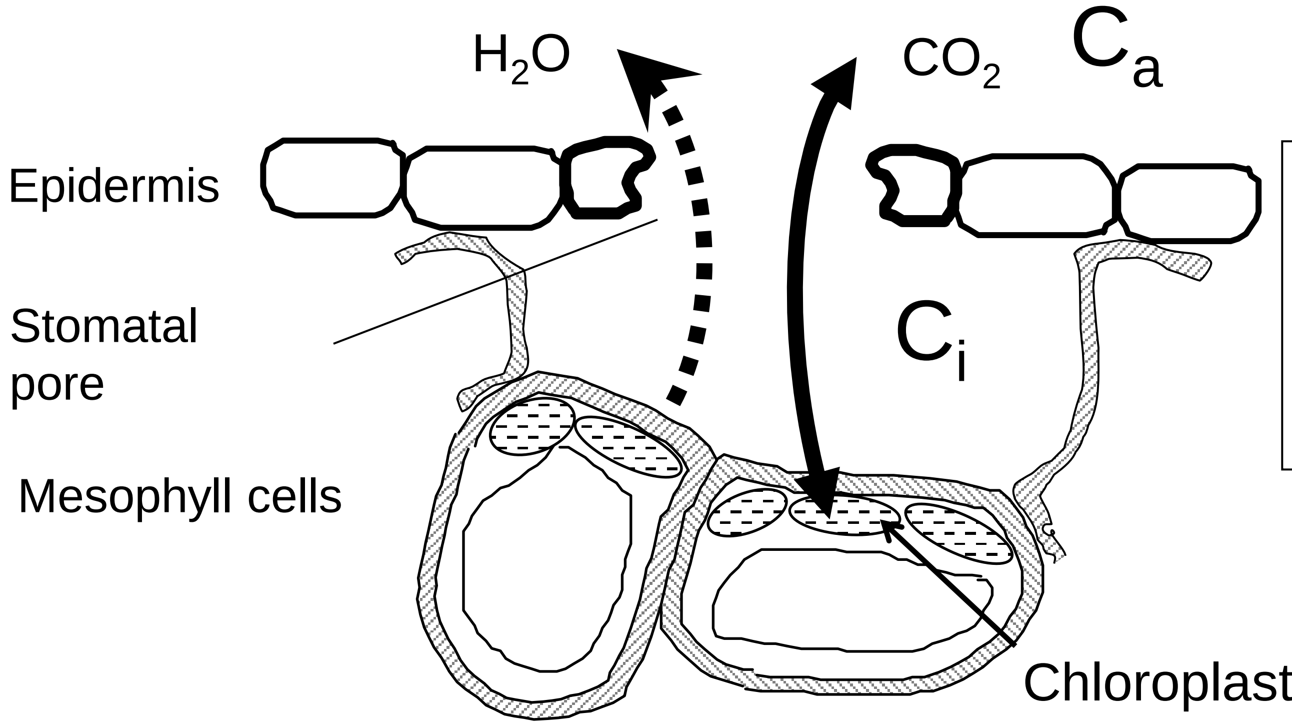
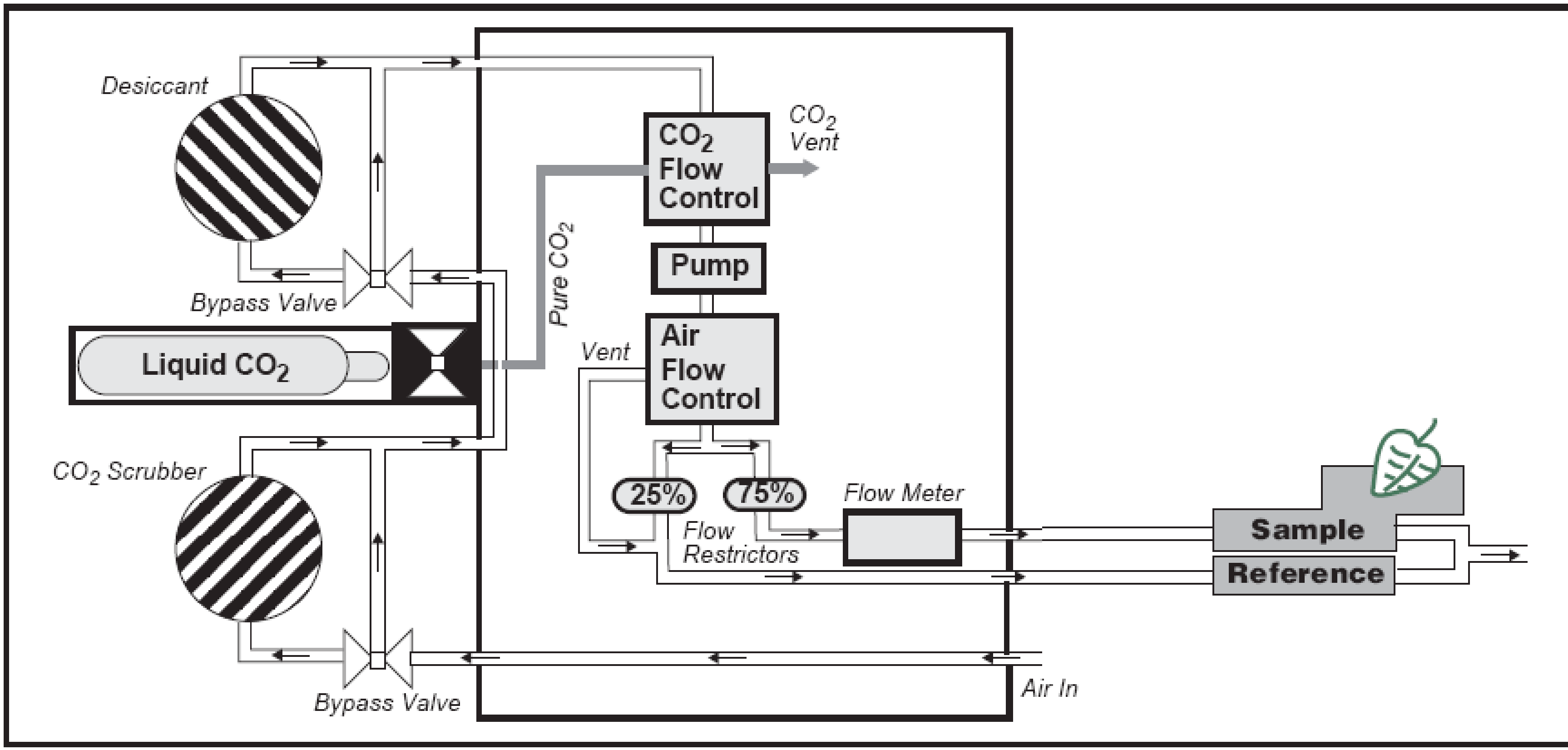


Diagram of part of a leaf viewed in cross section

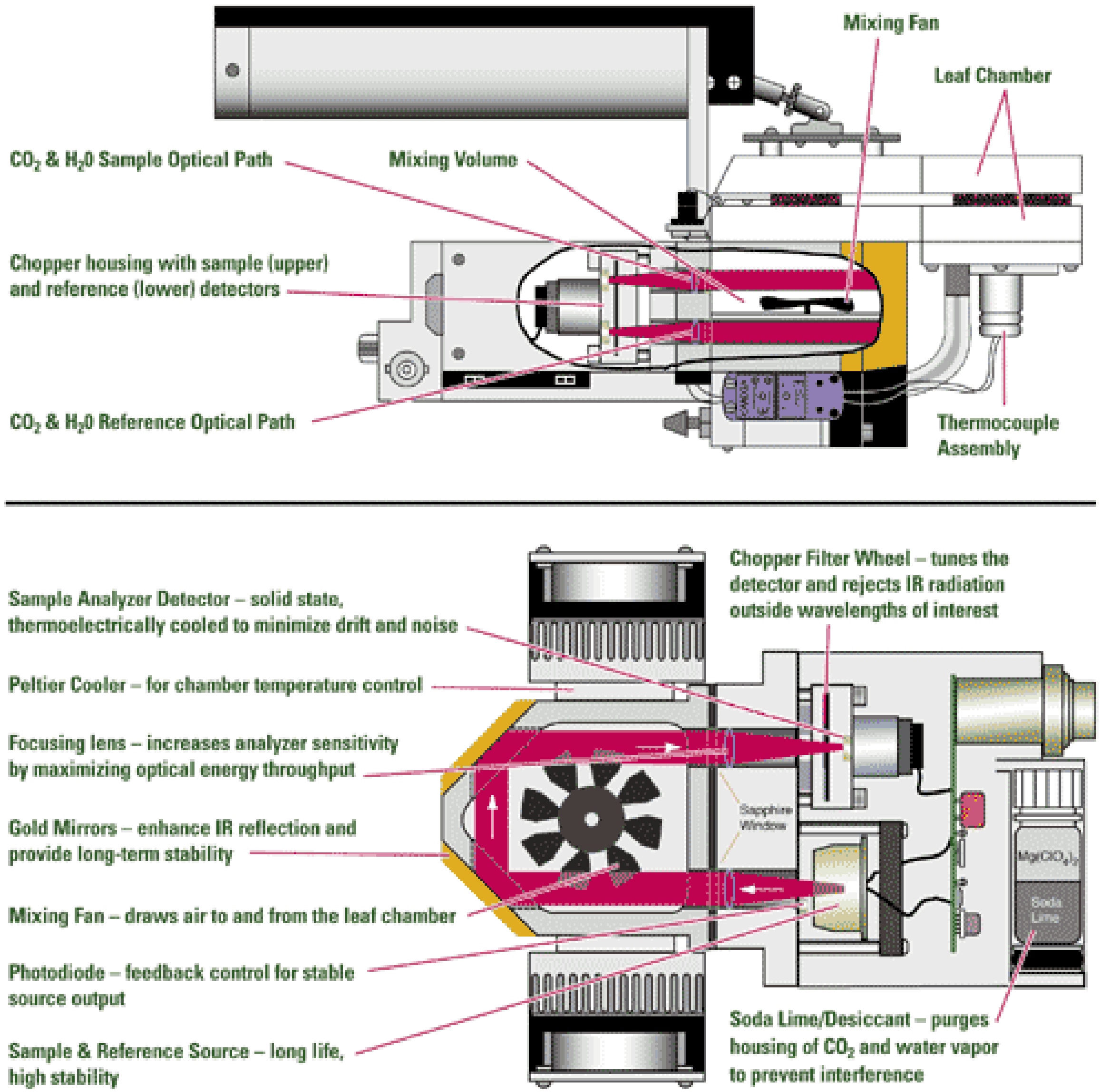
Licor 6400 gas flow diagram

Page 1-5

Schematic with a 6400-01 CO2 Mixer



Cross section through the LI6400 head showing the optical path of the two infra red gas analyzers



To reduce errors, sample cell gas is passed through the reference cell and the outputs from the two IRGAs are set to the same value. This is called matching. (f5 level 1 in New Measurements mode)

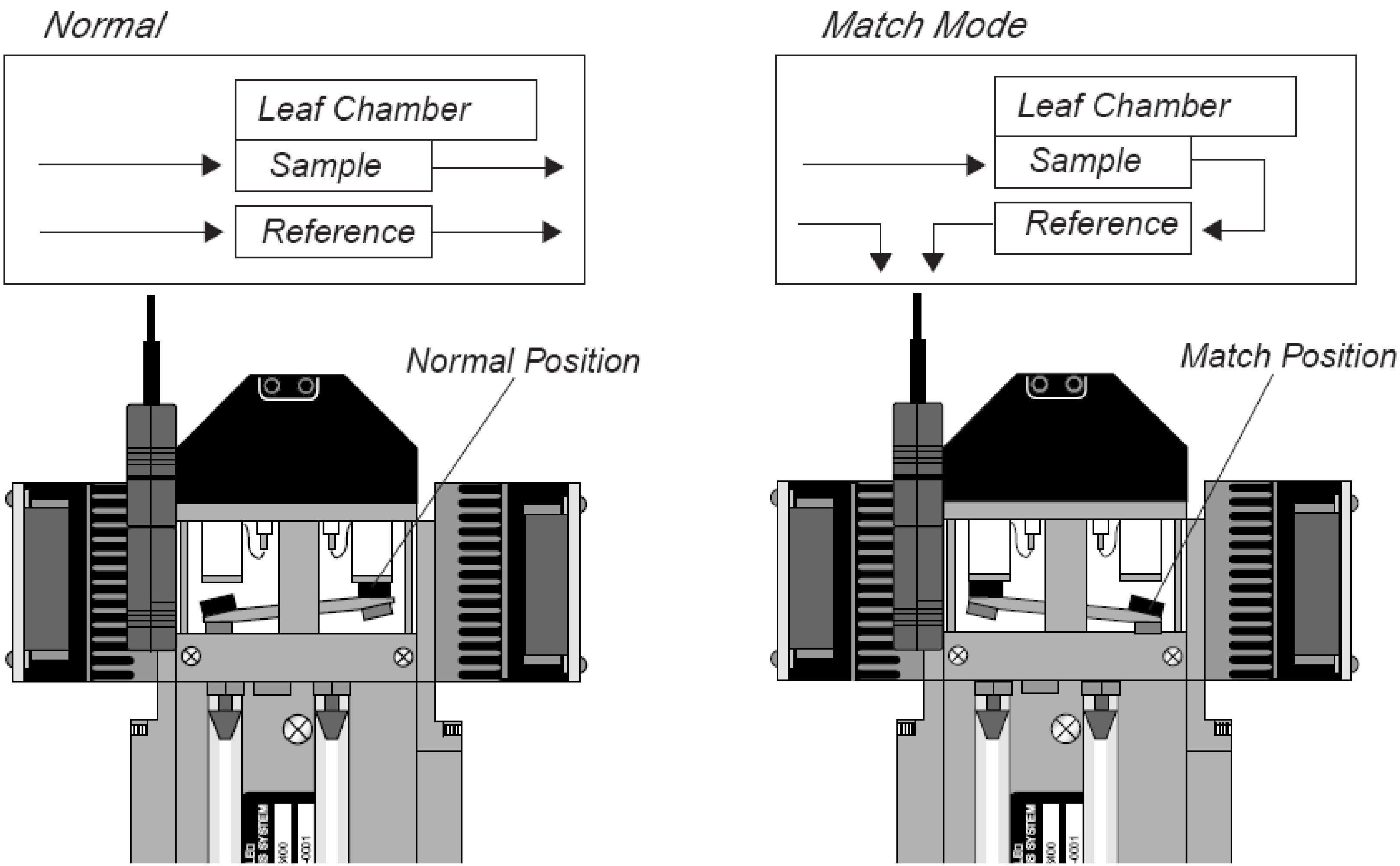
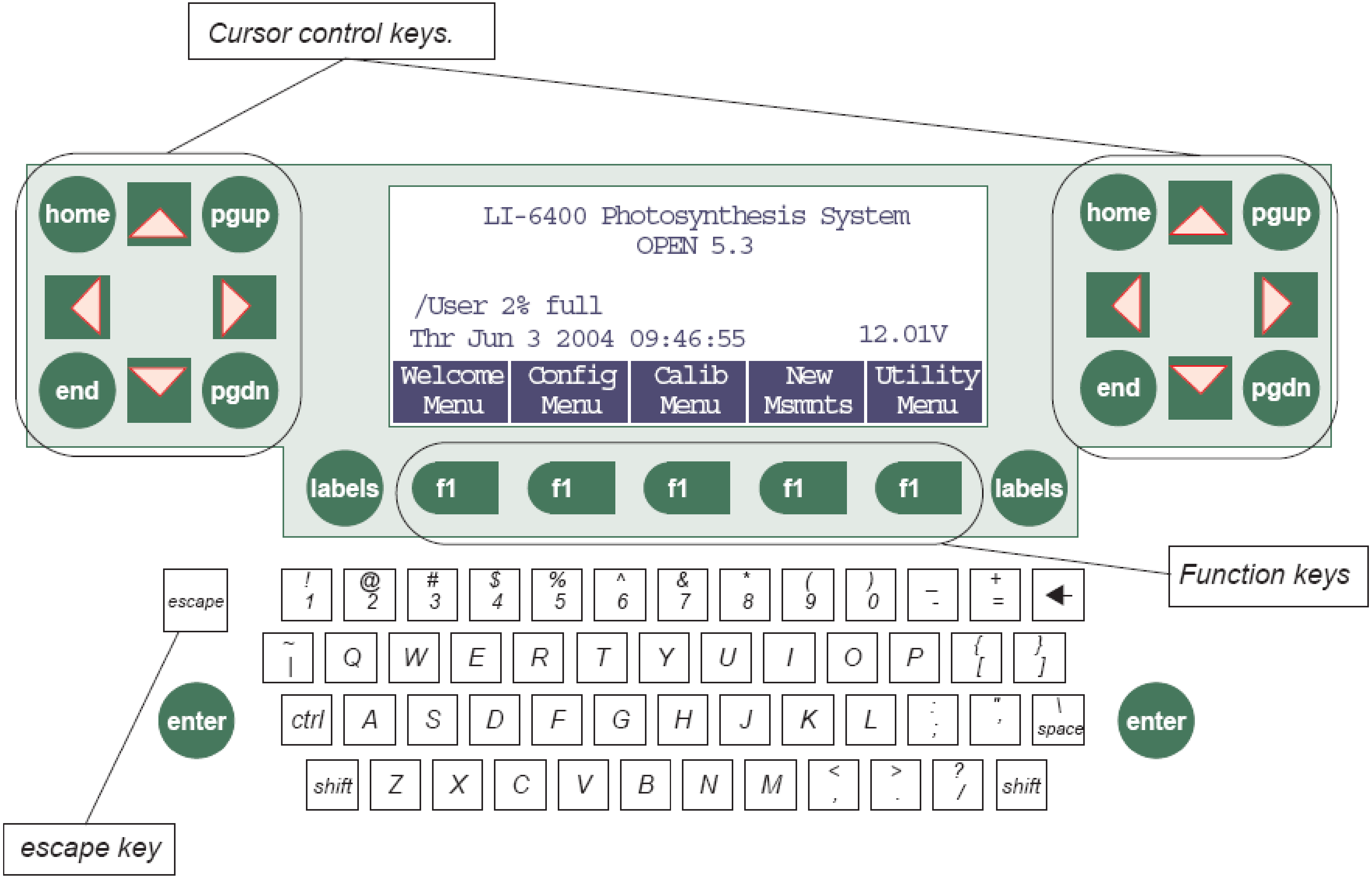
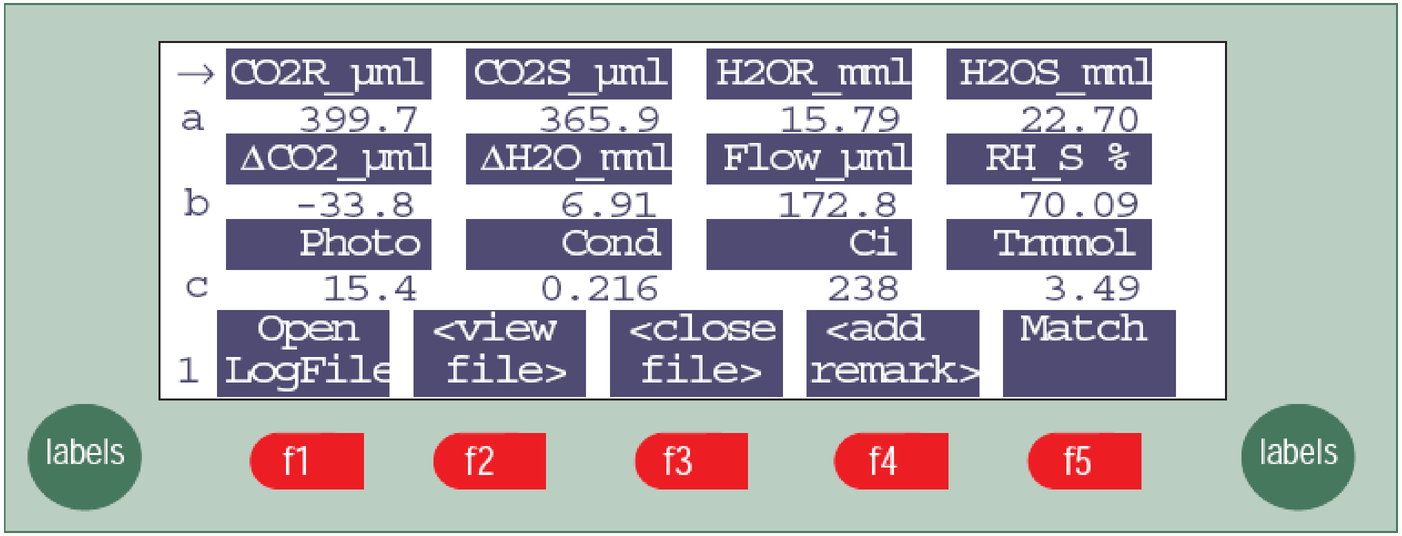


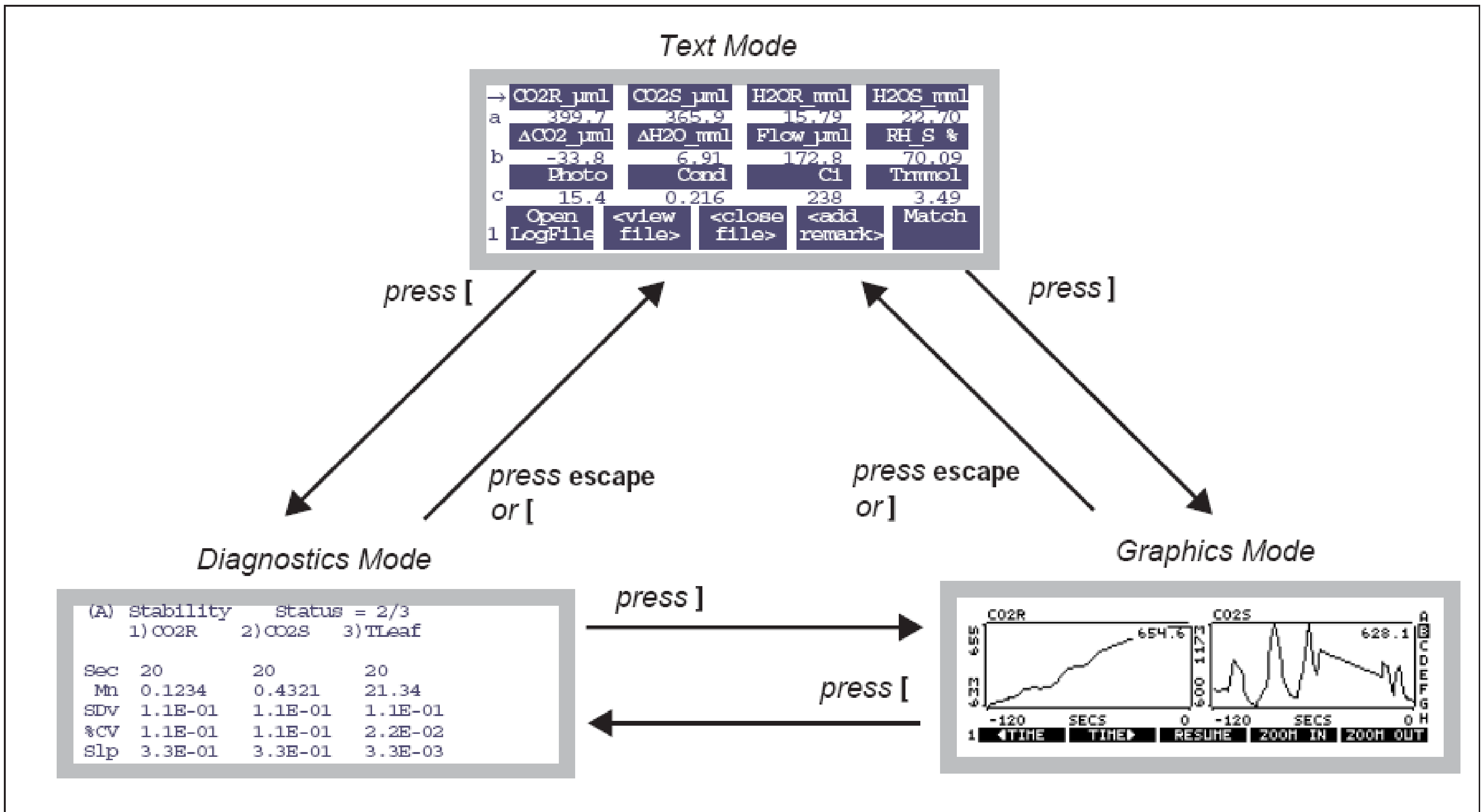
Figure 4-2. The match valve puts exhaust air from the sample cell into the reference cell, allowing both cells to be matched without altering conditions in the leaf chamber.


Cursor control keys.
home
pgup
end
pgdn
LI-6400 Photosynthesis System
OPEN 5.3
/User 2% full
Thr Jun 3 2004 09:46:55
12.01V
Welcome Menu
Config Menu
Calib Menu
New Msmnts
Utility Menu
home
pgup
end
pgdn
labels
f1
f1
f1
f1
f1
labels
Function keys
escape
enter
ctrl
space
shift
shift
enter
escape key



CO2R_µml
CO2S_µml
H2OR_mml
H2OS_mml
a 399.7 365.9 15.79 22.70
ΔCO2_µml
ΔH2O_mml
Flow_µml
RH_S %
b -33.8 6.91 172.8 70.09
Photo
Cond
Ci
Trmmol
c 15.4 0.216 238 3.49
1
Open LogFile
<view file>
<close file>
<add remark>
Match
labels
f1
f2
f3
f4
f5
labels

Page 3-16



Text Mode
press [
press]
press escape or [
press escape or]
Diagnostics Mode
Graphics Mode
press]
press [
(A) Stability Status = 2/3
1) CO2R 2) CO2S 3) TLeaf
Sec 20 20 20
Mn 0.1234 0.4321 21.34
SDv 1.1E-01 1.1E-01 1.1E-01
%CV 1.1E-01 1.1E-01 2.2E-02
Slp 3.3E-01 3.3E-01 3.3E-03
CO2R
CO2S
-120 SECS 0

0

Parameters, their units and location

Group	Label	Description
A	CO2R_µml	Reference cell CO_2 (µmol CO_2 mol^{-1})
	CO2S_µml	Sample cell CO_2 (µmol CO_2 mol^{-1})
	H2OR_mml	Reference cell H_2O (mmol H_2O mol^{-1})
	H2OS_mml	Sample cell H_2O (mmol H_2O mol^{-1})
B	ΔCO2_µml	CO_2 delta (sample - reference) (µmol CO_2 mol^{-1})
	ΔH2O_mml	H_2O delta (sample - reference) (mmol H_2O mol^{-1})
	Flow_µml	Flow rate to the sample cell (µmol s^{-1})
	RH_S_%	Relative humidity in the sample cell (%)
C	Photo	Photosynthetic rate (µmol CO_2 m^{-2} s^{-1})
	Cond	Conductance to H_2O (mol H_2O m^{-2} s^{-1})
	Ci	Intercellular CO_2 concentration (µmol CO_2 mol^{-1})
	Trmmol	Transpiration rate (mmol H_2O m^{-2} s^{-1})
D	Ci/Ca	Intercellular CO_2 / Ambient CO_2
	VpdL	Vapor pressure deficit based on Leaf temp (kPa)
	VpdA	Vapor pressure deficit based on Air temp (kPa)
E	Stable	Stability status: # Stable / # Checked
	StableF	Stability status as a decimal value
	<letters>	Stability flags: 1's and 0's for each variable
	TotalCV	Sum of the CVs of the stability variables
F	RH_R_%	Relative humidity in the reference cell (%)
	RH_S_%	Relative humidity in the sample cell (%)
	Td_R_%	Dew point temp in the reference cell (C)
	Td_S_%	Dew point temp in the sample cell (C)

Group	Label	Description
G	Prss_kPa	Atmospheric pressure (kPa)
	ParIn_µm	In-chamber quantum sensor (µmol m^{-2} s^{-1})
	ParOutµm	External quantum sensor (µmol m^{-2} s^{-1})
	BLC_mol	Total boundary layer conductance for the leaf (includes stomatal ratio) (mol m^{-2} s^{-1})
H	Tblock°C	Temperature of cooler block (C)
	Tair°C	Temperature in sample cell (C)
	Tleaf°C	Temperature of leaf thermocouple (C)
I	HH:MM:SS	Real time clock
	Program	Shows AutoProgram status
	CHPWMF	Status word (summary of line J)
	Battery	Battery voltage (V)
J	CO2	Status of CO_2 IRGAs
	H2O	Status of H_2O IRGAs
	Pump	Status of pump
	Flow	Status of Flow controller
	Mixr	Status of CO_2 mixer
	Fan	Speed of chamber fan
K	Program	Shows AutoProgram status
	ProgPrgs	AutoProgram step counter
	FwMxCrLp	Numerical summary of the four stability flags
	Stable	Stability status
L	CRagc_mv	Reference CO_2 AGC (automatic gain control) signal, in mV
	CSagc_mv	Sample CO_2 AGC signal
	HRagc_mv	Reference H_2O AGC signal
	HSagc_mv	Sample H_2O AGC signal

1	*Logging control; IRGA matching*	Open LogFile	<view file>	<close file>	<add remark>	Match
2	*Environmental control manager keys (CO_2, humidity, temp, light)*	<rspns>	Flow= 500µms	Mixer OFF	Temp OFF	Lamp= OFF
3	*Chamber fan speed; system and user-defined constants*	AREA= 6.00	STOMRT= 1.00	LeafFan Fast	Prompts off *	Prompt All *
4	*Real Time Graphics control*		GRAPH QuikPik	View Graph	GRAPH Setup	
5	*AutoProgram control; defining what's logged.*	AUTO PROG		Log Options	Define Stablty	Define Log Btn
6	*Text display control*	Display QuikPik	Display List	What's What	Display Editor	Diag Mode

Pages 3-17, 27-18

8	Flr QuikPik	Flr Editor	Define Actinic	Flr Adjust	Rcrdng OFF	
9	Meas is ON	Flash	Dark Pulse	Actinic is OFF	FarRed is OFF	
0	Do Fo	Do Fm	Do FoFm		View Fsh/Drk	*When Actinic Off*
0	Do Fo'	Do Fs	Do FsFm'	Do Fs Fm' Fo'	View Fsh/Drk	*When Actinic On*

m	F	dF/dt	FlrCV_%	FlrEvent
n	Fo	Fm	Fv/Fm	
o	Fo'	Fm'	Fv'/Fm'	Fs
p	ΔF/Fm'	ETR	qP	qN
q	Adark	LeafAbs	PS2/1	PhiCO2
r	F	M:Int kHz Hz Gn		
s	FlrMax	F:Dur Int Blu kHz Hz		
t	FlrMin	D:Dur Far Bfr Aft kHz Hz		

Page 27-18

6400-40 LCF

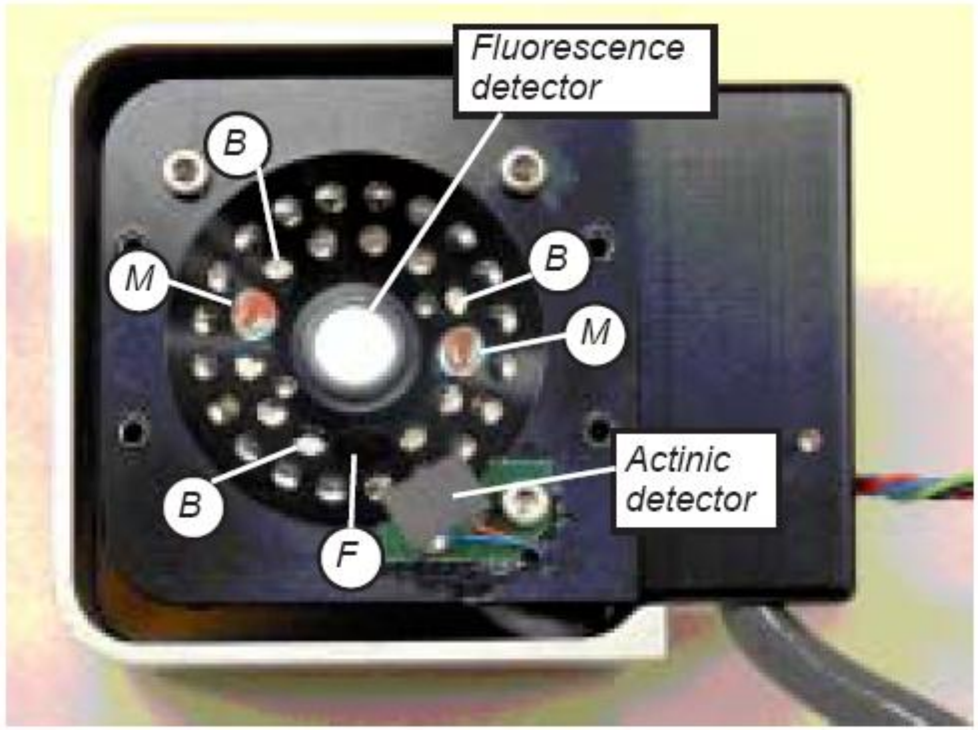


Figure 27-1. View of the LEDs of the LCF, showing the two red modulated measuring beam LEDs (M), the three blue actinic LEDs (B), and the far red LED (F). The remaining LEDs are red, and are used for actinic and saturating flashes.

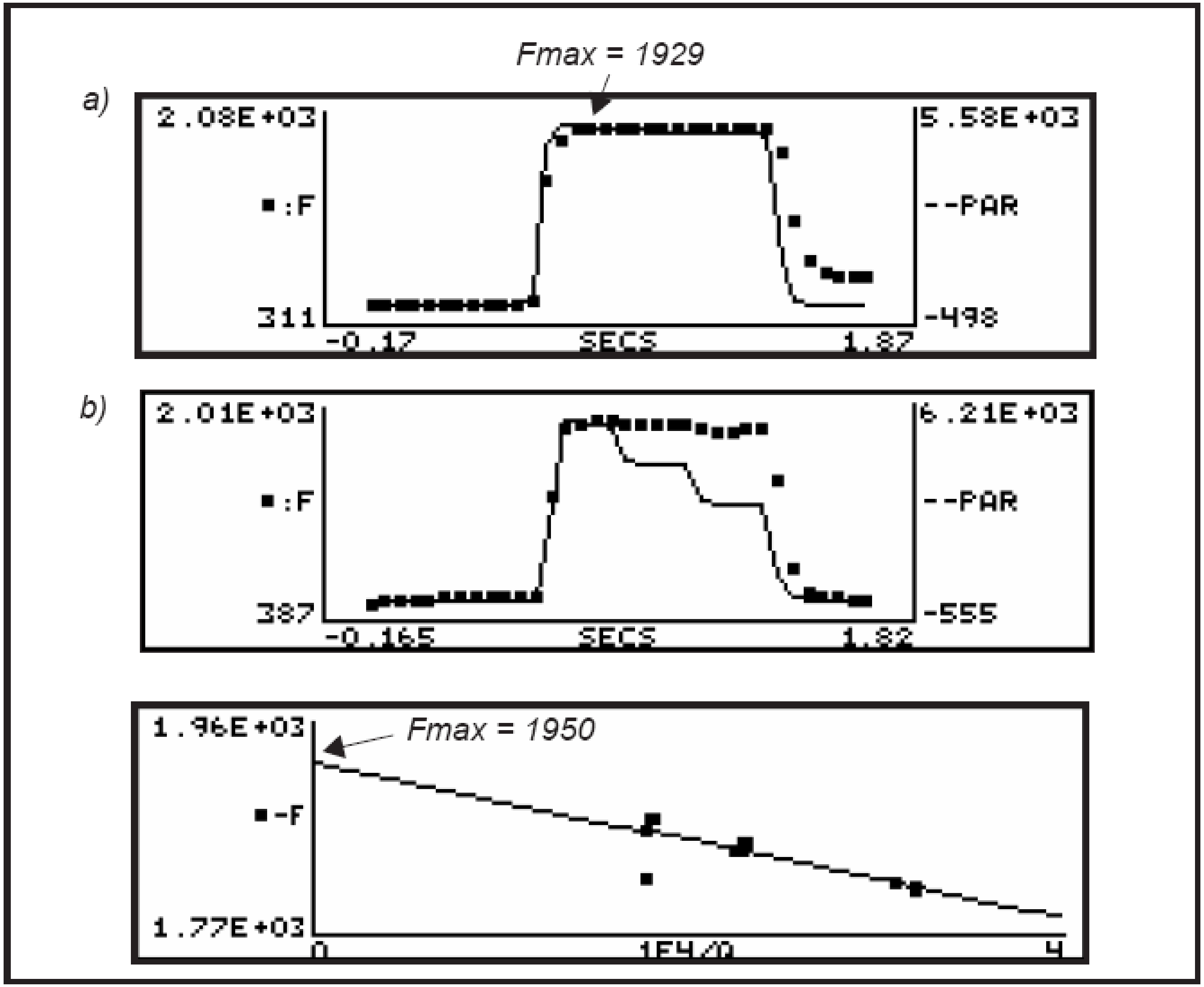
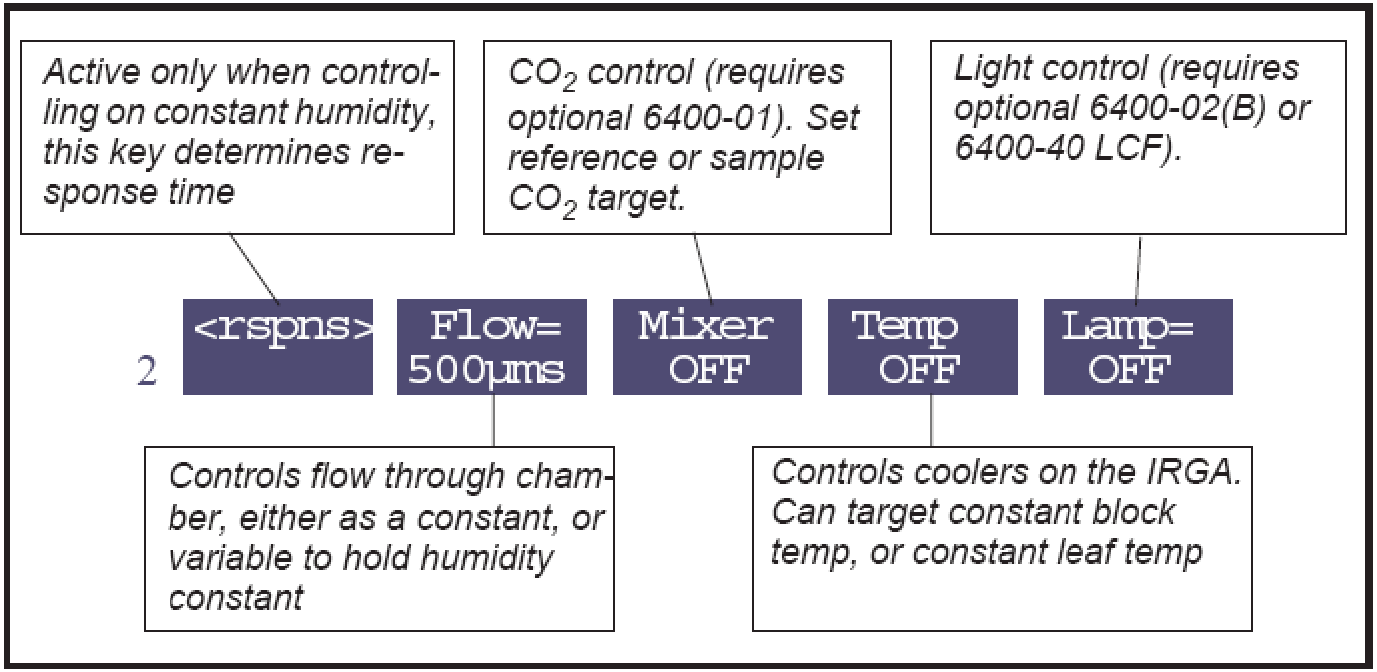


Figure 27-17. Two types of saturating flashes: a) single and b) multiple intensity with regression analysis.

Controlling Chamber Conditions

Chamber conditions are controlled from New Measurements mode via the function keys on level 2 (Figure 3-25).



Set Area and Stomatal Ratio

In New Measurements mode, press **3**, and set the leaf area and stomatal ratio for this leaf. Leaf area is simply the area exposed inside the chamber. If you are using a 2x3 chamber and filling it, the area is 6 cm^2. Stomatal ratio is an estimate of the ratio of stomata on one side of the leaf to the other. 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.

Checklist for using the LI6400

A. During Warm Up
1) Air Supply: Prepare CO_2 Mixer or Buffer Volume
2) Temperatures: Values OK? T_{leaf} responding?
3) Light Source, Sensors: Responding? Values OK?
4) Pressure Sensor: Value OK? Stable?
5) Leaf Fan: Running?
6) Flow Control: Max flow OK? Chemical tube restrictions?

B. After Warm-up
1) Check the flow zero
2) Adjust latch, close chamber
3) Check CO_2 zero
4) Check H_2O zero
5) Mixer Calibration (optional)
6) Lamp Calibration (optional)
7) Check T_{leaf} zero
8) Set Reference CO_2 and H_2O
9) Test for leaks
10) Match the IRGAs. Valve working?

C. Measuring the First Leaf
1) Set Light
2) Set Flow to 400 $\mu mol\ s^{-1}$
3) Set Reference CO_2
4) Temperature?
5) Clamp onto leaf
6) Set Area and Stomatal Ratio
7) Set constant humidity?