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## 16 Calibration

Calibration of the stem psychrometer is accomplished by preparing a range of standard sodium chloride (NaCl) solutions of known molality in the range 0.1, 0.2, 0.3, 0.4, 0.5 and 1.0 Molal. The exact NaCl volumes per 50ml volume of water are provided in the [Appendix Preparation of Calibration Solutions](#). These concentrations are derived from the work of Lang, A.R.G, *Osmotic Coefficients and Water Potentials of Sodium Chloride Solutions from 0 to 40°C*, 1967. Australian Journal of Chemistry, 20, 2017-23. ([Appendix Osmotic Coefficients and Water Potentials of Sodium Chloride Solutions](#)). This range of concentrations correspond well to the typical range of water potentials experienced by crop plants and trees (-0.642, -0.915, - 1.368, -1.823, -2.281 and -4.640 MPa).

Saturated filter paper disks (Watman's No.1) are used for calibration purposes. These are supplied by ICT already cut to the correct 6mm diameter size required for the calibration lid holder. Replacements are conveniently obtained using a standard paper punch. Measurement of thermocouple output in the suggested range is essentially linear at a given temperature.

**NOTE 27:** Always use freshly made NaCl calibration solutions using the “recipe” as per [Appendix Preparation of Calibration Solutions](#). If you are making calibration solutions for the first time, we recommend watching: VIDEO 16 – [Making Calibration Solutions](#)

### 16.1 Pre-Calibration Cleaning

The slope and intercept will be different for each psychrometer as the thermocouples are hand made using a very thin 25 µm wire. The thermocouple surface will have a natural variation that may have more or less dimpling providing different physics for the condensation of water. This is further impacted by the cleanliness of the thermocouple hence, it is imperative that the chamber is cleaned prior to calibration.

**NOTE 28:** When the chambers are cleaned, and calibrated at 25°C using fresh Molal calibrations solutions the calibration protocol reliably returns  $R^2$  of 0.999 to 0.9999.

It is advisable to enhance the vapour seal between the faces of the chamber and the calibration disk holder with a small amount of silicon grease. It is also helpful if calibration is carried out in a stable thermal environment or temperature controlled chamber.

Watch a demonstration: VIDEO 17 – [Sealing the Calibration Lid to the Psychrometer to perform a calibration measurement](#)

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## 16.2 Calibration Temperature

Calibration is usually done at, or corrected to, 25°C. Temperature correction for Psychrometric measurements is given by:

$$\text{Corrected Reading} = \text{Reading} / (0.027T + 0.325) \text{ Equation 3}$$

**Where:** T is chamber temperature in °C.

The calibration routine should be carried out in a temperature controlled chamber if possible. However, if you can only manage crude temperature control, then this is often worse than none at all. Rapid cycling of temperature caused by heating and cooling systems initiates transient gradients in the instrument and confound accurate calibration procedures. The relatively stable temperature of a large room is preferable. Corrections for ambient conditions can be made using the formula above.

## 16.3 Calibration Chamber

Here is a simple and effective means to achieve stable temperature control: You will require a good quality circulating water bath, preferably with proportional control of water temperature. This eliminates the typical "sawtooth" temperature control of less sophisticated baths. Circulate the water through a flexible copper tube (1-2 cm diameter and 2-3 m long) which is then coiled and placed inside a box close to the inside edges. Construct the box of dense styrofoam (eg. 5 cm. thickness, Styrofoam SM) and line all inside surfaces with a reflective foil (eg. aluminium foil). The dimensions of the box should accommodate the number of psychrometers you intend to calibrate as well as one or two small electric fans to facilitate air mixing. Internal dimensions of approximately 50x50x50 cm are appropriate. Suspend a light plastic grid in the centre of the box to serve as a platform for the psychrometers. This allows air movement around the instruments and eliminates temperature gradients from conductive surfaces. Normal equilibration times for salt solutions are quite brief but it is safest to allow between 30 to 60 minutes for each solution to equilibrate depending upon how intensively the psychrometer was handled while loading the calibration solution.

**VIDEO 18:** A demonstration detailing the components used in the construction and use of a simple [calibration chamber for psychrometers](#).

## 16.4 Calibration Protocol

Prior to attempting a calibration of the psychrometer for the first time it is recommended that you watch VIDEO 19: [Calibration Protocol](#), this video clearly details all the practical steps involved in the successful calibration of the PSY1 Stem Psychrometer. An additional video, VIDEO 20, demonstrates the software [Calibration Function](#), used to initiate a measurement and process the calibration data.

The calibration routine is designed to be semi-automated and all inclusive within the PSY1 software, from making measurements of calibration solutions to plotting the data, generating a slope and intercept and storing the calibration in the firmware of the PSY1 Instrument for real time data processing.

### 16.4.1 Start Calibration

To begin a calibration, start by opening the software and connecting to the PSY1 instrument. Then click on the Start Calibration button, the software is now in calibration mode and you will only be able to perform a calibration.

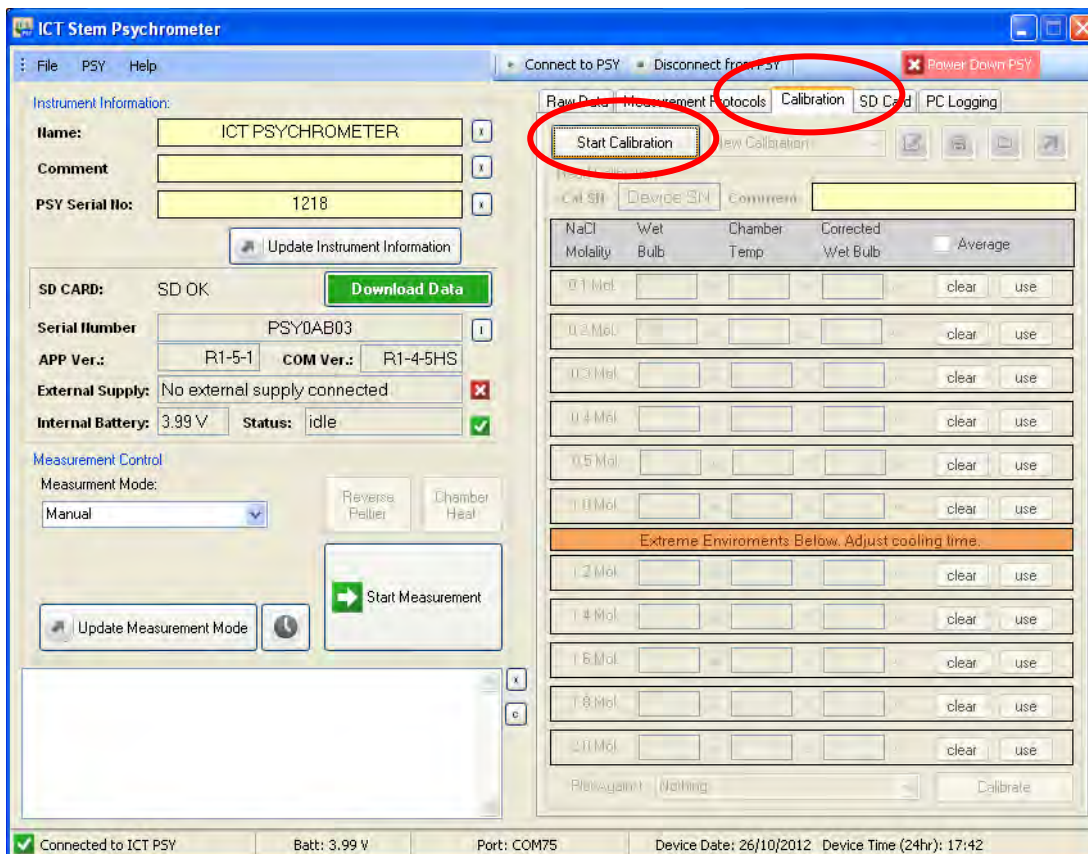
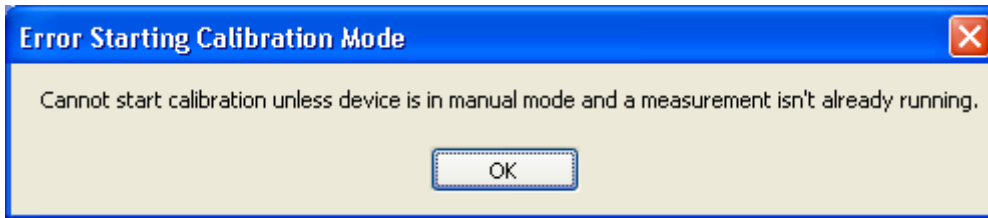


Figure 35: To begin a calibration, click the Start Calibration icon in the Calibration Tab.

**Note 29:** A calibration can only be performed in Manual mode. If you attempt to “Start a Calibration” while in Live Mode or while the instrument is logging, an error message is displayed.



**Figure 36: Error message displayed if calibration mode is attempted when the instrument is not in Manual Mode.**

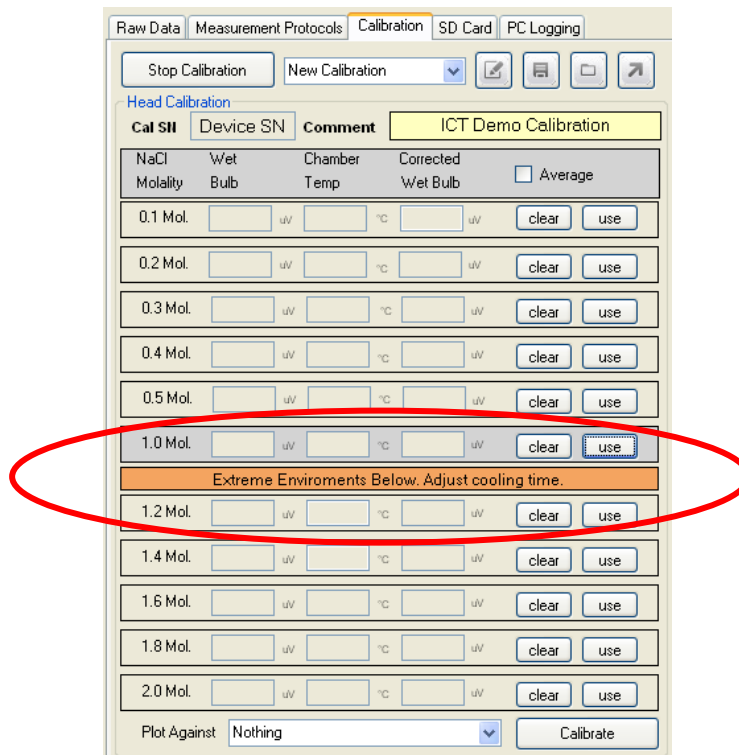
### 16.4.2 Loading the Calibration Filter Paper

Place a known calibration solution on a filter paper inside the calibration lid of the Psychrometer. Place the Psychrometer into an isothermal calibration chamber ideally set and regulated to 25°C and leave for at least 30 minutes to preferably 1 hour to equilibrate before making the measurement.

**NOTE 30:** It is advised to start with a 1.0 molal solution to test the integrity and cleanliness of the thermocouple. See diagnosing a [dirty thermocouple](#). Run a manual measurement on this solution first and plot the Peltier cooling curve before starting the calibration. If the plateau is not in the general range of 17-20  $\mu\text{V}$  for about 10 seconds then falls sharply to the zero reference and remains there then it is advised that the psychrometer be cleaned before proceeding with the calibration. Always good to know this before performing a calibration!

### 16.4.3 Selecting the Calibration Range

Click on the “use” button corresponding to the calibration solution you have placed in the psychrometer calibration lid. This measurement range is now active denoted by the subtle change in colour of all the input cells in that row.



**Figure 37: Selecting the calibration range.**

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#### 16.4.4 Start Measurement

Click the “Start Measurement” button, and a measurement is commenced.

Upon completion of the measurement the fields will automatically be populated with the relevant data:

- i. Wet Bulb Depression;
- ii. Chamber Temp;
- iii. Wet Bulb corrected to 25°C.

Remove the calibration solution and replace it with the next solution.

Repeat this procedure for at least 3 calibration solutions. The software is locked to ensure a minimum of any 3 points are recorded satisfy a calibration. 6 points are preferred.

**NOTE 31:** If you check the Average box you will be able to take multiple measurements at each cal solution and automatically average the values for use as the data point for that cal solution on the graph. Allow at least 10 minutes between replicate readings. Whilst this option is provided, if readings show sufficient variation to warrant using an average function it is recommended that you review your calibration procedure and clean the psychrometer.

The screenshot shows the 'Calibration' tab of the software interface. At the top, there are tabs for 'Raw Data', 'Measurement Protocols', 'Calibration', 'SD Card', and 'PC Logging'. Below these are buttons for 'Stop Calibration', 'New Calibration', and several icons. The main area is titled 'Head Calibration' and contains a table with columns: 'Cal SN', 'Device SN', 'Comment', 'ICT Demo Calibration', 'NaCl Molality', 'Wet Bulb', 'Chamber Temp', 'Corrected Wet Bulb', and an 'Average' checkbox. The 'Average' checkbox is checked and circled in red. Below the table are rows for different molalities (0.1, 0.2, 0.3, 0.4, 0.5, 1.0, 1.2, 1.4, 1.6, 1.8, 2.0) with input fields for 'uW' and '°C' and 'clear'/'use' buttons. A warning bar reads 'Extreme Enviroments Below. Adjust cooling time.' At the bottom, there is a 'Plot Against' dropdown menu set to 'Nothing' and a 'Calibrate' button.

**Figure 38:** Tick the average box if you wish to make multiple measurements at each calibration solution and average them.

#### 16.4.5 Generate Calibration Curve

When at least 3 data points have been measured click on the Calibrate button at the bottom of the screen.

Raw Data Measurement Protocols Calibration SD Card PC Logging

Stop Calibration New Calibration

Head Calibration

Cal SN Device SN Comment ICT Demo Calibration

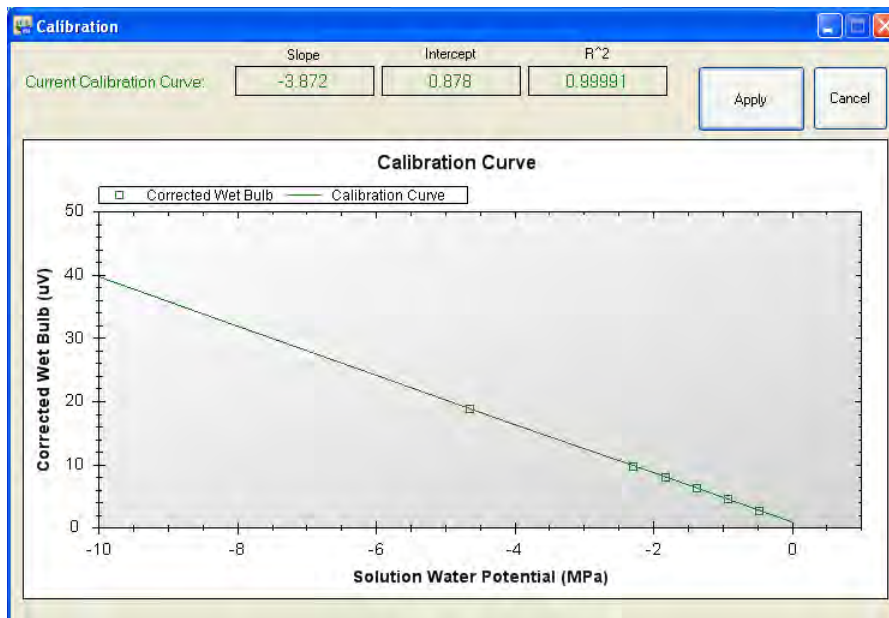
NaCl Molality	Wet Bulb	Chamber Temp	Corrected Wet Bulb	Average
0.1 Mol	2.60 $\mu\text{V}$	25.00 $^{\circ}\text{C}$	2.60 $\mu\text{V}$	<input type="checkbox"/>
0.2 Mol	4.50 $\mu\text{V}$	25.10 $^{\circ}\text{C}$	4.49 $\mu\text{V}$	<input type="checkbox"/>
0.3 Mol	6.20 $\mu\text{V}$	24.90 $^{\circ}\text{C}$	6.22 $\mu\text{V}$	<input type="checkbox"/>
0.4 Mol	7.90 $\mu\text{V}$	24.80 $^{\circ}\text{C}$	7.94 $\mu\text{V}$	<input type="checkbox"/>
0.5 Mol	9.60 $\mu\text{V}$	24.80 $^{\circ}\text{C}$	9.65 $\mu\text{V}$	<input type="checkbox"/>
1.0 Mol	18.80 $\mu\text{V}$	24.90 $^{\circ}\text{C}$	18.85 $\mu\text{V}$	<input type="checkbox"/>
Extreme Enviroments Below. Adjust cooling time.				
1.2 Mol	<input type="text"/> $\mu\text{V}$	<input type="text"/> $^{\circ}\text{C}$	<input type="text"/> $\mu\text{V}$	<input type="checkbox"/>
1.4 Mol	<input type="text"/> $\mu\text{V}$	<input type="text"/> $^{\circ}\text{C}$	<input type="text"/> $\mu\text{V}$	<input type="checkbox"/>
1.6 Mol	<input type="text"/> $\mu\text{V}$	<input type="text"/> $^{\circ}\text{C}$	<input type="text"/> $\mu\text{V}$	<input type="checkbox"/>
1.8 Mol	<input type="text"/> $\mu\text{V}$	<input type="text"/> $^{\circ}\text{C}$	<input type="text"/> $\mu\text{V}$	<input type="checkbox"/>
2.0 Mol	<input type="text"/> $\mu\text{V}$	<input type="text"/> $^{\circ}\text{C}$	<input type="text"/> $\mu\text{V}$	<input type="checkbox"/>

Plot Against Nothing

Calibrate

**Figure 39: Click calibrate once all measurements have been recorded.**

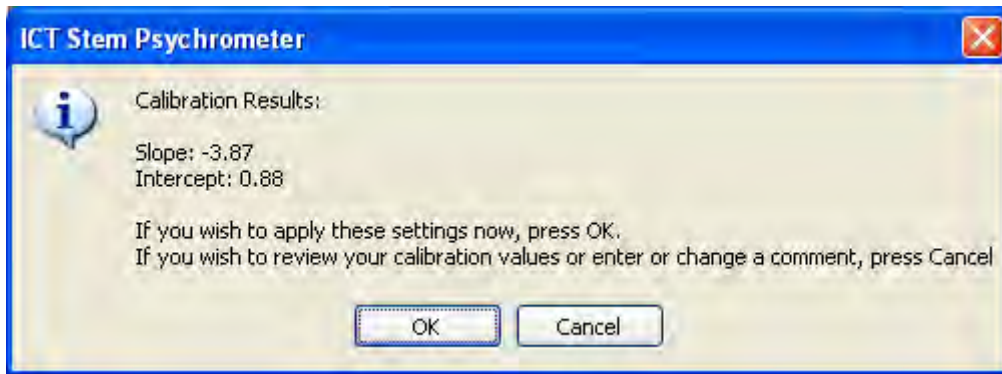
The data points will be automatically plotted, a regression line fitted, the slope, intercept and  $R^2$  calculated. If the calibration is of suitable accuracy, an  $R^2$  of at least 0.999, preferably 0.9999, should be possible and the slope should be within the general range of -3.8 to -4.2  $\mu\text{V}/\text{MPa}$  with an intercept of between 0 to 1  $\mu\text{V}$ .



**Figure 40: The calibration results are automatically processed and plotted.**

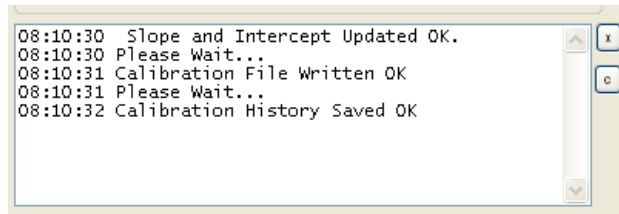
#### 16.4.6 Write Calibration to Firmware

Click on the Apply icon located in the top right hand corner of the plot. This will automatically write the calibration slope and intercept to the firmware of the PSY1 and then take you to the measurement protocol tab and stop the calibration.

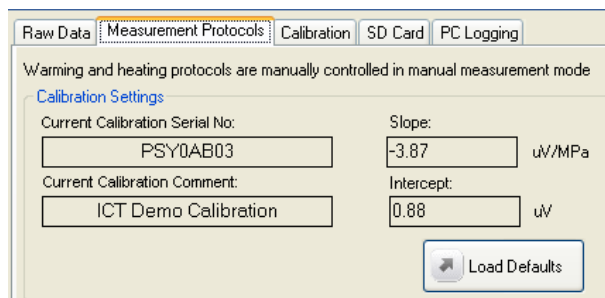


**Figure 41: Confirmation Window before writing PSY1 Calibration settings to firmware.**

Once the results are confirmed they are automatically written to firmware and displayed in the Calibration Settings on the Measurement Protocols tab. Confirmation that the calibration has been written correctly and the calibration history saved correctly (OK) is given in the dialogue box. The calibration is now active for this Psychrometer and all measurements made with the PSY1 will use this calibration.



**Figure 42: Dialogue box - confirmation of calibration change.**



**Figure 43: New calibration slope and intercept saved to firmware and displayed in calibration settings.**

#### 16.4.7 Calibration storage location

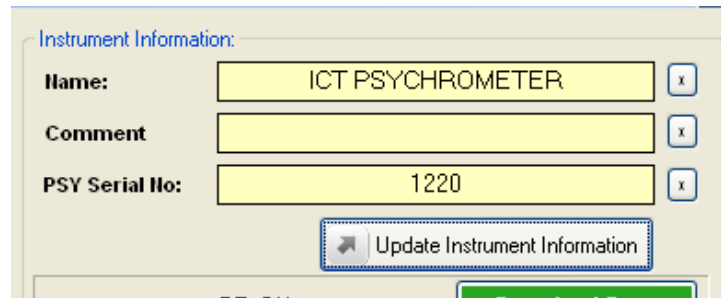
The calibration data is saved to a \*.rdf calibration file on the MicroSD card using the same name as the number located in the PSY Serial No. Field. This should correspond to the four digit Serial Number of the psychrometer that has been calibrated.

**NOTE 32:** You should enter the 4-digit serial number of the Psychrometer chamber into this field at the commencement of the calibration and click the update sensor information button.

#### 16.4.8 Loading a Different Calibration

A new or different calibration can be activated by entering a different chamber serial number in the PSY Serial No. field and clicking the update sensor information button. If the calibration file exists on

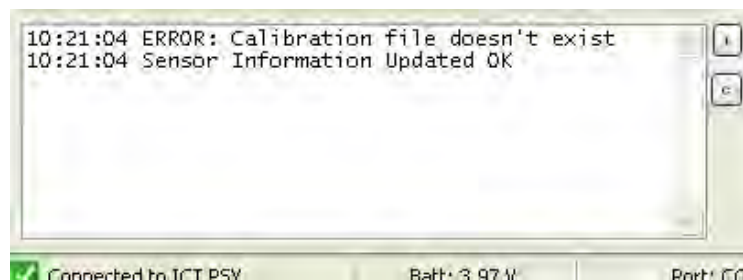
the MicroSD card, it will be loaded as the active calibration and all measurements made with the PSY1 will have the raw Wet Bulb Depression data converted to Water Potential (MPa) using this calibration.



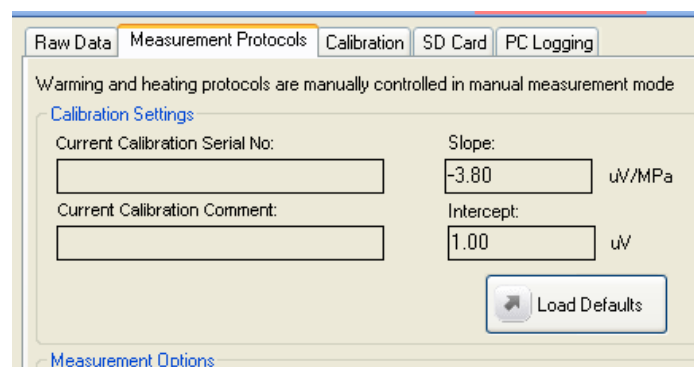
**Figure 44: PSY Serial No. field, located in the instrument information panel. Change this number to load a different calibration file.**

#### 16.4.9 Default Calibration

The default calibration is a historical average slope and intercept of a small sub sample of all psychrometers ever made. It is a good general calibration, that will always place the measured water potential within the right range, but it cannot be directly relevant to the specific output of any one psychrometer. If a psychrometer is being used and a calibration file does not exist, the PSY1 will display a warning in the Dialogue box and the default calibration will be loaded automatically.



**Figure 45: Dialogue Box notification that the calibration file does not exist.**



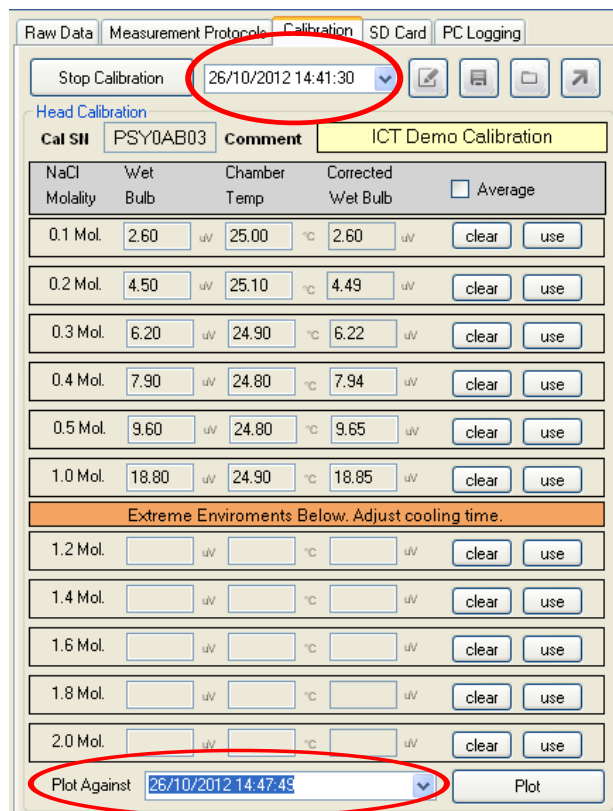
**Figure 46: Default Calibration Loaded.**

**Warning 11 – Any data processed with the default calibration will not be accurate. This data can only be considered relative and may display improbable positive water potential values when measurements are made close to zero MPa.**



## 16.5 Checking for calibration drift

Calibration drift should be checked by overlaying the existing calibration file. This is stored in the PSY1 and used for a 3 point check, plotting the points against each other every time the chamber is cleaned. To check for drift, select new calibration in the top drop down box and select the previous calibration in the Plot Against drop down menu at the bottom of the window. Then click Plot.



The screenshot shows the 'Calibration' window of the software. At the top, there are tabs for 'Raw Data', 'Measurement Protocols', 'Calibration', 'SD Card', and 'PC Logging'. Below the tabs, there is a 'Stop Calibration' dropdown menu set to '26/10/2012 14:41:30'. Underneath, the 'Head Calibration' section shows a table with columns for 'Cal S#', 'PSY0AB03', 'Comment', and 'ICT Demo Calibration'. The table contains several rows of calibration data for different molalities (0.1 to 2.0 Mol) with columns for 'Wet Bulb', 'Chamber Temp', and 'Corrected Wet Bulb'. A warning banner reads 'Extreme Enviroments Below. Adjust cooling time.' At the bottom, there is a 'Plot Against' dropdown menu set to '26/10/2012 14:47:49' and a 'Plot' button. Red circles highlight the 'Stop Calibration' and 'Plot Against' dropdown menus.

Cal S#	PSY0AB03	Comment	ICT Demo Calibration	
NaCl Molality	Wet Bulb	Chamber Temp	Corrected Wet Bulb	<input type="checkbox"/> Average
0.1 Mol.	2.60	25.00	2.60	<input type="button" value="clear"/> <input type="button" value="use"/>
0.2 Mol.	4.50	25.10	4.49	<input type="button" value="clear"/> <input type="button" value="use"/>
0.3 Mol.	6.20	24.90	6.22	<input type="button" value="clear"/> <input type="button" value="use"/>
0.4 Mol.	7.90	24.80	7.94	<input type="button" value="clear"/> <input type="button" value="use"/>
0.5 Mol.	9.60	24.80	9.65	<input type="button" value="clear"/> <input type="button" value="use"/>
1.0 Mol.	18.80	24.90	18.85	<input type="button" value="clear"/> <input type="button" value="use"/>
Extreme Enviroments Below. Adjust cooling time.				
1.2 Mol.				<input type="button" value="clear"/> <input type="button" value="use"/>
1.4 Mol.				<input type="button" value="clear"/> <input type="button" value="use"/>
1.6 Mol.				<input type="button" value="clear"/> <input type="button" value="use"/>
1.8 Mol.				<input type="button" value="clear"/> <input type="button" value="use"/>
2.0 Mol.				<input type="button" value="clear"/> <input type="button" value="use"/>

Figure 47: Plotting a new calibration against an existing calibration from file.

If the calibration curves are significantly different (>5%), a full 6 point calibration should be performed and saved to the firmware of the PSY1 to replace the old calibration. The psychrometer should be cleaned every time it is uninstalled and/or prior to deployment. If the psychrometer has been stored for any length of time, even if it has been stored correctly (a smear of vacuum grease around the inside of the calibration lid, taped up) to prevent oxidation of the thermocouples. Oxidisation of the thermocouples will still occur, even if only mildly, but it has the potential to influence readings and be the cause of potential errors in calibration even if the chamber is perceived to be "Clean". Always [Clean the Psychrometer](#) before calibration.

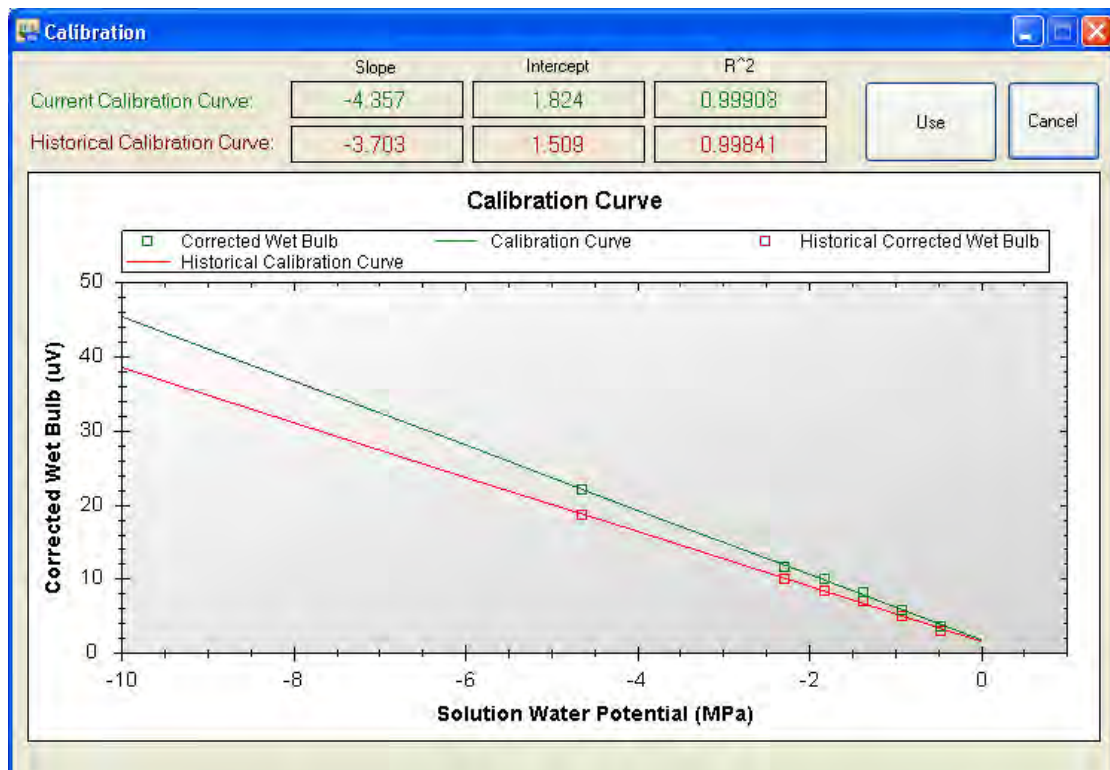


Figure 48: Comparison of a new calibration against an existing calibration from file.

## 16.6 Manual Editing of the Calibration

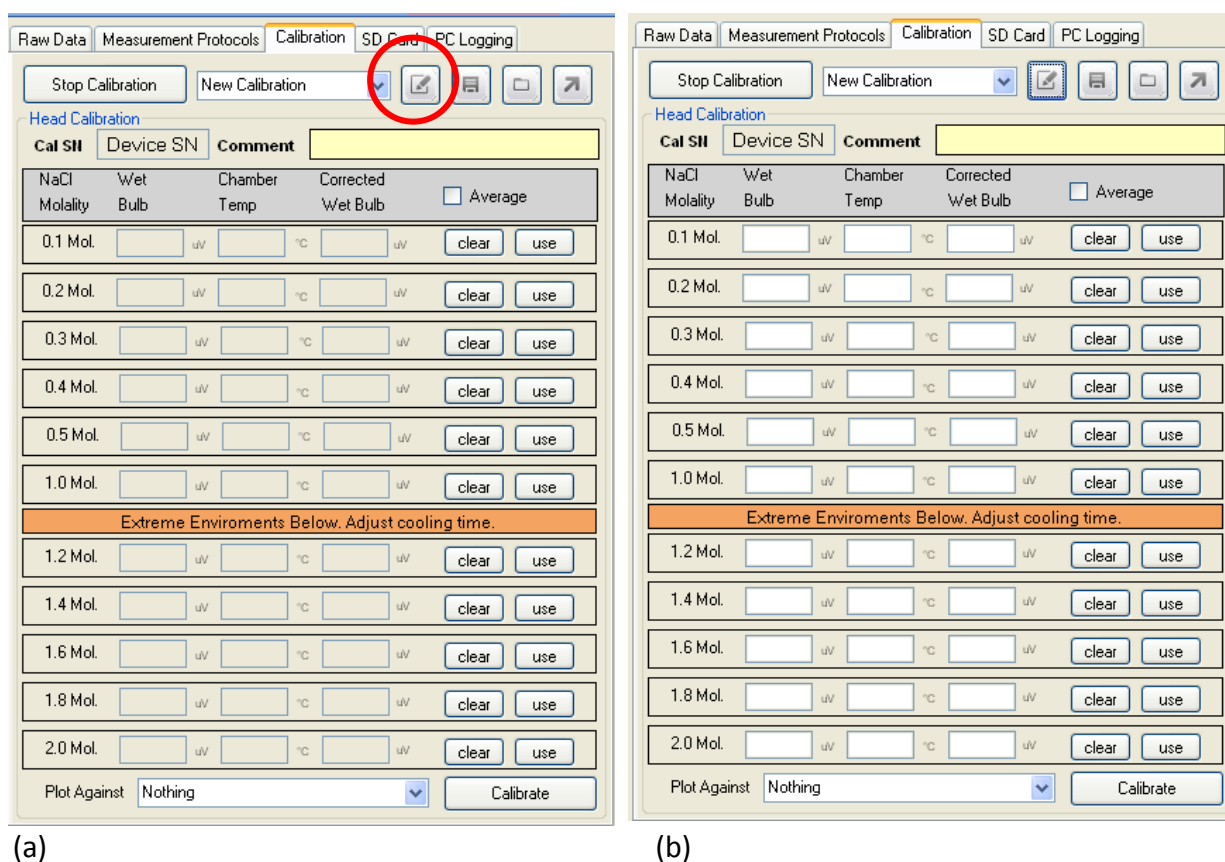
For best results, calibrating a psychrometer should be done within a short period of time, under the same, stable environmental conditions. A full 6 point calibration can take 6 hours, allowing a full hour of thermal equilibration time between the measurements of each data point. This may not always be practical, so an option to manually edit the calibration is included. This feature enables outlying data points of concern to be checked and remeasured without the need to repeat the whole calibration.

**NOTE 33:** The integrity of a calibration is imperative to getting good, accurate water potential data. This function should not be used to modify individual points of an existing or old calibration in isolation. It should only be used to check individual points during a full calibration run should there be any doubt over the accuracy of an individual data point.

### 16.6.1 Edit the Calibration

When a new calibration is started, by default, the calibration input cells are locked, signified by the cells being greyed out (Figure 35 (a)). This is to ensure only directly measured values from the psychrometer can be automatically entered immediately following each calibration measurement.

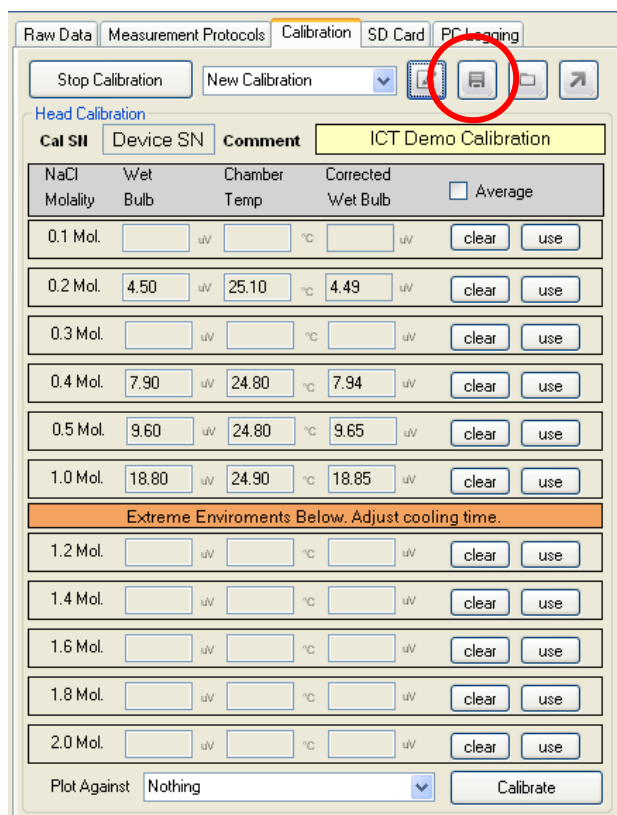
To edit a calibration you must first “open” the input cells of the calibration interface. This is done by clicking on the “edit the calibration” icon. After clicking the icon the cells change from being greyed out to white signifying that they are open and able to receive manual entries.



## 16.6.2 Save Current Calibration to Computer

Calibration data can be saved to your computer at any time by clicking on the Save Current Calibration icon. This process should be performed after each data point is measured to ensure that no data is lost in the event of a software or OS crash. Clicking on the icon opens a "Save As" Window that defaults to the directory: C:\Program Files\ICT\ICT Stem Psychrometer\\*.cqs. This directory can be changed to your preferred location.

**NOTE 34:** The calibration file should always be saved as the four digit Serial Number of the psychrometer being calibrated. This ensures a tracking reference in the future.



(a)



(b)

**Figure 50: (a) Click the Save Current Calibration icon to save the calibration data for later retrieval.**

**(b) Use the Save As Window to select a preferred location on your computer.**

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### 16.6.3 Load saved Calibration

Any calibration file (\*.cqs) saved to your computer can also be loaded within the calibration interface. This enables a calibration to be opened, edited, saved as an archived file name or transferred between PSY1 instruments should the psychrometer chamber be used on more than one instrument.

To load a saved \*.cqs calibration file, click on the Load Saved Calibration icon. Clicking on the icon opens an Open ... Window that defaults to the directory C:\Program Files\ICT\ICT Stem Psychrometer\\*.cqs or the directory where the last \*.cqs calibration file was saved.



**Figure 51: Loading a \*.cqs calibration file from a computer.**

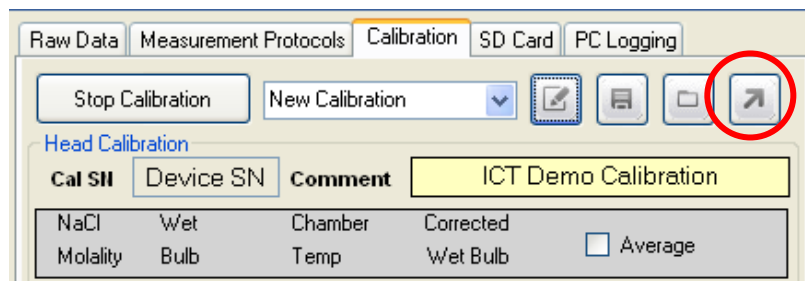
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### 16.6.4 Calibration Live View Mode

It is recommended that a minimum 30 to preferably 60 minutes thermal equilibration time be left between the handling and loading of each calibration solution. These figures are arbitrary in nature and designed to err on the side of caution to ensure all thermal gradients introduced to the psychrometer through handling of the chamber, introduction of samples of different temperatures, be allowed to dissipate so as not to cause errors in the calibration.

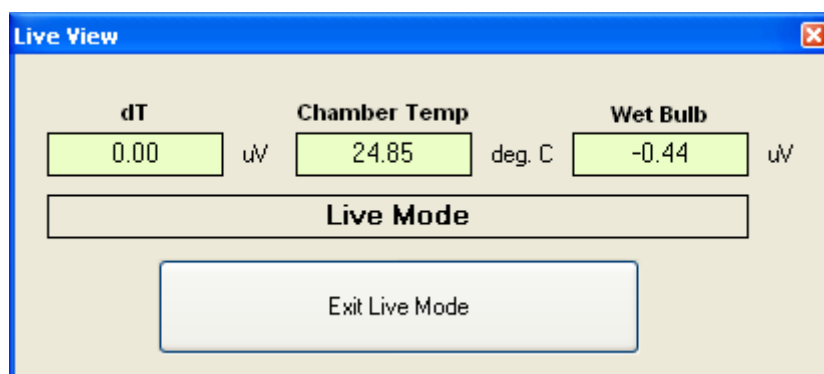
This can cause a calibration to be a long and time consuming process. An alternative protocol is to utilise the Live View feature incorporated within the calibration function. This is a more intensive protocol based on active interpretation of empirical data to determine how long the psychrometer takes to return to thermal equilibrium after loading a calibration solution.

To activate the Calibration Live View click the Live View icon in the upper right hand corner of the calibration tab:



**Figure 52: Calibration Live View icon.**

The Live View window initialises the microprocessor before continuously displaying the live values from all three thermocouples inside the psychrometer. Both dT and Wet Bulb will be elevated well above zero  $\mu\text{V}$  after loading the calibration solution. What must be determined is how long it takes for both values to reach and remain stable at zero  $\mu\text{V}$ . The user can apply discretion as to what constitutes a stable zero  $\mu\text{V}$  in relation to thermal equilibrium being reached. The values displayed below would constitute an adequate thermal equilibrium to initiate the next calibration data point measurement.



**Figure 53: Live View Window – thermal equilibrium achieved.**

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### 16.6.5 Calibration Summary file (\*.rdf)

The calibration summary file is attributed the name of the psychrometer chamber that was calibrated. The file consists of the calibration intercept and slope that was generated after performing the minimum 3-point calibration using known NaCl solutions. It also contains the basic reference information related to the calibration such as the serial number of the PSY1 instrument that the calibration was conducted with; the calibration comment and the date and time the calibration was completed.

0.88

-3.87

PSY0AB03

ICT Demo Calibration

26/10/2012\_14:48:36

### 16.6.6 Calibration File

The \*.cqs calibration file is simply a text (\*.txt) file that records and stores the raw calibration values for every calibration solution measured for a specific calibration run of a psychrometer. This file is stored on the Micro SD card or can be saved to your computer to allow loading it to the psychrometer or editing it at a later date. This can be a useful feature if a calibration cannot be finished or the user wishes to save each data point for fear of the OS crashing and the calibration data being lost.

The first line of the \*.cqs file is the header line or comment entered by the user. Then data is stored in a simple table. The left hand column is the Molal solution concentration: 0.1, 0.2, 0.3, 0.4, 0.5, and 1.0 Molal, the standard calibration concentration range. The column continues for 1.2, 1.4, 1.6, 1.8 and 2.0 Molal concentrations, or the Extreme environment calibration range. The subsequent 3 columns of the table refer to the direct measurements recorded during calibration process: Wet Bulb, Chamber Temp and Corrected Wet Bulb.

Values are only entered if measured by the PSY1. Because the data file can be [edited](#), there may be instances where only 1 value appears in a column, most commonly this would be the far right column for corrected Wet Bulb if the user has entered a nominal value for the purpose of demonstrating the calibration function or has values from a previous calibration that were manually entered into the calibration file.

**NOTE 35:** A minimum of 3 data points must be measured (or manually entered) to generate a calibration slope and intercept. Any fewer data points cannot yield a suitably linear calibration.

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### 16.6.7 Calibration History File

The calibration history file maintains a chronological record of all the calibrations performed on the psychrometer over the life of the instrument. Every time a calibration is made and saved using the four digit serial number of the psychrometer, the raw data for each calibration solution is stored in the file, including:

- i. Wet bulb depression,
- ii. Chamber temperature at which the measurement was made
- iii. Corrected wet bulb depression
- iv. Serial Number of the PSY1
- v. Comment
- vi. Date & Time

This provides a good reference to check for calibration drift over time.

#### **HCDATE\_26/10/2012\_14:47:49**

CC\_Check Calibration Drift

CS\_PSYOAB03

01\_2.45\_25.00\_2.45

02\_4.55\_25.00\_4.55

03\_6.40\_25.00\_6.40

04\_7.90\_25.00\_7.90

05\_9.80\_25.00\_9.80

10\_19.00\_25.00\_19.00

12\_ \_ \_

14\_ \_ \_

16\_ \_ \_

18\_ \_ \_

20\_ \_ \_

#### **HCDATE\_26/10/2012\_14:41:30**

CC ICT Demo Calibration

CS\_PSYOAB03

01\_2.60\_25.00\_2.60

02\_4.50\_25.10\_4.49

03\_6.20\_24.90\_6.22

04\_7.90\_24.80\_7.94

05\_9.60\_24.80\_9.65

10\_18.80\_24.90\_18.85

12\_ \_ \_

14\_ \_ \_

16\_ \_ \_

18\_ \_ \_

20\_ \_ \_

**NOTE 36:** If the thermocouple is broken and replaced, the relevance of the chronological calibration data is lost. A new thermocouple will have no correlation with the calibration history of a previous thermocouple. It is effectively now a different instrument.