MINI-PAM-II/ POROMETER Manual

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Heinz Walz GmbH • Eichenring 6 • 91090 Effeltrich • Germany Phone +49-(0)9133/7765-0 • Telefax +49-(0)9133/5395 E-mail info@walz.com • Internet www.walz.com

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2 Safety Instructions

2.1 General Safety Instructions

- Read safety instructions and the operating instructions prior to operation of the device and its accessories.
- Pay attention to all safety warnings.
- Keep device and its accessories away from water or high moisture areas.
- Keep the device and its accessories away from dust, sand and dirt.
- Do not put the device and its accessories near sources of heat.
- Ensure that neither liquids nor foreign bodies get inside the device or its accessories.
- Ensure sufficient ventilation.
- Connect the device only to the power source indicated in the operating instructions or on the device. If the device is not in use, remove the mains plug from the socket.
- The device and its accessories should only be repaired by qualified personnel.

2.2 Special Safety Instructions

- Turn off MINI-PAM-II before connecting or disconnecting MINI-PAM-II/POROMETER.
- The MINI-PAM-II/POROMETER is a highly sensitive instrument which should be only used for research purposes. Follow the instructions of this manual in order to avoid potential harm to the user and damage to the instrument.
- The MINI-PAM-II can emit very strong light! In order to avoid harm to your eyes, never look directly into the light port of the MINI-PAM-II or its fiberoptics.

Turn off MINI-PAM-II before connecting or disconnecting MINI-PAM-II/POROMETER or 2054-L External LED Light Source.

3 Introduction

The MINI-PAM-II/POROMETER is a new leaf-clip for MINI-PAM-II instruments. It combines measurements of stomatal conductance and chlorophyll *a* fluorescence for the assessment of PSII. As a lightweight, compact device, it is ideally suited for use in the field as well as in greenhouses and laboratories. The porometer provides precise information on evaporation and stomatal conductance for rapid screenings, and high-through-put stress assessment, but also analysis of stomatal movement in combination with well-established protocols like induction-curves or light-curves. Together with chlorophyll *a* fluorescence, these are important aspects for describing the photosynthetic activity.



The porometer is equipped with:

- humidity sensors for the determination of H_2O evaporation of the leaf and ambient humidity
- leaf temperature sensor
- pressure sensor
- flow sensors and a very silent pump
- adjustable pressure of the closing mechanism
- a GPS-module for tracking the sample and sun position in field applications and determination of angle of sunincidence on the leaf
- a sensor to measure photosynthetically active radiation (PAR) at leaf level with high accuracy, providing reliable light intensity data for electron transport rate (ETR) calculations
- a dark shield for easy determination of $\mathsf{F}_0,\mathsf{F}_M$ and measurements with controlled actinic light intensities
- ambient CO₂-sensor to monitor ambient CO₂-levels which are of interest in laboratory or greenhouse conditions

3.1 Description of the pneumatic pathway and its components

The MINI-PAM-II/POROMETER is an open system porometer. Fig. 1 displays the pneumatic pathway through the porometer. Air is drawn in from the surroundings and passes through a buffer (Air-In-Buffer) that compensates for peaks fluctuations. For system safety, the incoming air is filtered through a 1-2 µm filter. The pump follows, delivering the air at the preset flow rate. This is controlled by the following flowmeter (Mass-Flowmeter-In). The air is now analyzed in the reference analytic cell to determine the water vapor concentration within the ambient air. Subsequently this air passes by the leaf in the leaf chamber and absorbs the humidity released by the leaf. In the leaf chamber, additionally the leaf temperature is determined by a non-contact infrared temperature sensor (Tleaf-IR-sensor) and the PSII fluorescence is analyzed by PAM-technique (the fiber MINI-PAM/F is positioned in a 60° angle to the leaf for PSII fluorescence analysis). The air enriched with humidity evaporated from the sample is then analyzed in a second analytic cell (Sample Analytics). The difference in water vapor concentration of incoming air and sample enriched air is the basis for the porometer parameter calculation (see chapter 10). With a second mass flow meter (Mass-Flowmeter-Out) the system provides information about the amount of air coming from the leaf chamber and thus about the system's airtightness. Finally, the air is released through exhaust.



Fig. 1: Pneumatic pathway of MINI-PAM-II/POROMETER

4 Components

4.1 Components

Porometer	MINI-PAM-II/POROMETER	
Leaf clip cable	PORO/AL	
Air-in buffer	PORO/B	
Gasket (5x) white	000246900414	
Gasket (5x) black	000246903214	
Dark Shield	000246901014	
Tripod Mount	000246901114	
Pore standard	PORO/POR	
Filter paper (20x)	PORO/FP	
Transport Case	PORO/T	
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4.2 Optional Components

CO ₂ -calibration kit (zero)	PORO/CAL
External LED Light Source	2054-L

5 Setup

The porometer is ready for immediate use. As MINI-PAM-II leaf clip, it only needs to be connected to the MINI-PAM-II. The instrument recognizes the porometer and provides the required operation elements. Together with the MINI-PAM-II it can be used in stand-alone configuration e.g. in field experiments and controlled by an external computer.

Please connect the leaf clip cable to the MINI-PAM-II/POROMETER and LEAF CLIP socket (#1) of the MINI-PAM-II instrument (see Fig. 2).



Fig. 2: Back panel of MINI-PAM-II Power-and-Control-Unit

Note: Great caution should be exercised to prevent dirt or foreign matter from entering the ports or sockets of the MINI-PAM-II. Do not force a plug into the wrong socket. Orientate each plug so that the red dot on the plug coincides with the red dot of the socket. Do not try to disconnect a plug by pulling at the cable. Disconnect plug by pulling at the rippled bushing of the plug.

To protect the gasket, the porometer is stored with an open leaf chamber when not in use. For this purpose, a black ring can be slipped over the handle and keeps the chamber open. When the porometer is in use, this black ring can be tucked away at the end of the handle.



Fig. 3: Black ring holding the chamber in an open position (A) and tucked away for measurements (B)

If there is a strong humidity gradient between the experiment location and the porometer storage location, it is recommended to store the buffer column at the experiment location or to switch on the flow ahead of the experiment so that the humidity buffer can equilibrate.

5.1 Mounting fiberoptics

For PAM-measurements, the MINI-PAM/F fiberoptic is fixed at an 60° angle to the sample area. The bending of the fiber is secured by the mounting aid. Already purchased fibers can also be equipped with the mounting aid. To do this, please attach the mounting aid as shown in Fig. 4 at a distance of 30 cm to the fiber tip. Please use the black Ø 1.4 cm ring to attach the mounting aid to the porometer (see Fig. 5)



Fig. 4: MINI-PAM/F mounting aid



Fig. 5: Rear mounting of MINI-PAM/F

The light intensity at the leaf area level is determined among other things by the optical geometry between fiber and sample. Therefore, the internal light list of the MINI-PAM-II should be calibrated in the initial setup process. To calibrate the internal light list please follow the procedure described in chapter 9.6.

5.2 Mounting dark shield

The porometer can measure stomatal conductance quickly and accurately in single point measurements under ambient light conditions. Additionally, the porometer can combine typical MINI-PAM-II measurement protocols such as Actinic+Yield, induction- and light-curves with its data. If the experiments require controlled light conditions, shielding from ambient light, such as for assessing of F_V/F_M after dark acclimation, the dark shield can be used for easier handling. It prevents the sample area from being exposed to ambient light. The dark shield is mounted to the fiber between sample area and fiber holder see Fig. 6.



Fig. 6: Mounting MINI-PAM/F and dark shield.

5.3 Black gaskets for needles

In addition to the standard white gaskets, 2 mm black gaskets are included in the scope of delivery. These are for special applications where the sample does not cover the entire leaf chamber. The black gaskets are attached to the top of the leaf chamber for better sealing of the cuvette.

Setup



Fig. 7: Black gaskets for sealing specific samples.

Note: if the sample area is not covered entirely the parameter leaf area needs to be adjusted (see chapter 7.4 Parameter – Leaf area).

To attach the black gasket, remove the protective layer from the adhesive surface (Fig. 8; A). With the porometer open, place the black gasket with the adhesive surface facing up on the white gasket (Fig. 8; B). Carefully close the porometer to ensure that the black seal adheres precisely to the surface of the leaf chamber (Fig. 8; C).



Fig. 8: Mounting the black gasket

5.4 Tripod Mount

For experiments with longer measuring times, the porometer can be used with a tripod. For this purpose, the tripod mount is attached to the side of the porometer with two screws. There is a ¼" thread in the lower part of this adapter.



Fig. 9: Tripod Mount and Porometer (A); mounted (B)

5.5 External LED Light Source 2054-L

For actinic illumination of the leaf the LED Light Source 2054-L can be used. It offers the ability to adjust the colors red, green, blue, and white independently. Thus, the spectrum of the actinic illumination can be set individually for each experiment. The 2054-L light source is mounted to the side brackets of the porometer and connected to the SYNC port of the MINI-PAM-II. Adjustment of light color and intensity is done either via the MINI-PAM-II menu "Light Sources"- "Light Panel Sett." or via WinControl-3.

Further information about the External LED Light Source 2054-L can be found in the MINI-PAM-II manual.

6 Taking Measurements

The MINI-PAM-II/POROMETER is equipped with sensor technology that enables the calculation of stomatal conductance and the determination of stomatal movement. These parameters can be combined with PAM-measurements of MINI-PAM-II instruments, to get detailed information about physiological aspects of photosynthesis.

This chapter describes the measurement procedure and gives useful tips on how to perform porometer experiments. All operating keys are described in detail in the next chapter.

Two measurement procedures can be distinguished with the MINI-PAM-II/POROMETER: One for the rapid determination of stomatal conductance and another for the recording of stomatal conductance and stomatal movement over multiple data points. These general handling tips apply to both measurement procedures.

Before starting a series of measurements:

- Setup the MINI-PAM-II/POROMETER (chapter 5)
- **Check the porometer baseline.** Close the leaf chamber of the porometer to purge both analytical cells with the same air. If there are differences in humidity

(e.g. dH₂O), please follow the maintenance "Pressuresensor calibration" and "Match" chapter 9.1 and 9.2.

- If you are interested in the **ambient CO**₂ level, please calibrate CO₂ sensor prior your experiments (chapter 9.3.)
- Charge the **batteries** or take a set of charged batteries ies with you. 6 AA (Mignon) rechargeable batteries (Eneloop 1.2 V/2 Ah) within the MINI-PAM-II typically provide power for more than 8 hours continuous porometer operation at maximum flow. Fluorescence measurements shorten the operation time. The most power consuming elements are the sensors for the geospatial data. Please turn GPS off, when not in use.

Positioning of the leaf clip:

• Orient the leaf side of interest toward the white gasket.

Air flows through the bottom of the leaf chamber. When the sample area is completely covered, a onesided measurement is made. If the upper leaf side is to be analyzed, the porometer can be rotated so that the top of the leaf faces the white gasket.

• Avoid positioning onto the veins of the leaf. Mechanical force on the leaf vascular structure can affect the measurement results. If the vascular structure is pronounced and therefore the surface is uneven, please check seal. The outgoing flow should equal the incoming flow. You can adjust the mechanical force of the clamping mechanism with the slider.

• Fully fill the sample area with the leaf or adjust the leaf area parameter.

The standard leaf area is 0.79 cm². If the sample does not cover the complete leaf area the parameter leaf area needs to be adjusted in the parameter submenu of the porometer settings (page 38).

 For measurements in ambient light conditions match light sensor's light conditions to those of the leaf.

Do not cover your light sensor by your leaf. Make sure that the light sensor is only shaded when the leaf sample is shaded, too. (Further information page 35 f)

• Avoid measuring rain- or dew-wet leaves. Moisture on the leaf surface can lead to overestimation of stomatal conductance. Please dry wet leaves carefully and increase measuring time.

Workflow for rapid assessment of stomatal conductance:

- Scroll to the "Porometer" window in the MINI-PAM-II display. Decide to use auto stability determination or manual control (default setting is the measurement with stability determination, page 32)
- 2. Insert the leaf sample.

- If you are not using the auto stability determination, please wait until the system has stabilized and the parameter displayed on the graph has plateaued.
 If you are using the stability determination, you can immediately go to 4. the system will evaluate the stability before executing the triggered measurement.
- 4. Press SAT on the MINI-PAM-II display or the porometer's red button on the Porometer to execute a measurement including Y(II).

Or

Press Save on the MINI-PAM-II display or the porometer's black button for porometer data only.

5. Release the leaf and go to the next sample.

Workflows for measurement sequences can vary. Here are some hints for optimization:

- To save battery power, the **GPS** for geospatial data acquisition can be switched off after the first measurement of a sample.
- The **stability determination** is only applied to the first data point of a MINI-PAM-II standard protocol like actinic + yield, light-curve, induction-curve or clock. All subsequent measurements will record the current state and are performed without delay.

- Measurement protocols often start with the assessment of F_V/F_M under dark acclimated conditions. The **dark shield** can be used to easily shield the leaf sample from ambient light.
- The porometer can be mounted on a **tripod** for convenient use over longer periods of time –using the tripod can help to maintain the original orientation of the leaf and to minimize mechanical impact.
- For experiments investigating the influence of light conditions, we recommend the External LED Light Source 2054-L for variable actinic illumination and the Miniature Spectrometer MINI-SPEC/MP for characterizing the ambient light conditions.

7 Touchscreen Operation

This chapter first gives information about touchscreen operation, then about WinControl-3 operation using an external computer.



The MINI-PAM-II/POROMETER is designed for field use and can be easily operated using the build-in touchscreen of the MINI-PAM-II.

The porometer functions are available on a new top-level window, the "Porometer" window. It will appear in the MINI-PAM-II menu between the top-level windows "Primary Data" and "Quenching Analysis", when a porometer is connected to the MINI-PAM-II. It contains all porometer-specific functions and a link to the corresponding settings.

Titel	GPS Power	Mark SAT
Data and Graph		
		•
		MENU
Action I	Keys	MEM
Informa	ation line	

All top-level windows, including "Porometer", have a similar structure. At the top is the title bar, which also contains the power indicator and the currently used marker letter. In the middle are the data and

graphs, and below is a bar of action keys. In the bottom is the information line. The side panel provides a button e.g., for saturation pulses (<u>SAT</u>), navigation keys, access to the main menu, and memory or an action key, e.g. for actinic light. With a porometer connected to the MINI-PAM-II, all SAT pulse measurements will also include porometer data.

7.1 Porometer windows, data and graph



The middle part of the "Porometer" window is divided into two parts: the upper part displays the measurement graphically; the lower part displays the data numerically. The win-

dow shows either the stomatal conductance g_s , the total conductance g_t , or the difference in water vapor dH₂O. This can be selected in the porometer settings (chapter 7.4).

It is possible to limit the numerical values to two enlarged parameters, this basic view can be switched on in the porometer settings (page 34). To view more porometer parameters simultaneously, the detailed view of the numerical data "Porometer Values" can be used. By touching the "Porometer" window in the center, the

Porometer Values 🛡 95 A	SAT
gs* 87.4 gt* 85.5 E 0.975 VPD 11.40	FLIP
dHzU 0.761 HzUun 11.444 Match 0 HzOout 12.205 Tleaf 196 Press 9760	
CO₂ 587 Flow 100/100	MENU
Sett. Flush Pump Save 2023-11-20 9:39:49	BACK

display changes from the subdivided view to a window in which all relevant parameters are listed numerically. A similar function is already known from the "Induction Curve",

"Light Curve" and "Recovery" window of the MINI-PAM-II. The "Porometer Values" window displays the current porometer sensor data and calculated parameters. In this display the sidebar has changed to a <u>FLIP</u> key instead of the arrows. Touching

Geospatial Data ♥ 95 A 49.656534N 11.097957E	SAT
SAT 14DOP - Height 334	FLIP
Leaf Az. 275 Sun Az. 133	
A.o.I. 86 Inc. 7 6	MENU
Sett. Flush Pump Save 2023-11-20 9:39:56	BACK

<u>FLIP</u> gives access to the current geospatial data. The <u>BACK</u> key or touching the center of the window again will go return to the combination of graphical view and some numerical

values in the "Porometer" window.

7.2 Porometer action key bar



The action key bar contains four buttons. From left to right they are <u>Sett.</u>, <u>Flush</u>, <u>Pump</u>, and <u>Save</u>.

<u>Sett.</u> is a link to porometer relevant settings and is described in detail in chapter 7.4.

<u>Flush</u> The flush function is used to quickly purge the leaf chamber with ambient air e.g., after measuring heavily evaporating leaves. Pressing this button will increase pump power for 15 seconds. During this time no porometer readings will be displayed. While the flush function is active the key has a dark background. The flush function can be interrupted by pressing the <u>Flush</u> button again.

<u>Pump</u> is the key for turning the pump on and off. When the pump is on, this key has a dark background. The pump is controlled at the pump intensity set in the settings menu (see Sett. chapter 7.4) with a flow rate of 40-200 μ mol s⁻¹. When the porometer is turned on, the pump is automatically activated.

<u>Save</u> stores the porometer data **without** triggering a saturation flash. This button is called Store in WinControl-3. In general, if a porometer is connected to the MINI-PAM-II, all saturation pulse measurements will automatically store a porometer data set with the executed saturation pulse. Even if it is triggered by the SAT-, Fo,Fm-button from another MINI-PAM-II window or executed during a program routine like light-curve and induction-curve.

7.3 Porometer side panel



The five buttons of the porometer window side panel are the same as in other MINI-PAM-II top level windows. They are SAT, up and down navigation, MENU and MEM.

<u>SAT</u> This button is used to trigger a MINI-PAM-II measurement. If a porometer is connected, a set of porometer data is always included in the saturation pulse analysis. A MINI-PAM-II saturation pulse measurement takes 2 seconds. During this time the <u>SAT</u> button will be dark. If the beeper is on, a beep will indicate the start and end of the measurement. When the porometer stability criteria are applied (chapter 7.4), the instrument waits with the measurements until the porometer values are validated as stable. During this time, the background of the <u>SAT</u> button remains dark and the beep sounds at regular intervals.

The <u>up and down navigation keys</u> scroll through the main level windows, menu items, or memory window data, depending on the context in which they are used. <u>MENU</u> gives access to the MINI-PAM-II main menu. Many porometer settings are also available in the action key <u>Sett.</u> (chapter 7.4). The main menu items relevant to the porometer are explained in more detail in chapter 7.5.

<u>MEM</u> opens the memory window of porometer measurements. This window displays all porometer measured data, an-

Porometer Memory 80 I No. 1663	SAT
gs 42.5 gt 42.1 E 0.642 VPD 15.24	
Match 39 HzOout 15.716 Tleaf 238Press 9617	\bullet
CO₂ 1869 Flow 100/97 ETR 75.0 PAR 420	MENU
Y(II) 0.425 2023-10-26 13:44:34	BACK

notated by No., date and time. When a saturation pulse was performed also Y(II) and ETR information is given. Please use the navigation keys to scroll through the data. Back re-

turns to the porometer window. The stored porometer data is also available in the main menu memory (chapter 7.6).

7.4 Sett./Porometer Settings



In the Porometer window, on the left side of the action key bar, is a special action key: <u>Sett.</u> It is a link to all porometer related settings. The settings can also be set via <u>MENU</u> in the

main menu, but there these settings are divided into different subcategories like "Light Sources" and "Sensors/Porometer".

Therefore, please use the <u>Sett.</u> key for an easy access to all porometer relevant settings.

The porometer settings include:



- Pump setting,
- Stability criteria for measurements,
- Choice of chart display
- On/off switch of basic view of the porometer window,
- Used PAR, origin of PAR value (external sensor, internal sensor or light list)
- Parameter: Leaf Area, Emissivity, Pressure and CO₂, GPS, Cal Orient. Sens.
- Match procedure

Pump

The pump can be set to provide a flow of 40 – 200 μ mol s⁻¹ in 20 μ mol s⁻¹ increments. Please use the navigation keys and <u>SET</u> to change the flow rate.

Please consider that at higher flow rates the air flows faster through the leaf chamber. Therefore, the difference in water vapor concentration is less pronounced than with the same sample at lower flow rates. High flow rates are recommended only for samples with high transpiration rates. Standard pump flow setting is 100 μ mol s⁻¹.

Stability

The stability mode facilitates the porometer measurements, especially in the field. In stability mode, the slope of g_s and g_t over a given time are evaluated. After triggering the measuring (clicking <u>SAT</u> or <u>Save</u> on the MINI-PAM-II or using the red or black button on the porometer), the system logs the values internally until the set stability criteria are met. As soon as the porometer values are stable, the actual measurement is performed, and the data is stored.

If a program protocol, e.g., a light curve or induction curve, is started the stability criteria are applied only to the first measurement. All sequential measurements will be executed as given in the respective protocol.

Stability gives access to the submenu for selecting and applying stability criteria to measurements.



The first line of the stability criteria window is the on/off switch for the stability mode. When the stability mode is activated, the following stability criteria are applied to the meas-

urements. To switch off the stability mode, please use the navigation keys to select the stability mode line and press <u>SET</u>, the label will change to off.

For the evaluation of stability, the change in conductance (g_t, respectively g_s) is taken into account. When the change in slope

of conductance is smaller than the value listed in delta g_t over the given period (Interval in seconds), the stability criteria are fulfilled, and the measurement stored.

Whereby:

- Delta g_t [mmol m⁻²s⁻¹] is the limit of slope; 0.2 20
- Interval (s) defines the time period in seconds over which the slope is determined; 3-10 seconds
- Timeout (s) defines the maximum time period 20 300 seconds until the measurement is executed

When the stability mode is activated unstable defined g_s and g_t values are indicated with $^+$ (g_s^+ ; g_t^+). When the stability criteria are met, the $^+$ disappears. After triggering a measurement, the MINI-PAM-II gives an acoustic signal in form of a rhythmic beep as long as the system is waiting for the stability criteria to be met and the measurement has not yet been executed. The stability mode includes a timeout. If the system does not become stable within the set timeout period, a measurement is performed even under unstable conditions. The stability evaluation can be interrupted manually by pressing the <u>SAT</u> or <u>Save</u> key again. Then no measurement is executed! The evaluation time of the stability criteria begins as soon as the leaf chamber is closed, and the outgoing flow exceeds 75% of the incoming flow rate.

Chart

The porometer window in the MINI-PAM-II displays one of the following parameters in the chart and numerically below it:

 g_s , the stomatal conductance (mmol $m^{-2} s^{-1}$)

 g_t , the total conductance (mmol m⁻² s⁻¹)

 dH_2O the difference in water vapor (mmol mol⁻¹)

Chart Value	
g <mark>s Chart X</mark> gt Chart dHaO Chart	
	SET
	EXIT

The parameter to be shown, can be selected in the Chart Value submenu and is marked with X, e.g., here it would be g_s .

Basic View



In the basic view, the numerical display of the porometer window is limited to two values, which are displayed larger. When the basic view is activated, only the selected parame-

ter of g_s , g_t or dH_2O and the Flow (%) are given. The outgoing flow is indicated in % of the incoming flow rate, to check if the leaf chamber is closed well.

Used PAR



This submenu offers the possibility to select the origin for PAR. Depending on the experimental setup, the internal PAR sensor of the MINI-PAM-II, the external sensor of the

porometer, a combination of both, or just the light list, can be selected as source for the PAR parameter. This menu topic is identical to parts of the "Sensor Settings" menu of the MINI-PAM-II. The selected PAR value will be used in all subsequent calculations of photosynthesis parameters. The default setting for porometer applications is "Ext. PAR Sensor" on and "Add int. PAR" on.

Measurements in ambient light ± actinic light



The porometer standard condition: "Ext. PAR Sensor" on and "Add int. PAR" on,

should be selected for measurements under ambient light conditions. In this configuration, the ambient light condition is measured by the external light sensor next to the leaf chamber, and the PAR value of the internal light sensor is added when actinic illumination is applied onto the sample area by the MINI-PAM-II fiber.

Without actinic illumination from the MINI-PAM-II, the leaf in the sample area is only exposed to ambient light. This is determined by the external PAR sensor. The internal PAR sensor is positioned facing the beam path and measures the actinic light intensity given by the MINI-PAM-II. If the actinic illumination is off, the value of the internal PAR sensor is 0. The used PAR value then corresponds the PAR value of the ambient light sensor. However, when the actinic light is on, the leaf is exposed to increased light intensity due to the illumination via the MINI-PAM-II fiber. The external light sensor, located at the side, is not affected by this illumination. To determine the PAR at the leaf level the used PAR must be summed. Therefore, the actinic light intensity given via the MINI-PAM-II fiber is determined by the internal light sensor and added to the ambient PAR value.

Attention should be paid to the position of the external light sensor. If the leaf is in the shade, ensure the light sensor is shaded. If the leaf is in the sun, ensure the light sensor is also in the sun. A mismatch in light conditions may lead to significant error in ETR calculation.

Measurements with Dark Shield

Used PAR Ext. PAR Sensor off (Add int. PAR becomes irrelevant), should be selected for measurements with the Dark Shield.

The Dark Shield (page 34) shields the entire sample area from ambient light. This is very useful for the simple determination
of F_V/F_M and is therefore often used in classical saturation pulse experiment protocols. The actinic illumination is generated only by the MINI-PAM-II. Thus, using the Dark Shield, the ambient light must be ignored and the PAR values from the internal light sensor are used to calculate ETR.

Measurements using External LED Light Source 2054-L

Used PAR Ext. PAR Sensor off (Add int. PAR becomes irrelevant) should be selected for measurements using External LED Light Source 2054-L.

The External LED Light Source 2054-L provides actinic illumination with freely adjustable light colors. Before using the 2054-L, the light flux characteristics should be adjusted using the light calibration procedure (chapter 9.6). In an automatic procedure, all LEDs are set to meet the internal light list requirements. Actinic illumination by the external LED light source partially illuminates the ambient light sensor next to the sample area. Therefore, the external PAR sensor should be turned off for this application. Since the internal PAR sensor is not within the optical beam of the external LED light source, the used PAR is obtained from the light list when the LED Light Source 2054-L is connected to the MINI-PAM-II.

Parameter

The parameter submenu provides settings for leaf area, emissivity for leaf temperature determination, and calibration routines for pressure and CO₂-detection.

<u>Leaf area</u>

The sample area of the porometer is 0,79 cm². For special applications where not the complete sample area is covered by the sample (e.g. measurements of conifer needles) the leaf area can be altered for correct parameter calculation (please read chapter 10).

Emissivity

An important factor in temperature measurement with IR sensors is emissivity. This is the ratio of the energy emitted by the surface of a material to the energy emitted by a perfect radiator (black body). The emissivity factor for the leaf temperature sensor of the porometer can be adjusted in this settings menu. The default value for the leaf temperature measurement is set to 0.9800 (López et al; Scientia Horticulturae 137; 2012)

<u>Pressure</u>

Submenu providing the calibration routine for the pressure sensors. Please read chapter 9.1 Pressure-sensor calibration.

<u>CO</u>₂

Submenu providing the calibration routine for the ambient CO₂sensor. Please read chapter 9.3 CO₂-sensor calibration.

<u>GPS</u>

GPS on/off switch. If no geospatial data is necessary, turning off GPS sensor saves a significant amount of power.

When the GPS sensor is switched on, the GPS icon is present in the title line, next to the battery indication.

 Porometer
 Image: Set of the set

Cal. Orient. Sens.

The spatial position sensors (accelerometer, magnetometer, gyro sensor) automatically check their initial values when in use. If the room position is not recognized by the porometer, the initial data of these sensors can be calibrated manually. The calibration routine is described in chapter 9.4.

Match

Match is used to align the sensors for humidity detection of the analytical sample cell to the analytical reference cell. The sensor specific differences in water vapor concentration detection are recorded in mmol mol⁻¹ and listed in the report. For more information, please read chapter 9.2.

7.5 **Porometer topics in the Main Menu**

Most of the porometer settings can be accessed through the settings link <u>Sett.</u> on the porometer window. Two functions that are only available in the main menu is the clock function "Porom. Only" in the submenu of Program/Clock, and "Reset Porometer" in the Reset submenu.

The porometer settings summarized under Sett. can also be managed in the Main Menu. Here they are embedded in different submenus. This chapter will guide through these settings in the main menu of MINI-PAM-II.

In the main menu following submenus contain porometer relevant information or functions:

Main Menu	
PAM Settings -> Light Sources -> Program/Clock ->	
Sensors -> MINI-PAM-II Sett>	▼
Info -> Reset ->	SET
	EXIT

- Program/Clock
- Sensors
- MINI-PAM-II Sett.
- Memory
- Info
- Reset

Program/Clock

Program/Clock opens the submenu for program routines and automated measurements of MINI-PAM-II standalone operation e.g., light curves or the clock. The "Clock" triggers repetitively an event at a defined interval. The interval is specified in "Clock Interval" and the event in "Clock Item". With the porometer connected a new clock item is available. "Porom. Only". This executes repetitively storing of porometer data without executing a saturation pulse (like Save on the porometer window or Store in WinControl-3).

Sensors



The sensor menu contains the settings for the MINI-PAM-II sensors. Settings for light measurement as well as settings for the sensors of various MINI-PAM-II accessories can

be made here.

The first two lines are used to select the origin of the PAR value used for calculations, which can also be found within the used submenu of the porometer settings window.



The Ext. PAR Sensor submenu is for the choice of external PAR sensor when more than one external PAR sensor is connected to the MINI-PAM-II (e.g spectrometer and

porometer). If only one accessory is connected the MINI-PAM-II will automatically select the corresponding external PAR sensor.



The Porometer submenu is to adjust settings of the porometer. This settings menu is also available via the shortcut <u>Sett.</u> in the porometer window.

MINI-PAM-II Sett.

Calculating the sun position of the sun angle of incidence requires the correct date and time. The porometer's built-in GPS sensor provides accurate timing when operational (filled symbol displayed in the title line).



The internal clock of the MINI-PAM-II shows the local time. The offset to the UTC time can be set in the MINI-PAM-II Sett. submenu: Time/Date. Here you can either enter the offset to UTC directly, e.g. UTC Time (+/-) 2:00 for +2 hours offset for the local time of Effeltrich, or use the option to enter the local time manually in the submenu Local Time/Date.

The internal clock of the MINI-PAM-II is automatically corrected



to UTC time (± offset) if the time difference is less than 15 minutes. If there is a time difference of more than 15 minutes between GPS sensor UTC time (± given offset) and MINI-PAM-II clock time a message is

displayed in the info line.

Memory

is described in chapter 7.6.

Reset

The MINI-PAM-II retains its internal memory settings to be restored the next time the instrument is used. The default values of these settings can be reset in the Reset submenu, which contains three submenus: Touchscreen Operation



1. Reset Settings: MINI-PAM-II default settings (please read MINI-PAM-II manual for more details)

2. Reset Sys. Set.: MINI-PAM-II system settings (please read MINI-PAM-

II manual for more details)

3. Reset Porometer sets porometer relevant settings to default or system values. More details are listed in Tab. 1.

Tab. 1: Porometer Dela	Default Settings	et- Current Setting
	ting	(saved and restored)
Pump Flow	100 µmol mo	ol ⁻¹ Yes
Stability		Yes
Stability Mode	On	
Delta g _t	0.5 mmol m ⁻²	² S ⁻¹
Interval (s)	2 seconds	
Time out (s)	180 seconds	
Chart	g _s Chart	Yes
g _s Chart		
g _t Chart		
dH ₂ O Chart		
Geospatial Data	on	Yes
Basic View	off	Yes
Used PAR	Ext. PAR Sens	sor on Yes
Ext. PAR Sensor on	Add int. PAR	on
Add int. PAR on		
Parameters		
Leaf Area	0.79 cm ²	Yes
Emissivity	0.9800	Yes
Pressure	Sensor Data	No
CO ₂	Sensor Data	No
Match	0 mmol mol ⁻¹	Yes

Info

Info contains hardware and software information and is divided into three sections. Sections "MINI-PAM-II" and "Firmware" give information about MINI-PAM-II hardware version, serial number, battery status, and firmware version. Section two "Sensors" gives information about accessories connected to the MINI-PAM-II. The porometer serial number is listed here.

7.6 Memory

Memory		
Datasets	÷	
New Record Mark	-> A	
Record No. Measurement No.	7 1739	\bullet
		SET
		EXIT

memory Datasets.

 Stored Dataset
 Image: Stored Dataset
 Im

The combined analysis of porometer data, geospatial information and saturation pulse analysis provide a comprehensive set of parameters. These are consolidated in the main menu

The <u>FLIP</u> key in the top right corner can be used to switch between Saturation Pulse analysis data, porometer analysis data and geospatial information data of one measuring

point. These related data sets have the same No., date and time. The up and down arrow keys can be used to scroll through previous or subsequent measuring points of the chosen data set. If a measuring point lacks this data set it will be skipped. For example, data sets No. 1 and 5. contain porometer data, whereas data sets 2-4 were measured with the MINI-PAM-II without porometer. In the porometer data view, you can use the arrow keys to navigate from No.1 to No.5 and back, data from measurement No.2-No.4 are skipped. In the saturation pulse data view, also the measurements without porometer data are displayed. Use the arrow keys to scroll No.1-No.2-No.3-No.4-No.5.

8 WinControl-3 Operation

Like the MINI-PAM-II itself, the MINI-PAM-II/POROMETER can be controlled by an external computer running the WinControl-3 software. This manual is limited to the WinControl-3 functions for the porometer. Other WinControl-3 functions are described in the MINI-PAM-II manual.

8.1 WinControl-3 Status panel

When a porometer is connected to the MINI-PAM-II, the porometer functions will be extended in WinControl-3. A new porometer field appears in the Status panel next to the SAT Pulse/Chart field.



In this new porometer field the pump can be switched on and off and the flow level can be adjusted (40 μ mol s⁻¹ to 200 μ mol s⁻¹).

The Store button stores the porometer data without executing



a saturation pulse. In the MINI-PAM-II standalone operation this command is assigned to <u>Save</u> or the black porometer button. As in the MINI-PAM-II standalone configuration, the execution of SAT or Fo,Fm will always store a porometer data set.

The flush function increases pump power for 15 seconds to purge the leaf chamber. No porometer online values will be displayed in the meantime.



The online field also expands when the porometer is in use, displaying online

data from the porometer sensors and important calculated parameters. The online porometer values are marked with a blue background. Parameters which are also available with other MINI-PAM-II leaf clips stay at the designated online data fields e.g. leaf temperature is listed in Temp*, used PAR in PAR* and ambient humidity in AHum*. According to WinControl-3 designations, the * indicate that these data are online data. This * is not to be confused with the parameter indication ⁺ in the standalone configuration, which indicates unstable conditions during stability evaluation (page 32). Stability evaluations can be performed in WinControl-3 operation but are not indexed in the online values. The acoustic signal during the stability evaluation via the MINI-PAM-II is possible at any time. Stability determination can be canceled by pressing the SAT or Store key on the MINI-PAM-II.

8.1 WinControl-3 Porometer Values

WinControl-3 determines which parameters are displayed in

		Options	View Side	bar
Values	Chan.			
SAT-Puls	e X Fm' Ten ETR ETR Q NI	np -F. N PQ		*
Y (NO Fo,Fm-M Fo Fo Fo Fv/Fn) Y easure Fm	(NPQ)		
Sensors S AHun Oxyg. X1	Stored n 			
Poromet gs E dH ₂ O H ₂ O in Flow i Matcl Press	er Stored □ g ☑ V 0 C n □ H in □ Fl	i t PD O2 20 out low out		
GPS Long. Heigh DOP Orientat	Lat t Sta Sat	tus		
Head Head Roll Leaf Sun A A.o.I.	Pit Az. Le z. Su In	tch eaf SI. In EI. c. %		~

the graphs or listed in the report based on the "Values" in the "Sidebar". The values are grouped into different topics in the sidebar. New groups are displayed with the porometer connected: Porometer Online, Porometer Stored, GPS and Orientation. Again, parameters which are also available with other MINI-PAM-II leaf clips stay at the designated data group, e.g. leaf temperature (Temp) and PAR in the SAT-Pulse group, and ambient humidity (AHum) in the Sensors group.

The data value activated by the corresponding checkbox will be displayed. If an entire group is to be displayed, you can activate these values by double clicking on the group name.

8.1 WinControl-3 Porometer Settings

When the porometer is connected, the Settings page displays the porometer settings control panel. Porometer-specific settings and calibration procedures can be done here.

Porometer			
Pump	100 🔺 🔻		
Emissivity	0.9800 🔺 🔻		
Leaf Area	0.79 🔺 🔻		
N	fatch		
Ca	librate		
GPS			
Stability:			
Active			
Delta gt	0.5 🔺 🔻		
Interval(s)	2 🔺 🔻		
Timeout	180 🔺 🔻		

The top menu item is Pump, which corresponds to Pump in the status bar and determines the pump performance. The Flow can be set from 40-200 μ mol s⁻¹ in increments of 20 μ mol s⁻¹.

Emissivity is a leaf temperature sensor setting and refers to the efficiency of a material's ability to emit thermal radiation. It is a dimensionless value between 0-1, standard value is 0.9800. More information on page 38.

If the sample area is not completely covered, the Leaf Area can be modified here (page 38. The leaf area is used in the calculation of porometer parameters (chapter 10).

Match is the procedure to match the humidity sensors. Please read chapter 9.2.

Calibrate opens a calibration window for the pressure sensor calibration procedure (chapter 9.1). CO_2 calibration is done in standalone operation (chapter 569.3).

The GPS checkbox enables or disables the GPS receiver. This sensor is power consuming and can be turned off to conserve battery power.

Stability: If the Active checkbox is checked, the stability determination analysis is performed. After a measurement is triggered by Fo,Fm SAT or the Store button, the system automatically waits for the porometer data to meet the stability criteria listed. For example, if g_t changes less than 0.5 mmol m⁻² s⁻¹ in two seconds, the measurement is performed, and the data are stored. If the stability criteria are not met within the time defined under Timeout, the measurement is executed after this maximum waiting time, even if the values are not stable. The stability determination can be interrupted by pressing the SAT or Save key on the MINI-PAM-II. In this case no measurement will be performed.

9 Maintenance

9.1 Pressure-sensor calibration

Three pressures are considered in the calculation of the porometer (chapter 10): the pressure within the reference analytics, the sample analytics, and the pressure within the leaf chamber. For this purpose, two pressure sensors have been installed in the porometer (in both analytical cells). The pressure within the leaf chamber derives from these pressure sensors.

In the porometer settings these sensors can calibrated to a reference pressure.

Please switch off the pump, open the leaf chamber, and choose the pressure sensor calibration window in the MINI-PAM-II. Porometer Settings \rightarrow Parameters \rightarrow Pressure.



The upper line of the Pressure Window shows the Ref. Pressure, which is the raw data from the output pressure sensor at the time the porometer was turned on. Ref. Pressure is the value to which both pressure sensors are set during the calibration process. The Ref. Pressure can be altered manually when the value from an external pressure sensor is accepted. This can be done by adjusting the Ref. Pressure value with the arrow keys.



The calibration for the pressure sensors is initiated in the second line of the Pressure window. A confirmation prompt appears. <u>Yes</u> sets both pressure sensors to value specified

as Ref. Pressure.

If the pressure sensor difference exceeds a physical threshold, the MINI-PAM-II will issue a pressure sensor warning.

The pressure sensor calibration can also be done in WinControl-3. Calibrate in the porometer field of the Settings tab



opens the pressure sensor calibration prompt.

Please turn off the pump, open the leaf chamber and set the pressure value to the desig-

nated pressure. Press Calibrate to set the porometer pressure sensors to this calibration value.

9.2 Match

Matching is the process of reconciling the differences in humidity detection by the sensors between sample analysis cell and reference analysis cell, while the same air passes through both cells. This adjustment serves as a zero adjustment of the humidity sensors to increase the measurement accuracy. It should be performed with care before a series of measurements. Within a measuring sequence a new adjustment is not necessary. But if match is also to be carried out between measurements, care must be taken to ensure that the chambers are flushed with ambient air for a sufficiently long time.



Please close the **empty** porometer leaf chamber Switch on the pump and make sure that the leaf chamber is properly closed (the incoming flow equals outgoing flow)

- Wait for the system to get steady (recommendation is at least 5 minutes)
- Choose <u>Match</u> using the navigation keys in the "Settings" window, press <u>SET</u> and confirm by choosing <u>Yes</u> (using the navigation keys and <u>SET</u>).

The difference in water vapor concentration between sample and reference is stored as "Match" value (mmol mol⁻¹). This

sensor offset value is taken into account in the determination of dH₂O and H₂O_{out} (see Chapter 10).

9.3 CO₂-sensor calibration

The ambient CO₂ sensor determines the concentrations in the surrounding air. Calibration of this sensor against fresh air can be done without any additional equipment. The MINI-PAM-II with the connected porometer have to be outside in fresh air. In the Porometer Settings menu under Parameters, CO₂ leads to the corresponding submenu.



The designated CO₂ concentration is set under Ambient Level. Then the calibration procedure can be started in the Cal.Ambient. submenu.

Please stand back so as not to interfere with the measurement. The calibration takes 5 minutes. The lowest measured CO₂ concentration during this time is then calibrated to the set

ambient CO_2 value. The CO_2 calibration window can be closed with EXIT.

9.4 Calibration of Orientation Sensors

The gyroscope, accelerometer and magneto scope automatically calibrate themselves when in use. The initialization values can be calibrated manually using the Cal. Orient. Sens. submenu within the porometer "Parameters" window. The calibration routine starts when entering this submenu, it can be aborted with <u>EXIT</u>.

Orient.	Sensor	Calib.		
Gyro	— He	ead 🛛	Ø	
Accel	· Pi	tch	1	
Mag	י Re	oll	0	
Accel:	Hold th	e devic	e	
in va	rious p	ositions.		
for s	5s, move	slowly		
Mag: N	love the	device		
in a ho	prizontal	8 figur	e.	EVIT
				EVII

Accelerometer: Hold the device in various stable positions for 5 seconds each. Move slowly between two stable positions.

Magneto scope: Move the device in a

horizontal eight.

Gyroscope: Place the device in a single stable position for a few seconds to allow the gyroscope to calibrate.

The progress bars show the progress of the calibration. If all sensors have recognized their initialization parameters <u>SAVE</u> appears in the sidebar. Press <u>SAVE</u> to store these new calibration parameters.

Press <u>EXIT</u> to leave the calibration routine. The previously stored calibration values are used to initialize the sensors.

9.5 Cleaning

The basic cleaning and maintenance procedures for the porometer include:

- Cleaning of the leaf temperature sensor. The leaf temperature sensor is located at the bottom of the leaf chamber. To remove dirt particles that may contaminate it, use a cotton swab moistened with either distilled water or isopropyl alcohol.
- Cleaning of the leaf chamber window. For cleaning, please use a cotton swab moistened with either distilled water or isopropyl alcohol.
- Exchange of the Air-In Filter. If this filter is clogged, it can be replaced by a commercial 15 mm Ø glass fiber syringe filter with 1-2 μm pore size.

9.6 Light Calibration procedures

The porometer is equipped with a Micro-Quantum-Sensor. This external light sensor can be used for the calibration of the



internal light sensor, the internal light list of the MINI-PAM-II, and the light flux characteristics of the External LED Light Source 2054-L.

Please move the light sensor to the

center of the sample area.

The light calibration procedure is executed in standalone operation on the Actinic Light List window by pressing CAL.

Calibrate Light List	
using ext Sensor -> using int. Sensor ->	
	►
	SET
	EXIT
Calibrate Light List?	-
Calibrate Light List? No <u>Ves</u> Place the ext PAR	
Calibrate Light List? No Yes Place the ext. PAR sensor in the sensing area!	▲ ▼
Calibrate Light List? No <u>Ves</u> Place the ext. PAR sensor in the sensing area!	▲ ▼ SET

To calibrate the light conditions at the sample level, the external sensor should be used for actinic light calibration. <u>Using ext. Sensor</u> leads to the submenu where the automatic calibration procedure is started.

For calibrations with the MINI-PAM-II only (without an external light sensor from a porometer or another leaf

clip), the submenu using int. light sensor can be used for light calibration. In this case only the internal light sensor is used to set the actinic LEDs.

9.1 Gasket

The porometer leaf chamber needs to be airtight. To seal the leaf in the leaf chamber, there is a white silicone gasket in the lower part of the leaf chamber. Sustained pressure can affect the elasticity of this foam, therefore please store the unused porometer with an open leaf chamber. To keep the chamber open, please slip the black ring (Ø 31 mm) over the handle. When the porometer is in use, this black ring can be tucked away at the end of the handle.

If the elasticity of the foam is not given anymore and the leaf chamber troubles with air loss (Flow_{out} does not reach Flow_{in} values) please exchange the foam. Spare foams are within the delivery package or can be ordered with article number: 000246900414



Fig. 10: Black ring holding the chamber in an open position (A) and tucked away for measurements (B)

The black gaskets are for use in special applications where the sample does not cover the entire sample area (see chapter 5.3)

9.1 Pore Standard PORO/POR

With the pore standard PORO/POR the functionality of the porometer can be easily checked. It consists of two circular discs with a moist filter paper placed between them. The disc offers six positions, labeled 0-5 based on the number of pores in the lower disc. The g_s , g_t and dH_2O values measured with the porometer increase as the number of pores increase.

Please wet one sheet of filter paper (PORO/FP) with water at ambient temperature. Remove excess water by gently pressing the filter paper between two absorbent surfaces (e.g. paper towels). Store the moist filter paper in a sealed bag for approximately 10 minutes. The damp filter paper can now be inserted into the PORO/POR for measuring position 0-5.



Fig. 11: Preparation of PORO/POR

In accordance with the equation from Chapman and Parker describing diffusion through a concentric cylinder (Agricultural Meteorology. 23; 9-20; 1981) the following equation was used for the calculation of the theoretical total conductance of the pore standard.

EQ: 1

$$R_{(n)} = \frac{n}{A} \left(\frac{1}{r D}\right) \left(\frac{h}{\pi r} + \frac{1}{4}\right)$$

Whereby

R(n) = Resistance of the pore standard depending on the number of pores [s mm⁻¹]

n = number of pores

A = sample area (79 mm^2)

r = pore radius (mm)

D = water diffusion coefficient at 22°C and 98.12 kPa $[mm^{-2}s^{-1}]$

h = height of the pore [mm]

at 22 °C; 98.12 kPa, and 48% ambient humidity gives the following total conductance (g_t):

Numbers of pores	g _t [mmol m ⁻² s ⁻¹]
1	19
2	38
3	57
4	76
5	95

Minor deviations are possible due to the manufacturing tolerance of the PORO/POR and moist filter paper properties.

9.2 Signal LED

On the left side of the porometer is a signal LED indicating the operational status as listed in the following table.

Tab. 2: Signal Code of LED

LED action	Status
Flashing green	Normal operation
Continuous green	Communication from MINI-PAM-II to MINI-PAM- II/POROMETER interrupted. This happens tem- porarily during firmware update of MINI-PAM-II
Flashing red	Error: contact Walz.
Continuous red	Firmware update of MINI-PAM-II/POROMETER

10 Calculations

10.1 Parameters

The MINI-PAM-II/POROMETER is an open system porometer. An airstream with a constant flow rate passes a defined area of the sample. Sensors upstream and downstream of the leaf chamber detect the water vapor released from the leaf. A noncontact infrared thermometer records the leaf temperature. The following table lists all measured and calculated parameters.

Tab. 3: Parameters

Value	Description	Unit
Area	Sample area 1 cm Ø; 0,79 cm ²	cm ²
H_2O_{in}	Upstream H_2O mole fraction, air entering the cuvette	mmol mol ⁻¹
H_2O_{out}	Downstream H_2O mole fraction, air leaving the cuvette. taking sensor offset (Match) into account	mmol mol ⁻¹
dH ₂ O	Difference in H ₂ O mole fraction, up- stream and downstream of the leaf chamber,	mmol mol ⁻¹
Match	Offset of H ₂ O mole fraction between hu- midity sensors	mmol mol ⁻¹
Press.	Leaf chamber barometric pressure	kPa
Flow in/out	Air flow into/from leaf chamber	µmol s ⁻¹

Calculations

Temp;	Leaf temperature	°C
CO ₂	Ambient CO ₂ mole fraction	µmol mol ⁻¹
E	Transpiration rate	mmol m- ² s ⁻¹
VPD	Vapor pressure deficit between object (leaf) and air	Pa kPa ⁻¹
gt	Total water vapor conductance	mmol m ⁻² s ⁻¹
gs	Stomatal conductance	mmol m ⁻² s ⁻¹
g _b	Boundary layer conductance	mmol m ⁻² s ⁻¹
N	Latitude, GPS coordinates	0
E	Longitude, GPS coordinates	0
Height	Altitude above sea level	m
SAT	Numbers of Satellites (minimum 3)	
DOP	Dilution of precision of GPS sensor	
Leaf Az.	Leaf azimuth; 0-360° clockwise from north	0
Leaf Sl.	Leaf slope from horizontal; 0 - 180° (0 = horizontal; 180 =horizontal upside down)	0
Sun Az.	Sun azimuth; 0-360° clockwise from the north.	0
Sun El.	angle of the sun above the horizontal plane ±90°	0
A. o. l.	Angle of Incidence, angle at which sun- light strikes the surface of the leaf. $(\pm 90^\circ; 0 = direct incidence)$	0
Inc. %	Percentage of possible sun inclination which strikes the surface of the leaf	%

10.2 H₂O mole fractions

The H_2O mole fraction in the air stream is determined by humidity sensors before and after leaf interaction. Relative humidity, temperature and pressure data are considered for the calculation of upstream H_2O mole fraction, and the downstream H_2O mole fraction, with vapor release of the leaf into the air stream.

The relative humidity is the ratio of the actual water vapor pressure of the air to the saturation water vapor pressure.

EQ: 2

 $RH = \frac{Actual Vapor Pressure}{Saturation Vapor Pressure}$

The porometer humidity sensors detect RH, pressure and temperature within the analytical cells. The H_2O mole fraction is calculated as follows:

EQ: 3

 $H_20 \text{ mole fraction } [mmol \ mol^{-1}] = \frac{SVP(T) * RH/100 * \ 10^3}{P}$

Whereby

- SVP (T) = saturation vapor pressure at temperature of the analytical cell calculated according to Goff-Gratch [kPa] (chapter 0),
- RH = relative humidity in the analytical cell [%],
- T = analytical cell temperature [°C],
- P = analytical cell pressure [kPa],

In the porometer, humidity sensor data result in the upstream (H_2O_{in}) , and downstream water mole fractions, whereby a difference due the sensor offset (Match) is taken into account for H_2O_{out} .

EQ: 4

$$H_2 O_{out} = \left(\frac{SVP(T) * \frac{RH}{100} * 10^6}{P}\right) - H_2 O_{Match}$$

The water released into the air stream by plant interaction is calculated by the difference of the upstream (H_2O_{in}), and downstream (H_2O_{out}) water mole fractions.

EQ: 5

$$dH_2 O = H_2 O_{out} - H_2 O_{in}$$

10.3 Transpiration Rate (E)

According to Caemmerer and Farquhar (1981), the transpiration rate is calculated as follows:

EQ: 6

$$E = \frac{u_e * (w_o - w_e)}{LA * (1000 - w_o)}$$

Whereby

E = transpiration rate [mmol m⁻² s⁻¹],

 $u_e = molar$ flow rate at the inlet of the cuvette [mmol s⁻¹], $w_o = H_2O$ mole fraction at the outlet of the cuvette [mmol mol⁻¹], $w_e = H_2O$ mole fraction at the inlet of the cuvette

[mmol mol⁻¹],

LA = leaf area [m²].

The terms in the equation (EQ: 6) relate to the values provided by the porometer as follows:

$$u_e = Flow_{in}$$

$$w_o - w_e = dH_2O$$

$$w_o = H_2Oin + dH_2O$$

$$LA = 0,000079 m^2$$

Using the values provided by the porometer the transpiration rate E can be calculated as follows:

EQ: 7

$$\mathbf{E} = \frac{Flow_{in} * dH_2O}{0,00007854 m^2 * (1 - (H_2Oin + dH_2O))}$$

10.4 Vapor Pressure Deficit (VPD)

According to Caemmerer and Farquhar (1981) the ALVPD (or VPD) is calculated as follows:

EQ: 8

$$VPD = \frac{(w_i - w_a)}{1000 - \frac{(w_i + w_a)}{2}}$$

Whereby

VPD = (Air-to-Leaf-) Vapor-Pressure-Deficit [Pa kPa⁻¹],

 w_i = Intercellular H₂O mole fraction within the leaf [mmol mol⁻¹],

wa = H_2O mole fraction in the leaf chamber = wo [mmol mol⁻¹].

The intercellular H_2O concentration (w_i) is determined from the temperature of the leaf assuming 100% humidity in the intercellular air spaces:

EQ: 9

$$w_i = \frac{\text{SVP(Tleaf)}}{\text{Pressure}}$$

Whereby

SVP (Tleaf) = saturation vapor pressure at Tleaf calculated according to Goff-Gratch [kPa] (chapter 0), Pressure = total pressure in the leaf chamber [kPa],

$$VPD = \frac{\left(\frac{SVP(Tleaf)}{Pressure} - wa\right)}{1 - \frac{\left(\frac{SVP(Tleaf)}{Pressure} + wa\right)}{2}}$$

VPD can also be expressed in kPa. To obtain this unit, multiply the VPD value by the ambient pressure and divide by 1000.

10.5 Water Vapor Conductance (g_t)

According to Caemmerer and Farquhar (1981) the total water vapor conductance g_t is calculated as follows:

 g_t can be calculated from results of equations EQ: 7 and EQ: 10.

EQ: 11

$$g_t = \frac{\mathrm{E}}{\mathrm{VPD}}$$

Whereby

 g_t = total water vapor conductance [mmol m⁻² s⁻¹], E = transpiration rate [mmol m⁻² s⁻¹],

VPD = (Air-to-Leaf-) Vapor-Pressure-Deficit [Pa/kPa].

10.6 Stomatal Conductance (g_s)

Calculation of the one-sided stomatal conductance requires knowledge of the influence of boundary layer conductance as:

EQ: 12

$$\frac{1}{g_t} = \frac{1}{g_s} + \frac{1}{g_b}$$

whereby

 g_t = total water vapor conductance [mmol m⁻² s⁻¹], g_s = stomatal water vapor conductance [mmol m⁻² s⁻¹], g_b = boundary layer water vapor conductance [mmol m⁻² s⁻¹]

Solving this equation for the stomatal conductance:

EQ: 13

$$g_s = \frac{1}{\frac{1}{g_t} - \frac{1}{g_b}}$$

The boundary layer conductance g_b within the porometer was determined with an on water floating filterpaper, and ambient H₂O concentration at a dew point of 10°C. The flow dependency of the boundary layer is expressed by the following equation:

EQ: 14
$$g_{b(flow)} = -0.0298 flow^2 + 18.065 flow + 2358.1$$

Whereby flow is the porometer flow rate in μ mol s⁻¹.

With the flow specific boundary layer g_b (EQ: 14) the porometer calculates the stomatal conductance g_s according to equation EQ: 13.
10.7 Geospatial Data

The MINI-PAM-II/POROMETER gives detailed information about photosynthetic performance and provides insights into a plant's water-use strategy and stress response. Integrating this data with geospatial information, enhances the ability to elucidate the complex interplay between plants and their environment.

The MINI-PAM-II/POROMETER is equipped with GPS receiver, UTC time, accelerometer, gyroscope, magneto scope providing precise geographic positioning (latitude, longitude, altitude), solar position, exact orientation of the sample in the field, and in relation to the position of the sun. The latter is used to calculate the angle of incidence of the sun on the leaf.

EQ: 15

Inc.
$$\% = 100 \cos (A. o. I.)$$

Whereby:

A.o.I.= angle at which sunlight strikes the surface of the leaf (0= direct inclination)

11 Saturation Vapor Pressure above Water

The following three tables show the saturation water vapor pressure above water.

The table values were calculated with the formula of Goff-Gratch (List, Robert J.; Smithsonian Meteorological Tables; Smithsonian Institution Press; Washington, D.C.; Fifth reprint issued 1984):

$$\log_{10} \left[SVP(T) \right] = -7.90298 \cdot \left(\frac{Ts}{T} - 1 \right) + 5.02808 \cdot \log_{10} \left(\frac{Ts}{T} \right)$$
$$-1.3816 \cdot 10^{-7} \cdot \left(10^{11.344*(1-T/Ts)} - 1 \right)$$
$$+ 8.1328 * 10^{-3} * (10^{-3.49149*(Ts/T-1)} - 1) + \log_{10} \left(SP_{ws} \right)$$

- SVP(T) = saturation vapor pressure over a plane surface of pure ordinary liquid water dependent on T [hPa],
- T = absolute (thermodynamic) temperature [K],
- Ts = steam-point temperature [373.16 K],
- SP_{ws} = saturation pressure of pure ordinary liquid water at steam-point temperature [1013.246 hPa] (= 1 standard atmosphere).

The following two tables indicate the saturation vapor pressure in a temperature range between 0 and 100.9°C. The temperature can be read in steps of tenth degrees (add left side with head-line).

Temp.	.0	.1	.2	.3	.4	.5	.6	.7	.8	.9
°C	hPa	hPa								
0	6.108	6.152	6.197	6.242	6.288	6.333	6.379	6.426	6.472	6.519
1	6.566	6.614	6.661	6.709	6.758	6.807	6.856	6.905	6.955	7.004
2	7.055	7.105	7.156	7.207	7.259	7.311	7.363	7.416	7.469	7.522
3	7.575	7.629	7.683	7.738	7.793	7.848	7.904	7.960	8.016	8.072
4	8.129	8.187	8.245	8.303	8.361	8.420	8.479	8.538	8.598	8.659
5	8.719	8.780	8.842	8.903	8.966	9.028	9.091	9.154	9.218	9.282
6	9.346	9.411	9.477	9.542	9.608	9.675	9.742	9.809	9.877	9.945
7	10.013	10.082	10.152	10.221	10.291	10.362	10.433	10.505	10.577	10.649
8	10.722	10.795	10.869	10.943	11.017	11.092	11.168	11.243	11.320	11.397
9	11.474	11.552	11.630	11.708	11.788	11.867	11.947	12.028	12.109	12.190
10	12.272	12.355	12.438	12.521	12.605	12.690	12.774	12.860	12.946	13.032
11	13.119	13.207	13.295	13.383	13.472	13.562	13.652	13.742	13.833	13.925
12	14.017	14.110	14.203	14.297	14.391	14.486	14.582	14.677	14.774	14.871
13	14.969	15.067	15.166	15.265	15.365	15.466	15.567	15.668	15.770	15.873
14	15.977	16.081	16.185	16.290	16.396	16.503	16.610	16.717	16.825	16.934
15	17.044	17.154	17.265	17.376	17.488	17.600	17.714	17.827	17.942	18.057
16	18.173	18.289	18.406	18.524	18.643	18.762	18.881	19.002	19.123	19.245
17	19.367	19.490	19.614	19.739	19.864	19.990	20.116	20.244	20.372	20.500
18	20.630	20.760	20.891	21.022	21.155	21.288	21.421	21.556	21.691	21.827
19	21.964	22.101	22.240	22.379	22.518	22.659	22.800	22.942	23.085	23.229
20	23.373	23.518	23.664	23.811	23.958	24.107	24.256	24.406	24.557	24.708
21	24.860	25.014	25.168	25.323	25.478	25.635	25.792	25.950	26.109	26.269
22	26.430	26.592	26.754	26.918	27.082	27.247	27.413	27.580	27.748	27.916
23	28.086	28.256	28.428	28.600	28.773	28.947	29.122	29.298	29.475	29.653
24	29.831	30.011	30.192	30.373	30.556	30.739	30.923	31.109	31.295	31.483
25	31.671	31.860	32.050	32.242	32.434	32.627	32.821	33.017	33.213	33.410
26	33.608	33.808	34.008	34.210	34.412	34.615	34.820	35.026	35.232	35.440
27	35.649	35.858	36.069	36.281	36.494	36.709	36.924	37.140	37.358	37.576
28	37.796	38.017	38.239	38.462	38.686	38.911	39.138	39.365	39.594	39.824
29	40.055	40.287	40.520	40.755	40.991	41.228	41.400	41.705	41.940	42.187
30	42.430	42.674	42.920	45.100	45.414	43.663	43.914	44.165	44.418	44.672
22	44.927	45.164	43.442	45.701	43.901	40.225	40.460	40.750	47.010	47.265
22	47.551	47.621	46.092	46.304	40.057	40.912	49.100	49.400	49.743 52.610	52 004
33	53 200	53 407	53 706	54.006	54 307	54 700	55.004	55 310	55 618	55 026
35	56 237	56 5497	56.862	57 176	57 /02	57.810	58 130	58.450	58 773	50.007
36	59 422	59 749	60.077	60.408	60 739	61 072	61 407	61 744	62 082	62 421
37	62 762	63 105	63 450	63 796	64 143	64 493	64 844	65 196	65 551	65 907
38	66 264	66 624	66 985	67 347	67 712	68 078	68 445	68 815	69 186	69 559
39	69 934	70 310	70 688	71.068	71 450	71 833	72 219	72.606	72,994	73 385
40	73 777	74 172	74 568	74 966	75 365	75 767	76 170	76 575	76 982	77 391
41	77 802	78 215	78 629	79.046	79 464	79 885	80 307	80 731	81 157	81 585
42	82.015	82.447	82.881	83.317	83.754	84.194	84.636	85.080	85.525	85.973
43	86.423	86.875	87.329	87.785	88.243	88,703	89.165	89.629	90.095	90.563
44	91.034	91.506	91.981	92.458	92.937	93.418	93.901	94.386	94.873	95.363
45	95.855	96.349	96.845	97.343	97.844	98.347	98.852	99.359	99.868	100.380
46	100.894	101.410	101.929	102.449	102.972	103.498	104.025	104.555	105.088	105.622
47	106.159	106.698	107.240	107.784	108.330	108.879	109.430	109.984	110.540	111.098
48	111.659	112.222	112.787	113.355	113.926	114.499	115.074	115.652	116.233	116.816
49	117.401	117.989	118.579	119.172	119.768	120.366	120.967	121.570	122.176	122.784
50	123.395	124.009	124.625	125.244	125.865	126.489	127.116	127.745	128.378	129.012

Table 1: Saturating Vapor Pressure above water

Temp.	.0	.1	.2	.3	.4	.5	.6	.7	.8	.9
°C	hPa									
50	123.395	124.009	124.625	125.244	125.865	126.489	127.116	127.745	128.378	129.012
51	129.650	130.290	130.933	131.578	132.227	132.878	133.531	134.188	134.847	135.509
52	136.174	136.842	137.512	138.185	138.861	139.540	140.222	140.907	141.594	142.284
53	142.978	143.674	144.373	145.074	145.779	146.487	147.198	147.911	148.628	149.347
54	150.070	150.795	151.524	152.255	152.990	153.727	154.468	155.211	155.958	156.708
55	157.461	158.217	158.976	159.738	160.503	161.271	162.043	162.818	163.595	164.376
56	165.161	165.948	166.738	167.532	168.329	169.130	169.933	170.740	171.550	172.363
57	173.180	173.999	174.823	175.649	176.479	177.312	178.149	178.989	179.832	180.679
58	181.529	182.382	183.239	184.099	184.963	185.830	186.701	187.575	188.453	189.334
59	190.218	191.107	191.998	192.893	193.792	194.695	195.601	196.510	197.423	198.340
60	199.260	200.184	201.112	202.043	202.978	203.917	204.859	205.805	206.755	207.708
61	208.665	209.626	210.591	211.560	212.532	213.508	214.488	215.472	216.459	217.451
62	218.446	219.445	220.448	221.455	222.466	223.480	224.499	225.522	226.548	227.579
63	228.613	229.652	230.694	231.741	232.791	233.846	234.905	235.967	237.034	238.105
64	239.180	240.259	241.343	242.430	243.522	244.617	245.717	246.821	247.930	249.042
65	250.159	251.280	252.405	253.535	254.669	255.807	256.949	258.096	259.247	260.403
66	261.562	262.727	263.895	265.068	266.246	267.428	268.614	269.805	271.000	272.200
67	273.404	274.612	275.826	277.043	278.266	279.493	280.724	281.960	283.201	284.446
68	285.696	286.951	288.210	289.474	290.742	292.016	293.294	294.577	295.864	297.156
69	298.453	299.755	301.062	302.373	303.690	305.011	306.337	307.667	309.003	310.344
70	311.689	313.040	314,395	315.756	317.121	318,491	319.867	321.247	322.633	324.023
71	325.418	326.819	328.225	329.635	331.051	332.472	333.899	335.330	336.766	338.208
72	339.655	341.107	342.565	344.027	345.495	346,968	348.447	349.931	351.420	352.914
73	354.414	355.919	357.430	358.946	360.467	361.994	363.527	365.064	366.608	368.156
74	369.711	371.270	372.836	374.407	375.983	377.565	379.153	380.746	382.345	383.950
75	385.560	387.176	388.798	390.425	392.058	393.697	395.342	396.992	398.649	400.311
76	401.979	403.652	405.332	407.017	408.709	410.406	412.109	413.818	415.533	417.255
77	418.982	420.715	422.454	424.199	425.950	427.708	429.471	431.241	433.016	434.798
78	436.586	438.380	440.180	441.987	443.800	445.619	447.444	449.276	451.114	452.958
79	454.808	456.665	458.528	460.398	462.274	464.156	466.045	467.941	469.842	471.751
80	473.665	475.587	477.515	479.449	481.390	483.337	485.292	487.252	489.220	491.194
81	493.175	495.162	497.156	499.157	501.165	503.180	505.201	507.229	509.264	511.305
82	513.354	515.410	517.472	519.541	521.617	523.701	525.791	527.888	529.992	532.103
83	534.221	536.346	538.479	540.618	542.765	544.918	547.079	549.247	551.423	553.605
84	555.795	557.991	560.196	562.407	564.626	566.852	569.085	571.326	573.574	575.830
85	578.093	580.363	582.641	584.926	587.219	589.520	591.827	594.143	596.466	598.796
86	601.135	603.480	605.834	608.195	610.564	612.941	615.325	617.717	620.117	622.524
87	624.940	627.363	629.794	632.233	634.680	637.134	639.597	642.068	644.546	647.033
88	649.527	652.030	654.541	657.059	659.586	662.121	664.664	667.215	669.775	672.342
89	674.918	677.502	680.094	682.695	685.303	687.920	690.546	693.180	695.822	698.472
90	701.131	703.799	706.475	709.159	711.852	714.553	717.263	719.981	722.708	725.444
91	728.188	730.941	733.703	736.473	739.252	742.040	744.836	747.642	750.456	753.278
92	756.110	758.951	761.800	764.658	767.526	770.402	773.287	776.181	779.084	781.997
93	784.918	787.848	790.788	793.736	796.694	799.661	802.637	805.622	808.616	811.620
94	814.633	817.656	820.687	823.728	826.778	829.838	832.907	835.986	839.074	842.171
95	845.278	848.395	851.521	854.657	857.802	860.957	864.121	867.295	870.479	873.672
96	876.876	880.089	883.311	886.544	889.786	893.038	896.300	899.572	902.854	906.146
97	909.448	912.759	916.081	919.413	922.755	926.107	929.469	932.841	936.223	939.615
98	943.018	946.431	949.854	953.287	956.731	960.184	963.649	967.123	970.608	974.104
99	977.609	981.126	984.652	988.189	991.737	995.296	998.864	1002.44	1006.03	1009.64
100	1013.25	1016.87	1020.50	1024.14	1027.80	1031.46	1035.14	1038.83	1042.52	1046.23

12 Specifications

12.1 MINI-PAM-II/Porometer

Design: very compact leaf chamber with a circular 1 cm diameter sample area, one side ventilated with air flow at adjustable flow velocity. The amount of water vapor released to the air flow is determined with moisture sensors. The leaf temperature is determined by an IR sensor located in the chamber bottom. Geospatial data are provided by the built in GPS receiver, accelerometer, magneto scope and gyroscope. A mini-quantum-sensor can be positioned on the sample level by a movable Perpex arm. Ambient CO₂ values are monitored by a CO₂ sensor facing the outside at the lower left side of the porometer. For chlorophyll *a* fluorescence measurements a fiberoptics port aligns the MINI-PAM/F Fiberoptics at an angle of 60° relative to the measuring plane.

Power supply: MINI-PAM-II leaf clip socket;

6 AA (Mignon) rechargeable batteries (Eneloop 1.2 V/2 Ah) within the MINI-PAM-II typically provide power for more than 8 hours continuous porometer operation at maximum flow. Fluorescence measurements shorten the operation time (6 AA rechargeable batteries typ. provide power for up to 1000 yield measurements without porometer operation). Easy battery swap is possible.

Sample area: 1 cm diameter

Flow rates: 40; 60; 80; 100; 120; 140; 160; 180 or 200 µmol s⁻¹

RH sensor accuracy: typ. 20-70 % RH ±1.0 % RH; <20 % RH and >70 % RH ±1.5 % RH; ΔT = ±0.1 °C

Pressure sensor accuracy: ±1 hPa

Leaf temp. sensor accuracy: ±0.3 °C, emissivity adjustable 0.1000-1.000 without the need of recalibration

Ambient CO₂ sensor accuracy: ±(30 ppm, +3% of reading)

Flowmeter accuracy: ± (1.5 % RD + 0.15 % FS)

GPS receiver accuracy: 2.0 m CEP (circular error probable)

Micro quantum sensor: LS-B sensor for selective PAR measurement, range 0 to 7000 μ mol m⁻² s⁻¹, cosine corrected for light incident at an angle between -30 ° to +30 from surface normal,

Parameter: g_s [mmol m⁻² s⁻¹]; g_t [mmol m⁻² s⁻¹]; g_b [mmol m⁻² s⁻¹]; H_2O_{in} [mmol mol⁻¹]; H_2O_{out} [mmol mol⁻¹]; dH_2O [mmol mol⁻¹]; Match [mmol mol⁻¹]; chamber pressure [kPa]; TLeaf [°C; Flow_{in} [µmol s⁻¹]; Flow_{out} [µmol s⁻¹]; VPD [Pa kPa⁻¹]; E [mmol m⁻² s⁻¹]; PAR [µmol m⁻² s⁻¹];

Operating conditions: -5 to +45 °C; 0-90 % RH (non-condensing); 300-1100 hPa

Cable length: 75 cm

Dimensions: 24 cm x 7.5 cm x 14 cm (max L x W x H)

Weight: 450 g (excluding cable)

Subject to change without prior notice

12.2 Transport Case PORO/T

Design: Black case with custom foam packing

Dimensions: 31 cm x 26.5 cm x 11 cm (L x W x H)

Weight: 1,2 kg

Subject to change without prior notice

Warranty

All products supplied by the Heinz Walz GmbH, Germany, are warranted by Heinz Walz GmbH, Germany to be free from defects in material and workmanship for two (2) years from the shipping date (date on invoice).

12.3 Conditions

This warranty applies if the defects are called to the attention of Heinz Walz GmbH, Germany, in writing within two (2) years of the shipping date of the product.

This warranty shall not apply to

- any defects or damage directly or indirectly caused by or resulting from the use of unauthorized replacement parts and/or service performed by unauthorized personnel.
- any product supplied by the Heinz Walz GmbH, Germany which has been subjected to misuse, abuse, abnormal use, negligence, alteration or accident.
- to damage caused from improper packaging during shipment or any natural acts of God.
- to batteries, cables, calibrations, fiberoptics, fuses, gas filters, lamps, thermocouples, and underwater cables.

Submersible parts of the DIVING-PAM or the underwater version of the MONITORING-PAM have been tested to be watertight down to the maximum operating depth indicated in the respective manual. Warranty shall not apply for diving depths exceeding the maximum operating depth. Further, warranty shall not apply for damage resulting from improper operation of devices, in particular, the failure to properly seal ports or sockets.

12.4 Instructions

To obtain warranty service, please follow the instructions below:

- The Warranty Registration form must be completed and returned to Heinz Walz GmbH, Germany.
- The product must be returned to Heinz Walz GmbH, Germany, within 30 days after Heinz Walz GmbH, Germany has received written notice of the defect.
 Postage, insurance, and/or shipping costs incurred in returning equipment for warranty service are at customer expense. Duty and taxes are covered by Walz.
 Accompany shipment by the Walz Service and Repair form available at:

http://www.walz.com/support/repair_service.html

- All products being returned for warranty service must be carefully packed and sent freight prepaid.
- Heinz Walz GmbH, Germany is not responsible or liable, for missing components or damage to the unit caused by handling during shipping. All claims or damage should be directed to the shipping carrier.

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