

The CIRAS-2 CO<sub>2</sub>/H<sub>2</sub>O analyzer is an inherently stable instrument and does NOT require frequent calibration. The AutoZero function corrects for nearly all changes that result in calibration drifts. The most common causes of drifts in CIRAS-2 readings are i) incorrect Zeros due to exhausted chemicals or ii) leaks on the desiccant columns from rolled or damaged o-rings and iii) incorrect Diff Bal, applied while reference CO<sub>2</sub> (Cr) is changing or unstable.

If needed, recalibration can be accomplished most easily by using the internal CO<sub>2</sub> cartridge as the calibration gas source, or a laboratory standard CO<sub>2</sub> gas may be used if desired. For H<sub>2</sub>O recalibration the PP Systems water vapor calibrator is provided with each new system, or another standard humidity source may be used. We recommend use of the water vapor calibrator because the FeSO<sub>4</sub> salts have long-term stability and the calibrator requires no humidity or flow setting adjustments.

Our factory calibration ranges of 0-2000 ppm CO<sub>2</sub> and 0-50 mb water vapor are ideally suited for most typical applications. The system can also be calibrated to specific upper limits within these ranges, for example, be sure to use a certified gas standard with a concentration slightly greater than the upper limit of expected measurements, e.g. 1700-2000 ppm for A/Ci curves.

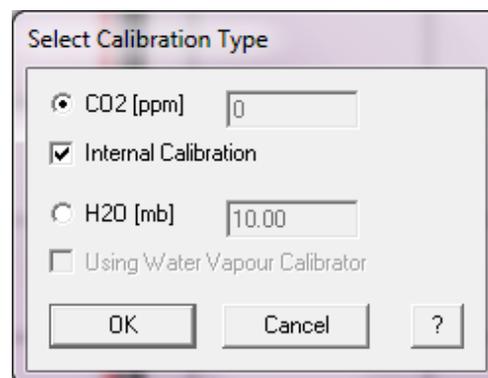
### Procedure

Allow the system to warm up for at least 30 minutes. The CO<sub>2</sub> cylinder should be fresh and the chemicals must be new or recent. Pay special attention to molecular sieve; be sure that it is recent. If the molecular sieve in its original container is older than 1 year consider replacing it – reopening and closing the container repeatedly exposes the chemical to air, and it will eventually lose its efficacy.

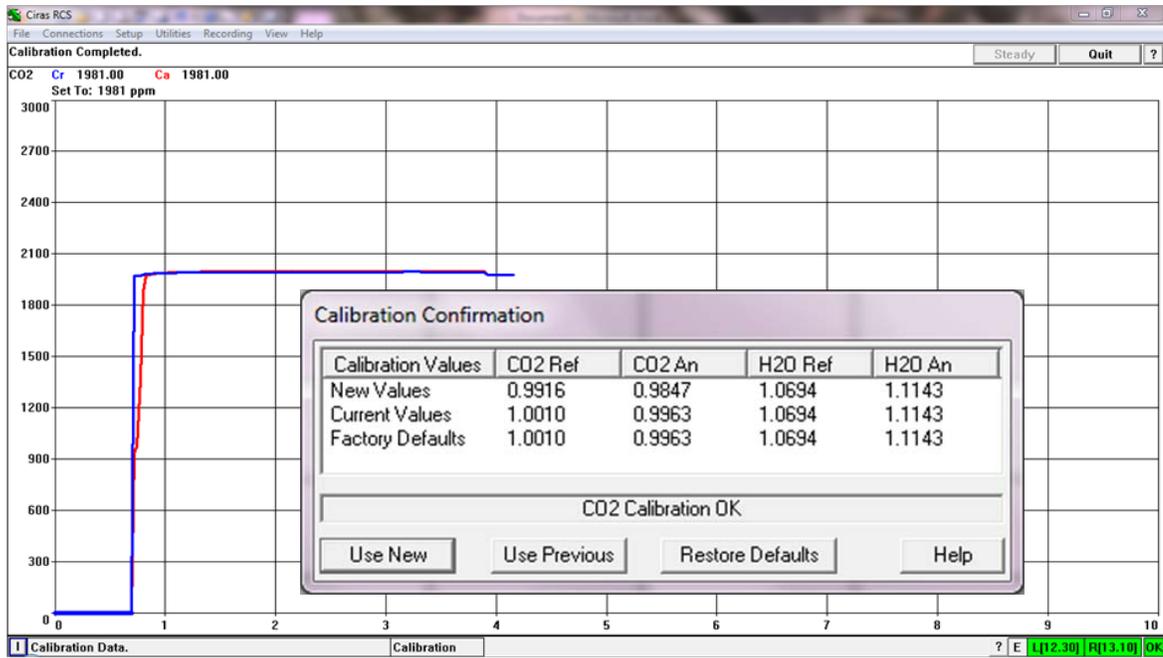
Run the Calibration Check function from the Utilities menu to determine if the system requires recalibration (see PP Systems Application Note AN2011-02 for instructions).

### CO<sub>2</sub> Calibration – internal source gas

1. Attach 3-way tubing with AIR OUT and REF IN gas ports sharing a single “T” or “Y” fitting and AIR IN as the third leg. The open leg of the tubing vents to atmosphere and should be about 30 cm in length. Use low-absorption tubing material, such as butyl rubber (See Fig.1 in Application Note AN2011-02).
2. Go to Utilities > Calibration in the CIRAS-2 main menu, then select ‘CO<sub>2</sub>’ and check the ‘Internal Calibration’ box (see below).
3. Click OK, system will perform Zero and return to measurement mode with the solenoid valves set to Diff-Bal (reference air sample passes through both the reference and analysis cells of the analyzer).
4. Allow system to stabilize for 2-3 minutes, noting ‘Expected’ ppm and difference of Cr (blue plot), Ca (red plot) status values ppm.
5. Once the readings remain stable click the **Steady** button.
6. Upon completion of calculating the calibration (some 20 seconds), CIRAS-2 sends the new calibration constants back to the firmware.



The following Dialog will appear:



Assuming that the calibration succeeded, three options are available:

- 1) **Use New** - use values determined by the new calibration
- 2) **Use Previous** - restore the previous calibration values (may be identical to Factory Defaults)
- 3) **Restore Defaults** - reset scaling factors to original factory values

Check that the Calibration Values (New Values, Recent Values, Factory Defaults) in the CO2 Ref, CO2 An, H2O Ref, H2O An columns seem reasonable. Typically, the new value should be within  $\pm 0.05$  of the Factory Default value. CIRAS-2 will reject calibrations that lie outside of a broader range.

#### CO<sub>2</sub> Calibration – external gas standard

1. Attach tubing between regulated gas tank to a “T” fitting and REF IN gas port - the open leg of the “T” vents to atmosphere and should be about 30 cm in length. Flow rate from the tank should be set so that there is small excess of approx. 150 cc min<sup>-1</sup> flowing to atmosphere. Set the tank regulator flow to between 300-400 cc min<sup>-1</sup>.
2. Go to Utilities > Calibration in the CIRAS-2 main menu, then select ‘CO2’ and enter the calibration concentration in the field to the right. Do not check the ‘Internal Calibration’ box.
3. Click OK, CIRAS-2 will perform Zero.
4. Allow system to stabilize for 2-3 minutes, noting ‘Expected’ ppm and difference of Cr (blue plot), Ca (red plot) status values ppm.
5. Once the readings remain stable click the **Steady** button.
6. Upon completion of calculating the calibration (some 20 seconds), CIRAS-2 sends the new calibration constants back to the firmware.

**WARNING: Over-pressurization of the analyzer cells will cause expensive damage to the system – take special precautions as in Step 1 above to avoid pressurizing the cells.**

Alternatively, fill a scientific-grade gas sampling bag with the calibration gas from the tank. Attach tubing from the gas sample bag’s valve to the REF IN port and CIRAS-2 will draw the gas sample without risk of pressurization.

As in the first example above, accept or reject the new calibration factors.

## H<sub>2</sub>O Calibration with PP Systems water vapor calibrator

If the H<sub>2</sub>O Calibration Check indicates that CIRAS-2 is out of calibration range you can proceed with recalibration as follows:

1. Go to Utilities > Calibration in the CIRAS-2 main menu, then select 'H<sub>2</sub>O' and check the 'Using Water Vapor Calibrator' box (see above).
2. Connect the water vapor calibrator to the PLC socket - the calibrator is automatically detected.
3. CIRAS-2 will confirm that the FeSO<sub>4</sub> salts in the calibrator are at an acceptable temperature. There is a risk of condensation inside the analyzer cells if the salts are too warm and ambient air temperature is low. Wait for 10 seconds to see if a warning appears "Temp. of FESO<sub>4</sub> too hot - Risk of Condensation [83]". If it is issued, allow the calibrator and CIRAS-2 to equilibrate to air temperature.
4. Connect the 3 gas lines on the calibrator as follows: lower tube to REF IN, upper tubes to REF OUT and AN OUT.
5. Allow 2 minutes for CIRAS-2 to settle to the generated water vapor pressure.
6. Note 'Expected' mb and difference Hr (blue plot), Ha (red plot) status values mb.
7. Once the readings remain stable click the **Steady** button.
8. Upon completion of calculating the calibration (some 20 seconds), CIRAS-2 sends the new calibration constants back to the firmware.

If Hr and/or Ha exceed  $\pm 0.5$  mb from expected a warning will appear in the Status field. As in the first example above, accept or reject the new calibration factors.

### References:

Parkinson K.J. and W. Day 1981. Water Vapour Calibration using Salt Hydrate Transitions. Journal of Experimental Botany 32:411-418.

CIRAS-2 CO<sub>2</sub> and H<sub>2</sub>O Calibration Check Function, Application Note # 2011-02

PP Systems Inc. 2011. CIRAS Water Vapor Calibrator Instructions, Version 1.02.

PP Systems Inc. 2010. CIRAS-2 Portable Photosynthesis System Operator's Manual. Version 2.04.

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