

International Committee for Controlled Environment Guidelines

# GUIDELINES FOR MEASURING AND REPORTING ENVIRONMENTAL PARAMETERS FOR EXPERIMENTS IN GREENHOUSES

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## Background

The importance of appropriate, accurate measurement and reporting of environmental parameters in plant sciences is a significant aspect of quality assurance for all researchers and their research. There is a clear need for ensuring research across the world can be compared, understood and where necessary replicated by fellow researchers. A common set of guidelines to educate, assist and encourage comparativeness is of great importance. On the other hand, the level of effort and attention to detail by an individual researcher should be commensurate with the particular research being conducted. For example, a researcher focusing on interactions of light and temperature should measure all relevant parameters and report a measurement summary that includes sufficient detail allowing for replication. Such detail may be less relevant when the impact of environmental parameters on plant growth and development is not the main research focus. However, it should be noted that the environmental experience of a plant during production can have significant impact when subsequent experiments investigate plants at a molecular, biochemical or genetic level or where species interactions are considered. Thus, researchers are encouraged to make a critical assessment of what parameters are of primary importance in their research and these parameters should be measured and reported.

This publication brings together a collection of parameters that should be recorded and reported when publishing scientific data from experiments conducted in greenhouses. It provides recommendations to end users on when, how and where these parameters should be measured along with the appropriate internationally standardized units that should be used. These guidelines have previously been published in the journal *Plant Methods* [1].

## Introduction

Research greenhouses are important tools for scientists and engineers studying the effects of environmental parameters on the growth and development of a large range of agronomic and ornamental plant species. In addition, the greenhouse environment allows for research to continue year-round so that external environmental influences have minimal effect on the experimental set points. The design and operation of research greenhouse facilities vary significantly from one facility to the next [2-5]. Therefore, accurate environmental control and data collection are essential to ensure that experiments can be adequately evaluated by research colleagues and replicated among different facilities [6].

These guidelines build on two previous ICCEG guidelines covering minimum sets of parameters that should be reported when working in plant growth rooms and chambers [7, Appendix 1] and in tissue culture facilities [8, Appendix 2]. The focus on greenhouses in this publication is complemented by earlier works relating to plant growth chambers [9,10].

It is the researcher's responsibility to become familiar with the operational principles of the sensor(s) used during experimentation. The principle of operation, the manufacturer's design of the sensor, and the specific sensor installation can all have an effect on the readings collected. These effects should be understood and considered when evaluating the measurements. Due to the spatial and temporal variability of many environmental parameters in greenhouse environments, it is important to carefully consider sensor placement, method of data processing, and the impact this variability may have on the experimental conclusions presented.

When reporting experimental results, it is recommended that the equipment and instrument information (model, manufacturer, town, country) is included, including information on precision and accuracy, when it is of specific importance to the experiment(s) conducted. In addition, the calibration procedure and the time of the most recent calibration should be reported. It is good practice to calibrate sensors before and after experiments so that any changes in calibration can be evaluated and incorporated into the data analysis. Periodic factory re-calibration of sensors is recommended (follow manufacturers' recommendations), and proper calibration can be verified by comparing sensor output with the output from calibrated sensors that are otherwise stored and only used for the purpose of calibration. Note that such calibration sensors may require periodic re-calibration also. In all cases, multi-point calibration of sensors is recommended.

These guidelines describe and discuss environmental parameters that affect plant growth and development. These parameters are divided into five categories: radiation, temperature, gases, water, and nutrients that are each discussed. In addition, greenhouse structure, growing system and environmental control system/strategies are included as a sixth comprehensive parameter. Greenhouse designers, managers and researchers conducting plant experiments in greenhouses are encouraged to review and implement the monitoring and reporting practices discussed in these guidelines. The guidelines conclude with a sample report describing how the environmental conditions during an experiment should be reported in the literature.

## Compliance and quality assurance

Whether growing conventional plants, genetically modified organisms (GMOs) or quarantine crops, producers (including those operating in research facilities) need to comply with an increasing range of regulations, codes of practice, legislation and/or standards. For example, compliance is often needed regarding hazardous material & waste management and/or environmental and health & safety regulations such as the Australian/New Zealand Standard [11]. Other regions of the world may have similar compliance requirements that need to be followed. In addition, sponsors of research increasingly expect a minimum standard of scientific practice, as described for example in the Research Councils' United Kingdom Policy and Guidelines on Governance of Good Research Conduct [12]. It specifies 'weak procedures, inadequate documentation of procedures or inadequate record-keeping' are inappropriate research behaviour. Furthermore in the UK, many grants by Research Councils and other government departments are made conditional on abiding by the Joint Code of Practice for Research [13] that includes the statement 'All equipment must be appropriate for the measurements to be made, calibrated at the appropriate interval, and be in good working condition'.

For greenhouse experiments, there are no standards that are equivalent to the International Organization for Standardization (ISO) ISO/IEC 17025 used for testing and calibration laboratories [14]. However, some of the practices that laboratories adopt to meet this standard's requirements could be beneficial for research in greenhouse facilities. Although they may appear daunting at first, they can be implemented with little effort and give rewards in terms of more effective operation. These include documenting methods (i.e. calibration and measurement), listing all staff competent in those methods, records of their training, and identification of sensors and equipment. For each item of measurement equipment there should be a log book recording when and how it was calibrated, who undertook the calibration and records of any observations on performance. Annotated records of all measurements should be stored in multiple locations.

It is recommended to utilise an independent set of sensors to measure environmental parameters in greenhouse environments, separate from those sensors used for monitoring and control. The reasons are:

- Sensors used for monitoring and control may not have the same specifications and/or operational requirements as the ones recommended for experimental measurements (e.g. response time, resolution, physical location).
- Even if sensors used for monitoring and control are serviced and calibrated, they may still malfunction during an experiment. Therefore, an independent set of sensors provides added assurance.
- Many sensors include, in addition to the actual probe, electronics and software that interpret and in some cases display the signals. Using different probes in the same hardware package does not offer truly independent assurance.
- Some regulatory bodies require that experiments are conducted using monitoring where the data recording the environmental conditions have assured integrity. This integrity includes not allowing any of the raw data collected to be altered by the user. Systems to monitor and control the greenhouse environment may not allow such a level of data integrity.

## Definitions

### I. Radiation

- Irradiance: The incident radiant energy flux per unit plane surface area ( $\text{J m}^{-2} \text{s}^{-1}$  or  $\text{W m}^{-2}$ ).
- Photon: The smallest packet ('particle') of electromagnetic radiation having no mass, and no electrical charge.
- Photon irradiance: The incident photon flux per unit plane surface area (also known as photon flux density;  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ).
- Photosynthetically active radiation (PAR): The part of the radiation spectrum that is by definition used by plants for photosynthesis (between 400 and 700 nm;  $\mu\text{mol m}^{-2} \text{s}^{-1}$ ).
- Radiation integral: The radiation flux summed over a period of time (typical units for plant experiments:  $\text{mol m}^{-2} \text{d}^{-1}$ ).
- Red:far red (R:FR) ratio: The red to far red ratio (ratio of radiation emitted at or close to 660 and 730 nm).
- Solar radiation (short wave radiation): The radiation emitted by the Sun (approximately between 280 and 3000 nm;  $\text{W m}^{-2}$ ).
- Spectrum: The distribution of energy (light) emitted by a radiation source in relation to wavelength.
- Waveband: A consecutive portion of the radiation spectrum.
  - a) 100 - 380 nm Ultra violet (UV)
    - I. 100 - 280 nm UVC
    - II. 280 - 315 nm UVB
    - III. 315 - 380 nm UVA
  - b) 380 - 770 nm Visible
  - c) 400 - 700 nm PAR
  - d) 770 - 3000 nm Near infrared (NIR)
  - e) 3000 nm<sup>-1</sup> mm Far infrared (Long wave)
- Wavelength: The distance between one peak (crest) of a wave of light and the next corresponding peak (nm).

## 2. Temperature

- Aspirated sensor: A sensor placed in an enclosure equipped with a downstream fan to provide continuous air movement around the sensor (with an air speed of approximately  $3 \text{ m s}^{-1}$ ). The sensor should be placed at a representative location near the crop canopy.
- Exposed sensor: A sensor fully exposed to the environment.
- Shielded sensor: A sensor shielded from solar radiation but exposed to passive air movement.
- Surface temperature: An object's surface temperature is one of the parameters that determines how much energy it radiates to its surroundings (Stefan-Boltzmann Law) ( $^{\circ}\text{C}$ ).

## 3. Gaseous concentration in air

### *Water vapour in air*

- Dew point temperature: The temperature to which the air must be cooled before the water vapour contained in the air begins to condense ( $T_{dp}$ ) ( $^{\circ}\text{C}$ ).
- Dry bulb temperature: The temperature measured by a thermometer freely exposed to air but shielded from radiation and moisture ( $T_{db}$ ) ( $^{\circ}\text{C}$ ).
- Humidity ratio: The ratio of the mass of water vapour to a unit mass of dry air (containing both water vapour and other gaseous constituents) ( $w$ ) ( $\text{kg/kg}$ )
- Partial vapour pressure: The pressure water vapour would exert if it alone occupied a volume of air (i.e. excluding the pressure resulting from other constituents of air) ( $e$ ) ( $\text{Pa}$ ).
- Relative humidity: The ratio of the partial vapour pressure ( $e$ ) to the saturated vapour pressure ( $e_{sat}$ ) at the dry bulb temperature ( $RH$ ) (%).
- Vapour pressure deficit: The difference between the pressure exerted by water vapour in a volume of air and the pressure that would be exerted by the water vapour that volume could hold if the air was saturated ( $VPD$ ) ( $\text{kPa}$ ).
- Wet bulb temperature: The lowest temperature that can be measured after adiabatic evaporation of pure water from a wick placed around an aspirated thermometer bulb ( $T_{wb}$ ) ( $^{\circ}\text{C}$ ).

### *Carbon dioxide in air*

- Atmospheric  $\text{CO}_2$  concentration: The concentration of carbon dioxide molecules in a mixture of air [ $\text{CO}_2$ ] ( $\mu\text{mol mol}^{-1}$ ).

### *Oxygen in aqueous solution*

- Dissolved oxygen concentration: The concentration of oxygen molecules in an aqueous solution [ $\text{O}_2$ ] ( $\text{mg L}^{-1}$ ).

## 4. Liquid water

- Volumetric water (or moisture) content: The volume of water contained in a unit volume of soil. Widely used and measured with a variety of soil moisture sensors ( $\theta_v$ ) (dimensionless fraction).
- Gravimetric water (or moisture) content: The mass of water in a unit mass of soil. Determined by weighing the soil before and after drying at  $105^{\circ}\text{C}$  ( $\theta_m$ ) (dimensionless fraction).
- Matric potential: The tension (suction) in the growing media that arises from the interaction between water and the matrix of solid particles ( $\psi_m$ ) ( $\text{kPa}$ ).

## 5. Nutrients

- Nutrient concentration: The concentration of an inorganic element in liquid solution or in a growing medium [*element*] ( $\text{mmol L}^{-1}$ ,  $\text{mol kg}^{-1}$  or  $\text{g L}^{-1}$ ).

## Recommended instruments and sensors (Table I)

### I. Radiation

The choice of radiation sensor depends on whether it is the energy content or photon content that is the quantity of interest.

- Net radiometer: Measures the difference between the incoming and outgoing radiation in  $\text{Wm}^{-2}$ . This sensor is typically used to evaluate the radiant energy environment in the greenhouse.
- Pyranometer: Measures total solar radiation in  $\text{Wm}^{-2}$ . This sensor is typically used to evaluate the incoming radiant energy from the sun.
- Quantum sensor: Measures photosynthetically active radiation in  $\mu\text{mol (photons) m}^{-2} \text{ s}^{-1}$ . (Figure I).
- Spectroradiometer: Measures the flux in discrete wavelength intervals across a specific waveband. This sensor is recommended when evaluating the radiation emitted from LED lighting systems because the narrow waveband emitted by LED sources is not always accurately measured by other radiation sensors.



**Figure I.** A quantum sensor on a leveling platform mounted at the top of the canopy for measuring photosynthetically active radiation (PAR). Radiation sensors should be chosen for the specific purpose required. Positioning should be carefully considered to avoid shading or noise from reflective surfaces. *Photograph courtesy of Lancaster Environment Centre, U.K.*

### 2. Temperature

- Infrared thermometer: A non-contact sensor appropriate for measuring surface (e.g. leaf) temperature. Infrared thermometers typically have a conical field of view that depends on sensor make and model, and distance from the surface being measured.
- Resistance temperature device (RTD): A sensor that operates on the principle that the electrical resistance in a thin wire or film is a function of temperature. The thin wire or film are often made of platinum and encapsulated in another material (e.g. ceramics or glass).
- Thermistor: A sensor that operates on the principle that the change in electrical resistance in a ceramic or polymeric material is a function of temperature. Thermistors typically achieve a higher precision across a more limited temperature range than RTDs.
- Thermocouple: A sensor made of two wires of dissimilar metals, fused at one end to form a junction, that produce a voltage, which is a function of the temperature difference between the junction and the other end of the two wires. An accurate reference temperature measurement is needed at the point where these two wires are connected to the meter (terminal).

### 3. Gases

#### Water vapour

- Capacitance hygrometer: A sensor that measures the resistive or capacitive changes in materials (Figure 2). A capacitive humidity sensor consists of a thin-film polymer sandwiched between two electrodes that record its capacitance. These sensors are inexpensive, small and require little maintenance. They have a wide measurement range (0-100%), a wide temperature range, high stability, fast response time, fully recover from condensation and are highly resistant to contamination. These sensors lose accuracy at low humidity (<5%) and require calculations (often accomplished using electronics) to convert capacitance to relative humidity.
- Dew point hygrometer (condensation hygrometer): A sensor that measures the scattering of light reflected from a mirror that is chilled to the point when condensation occurs. These dewpoint temperature sensors are expensive and require frequent cleaning because contamination on the mirror can cause errors. The measurements are precise ( $\pm 0.1^\circ\text{C}$ ) over a wide range ( $-100$  to  $+100^\circ\text{C}$ ). The measurement process does not distinguish between the formation of ice or dew on the mirror.
- Psychrometer: A device that consists of two matched thermometers over which air is drawn (Figure 2). One of the thermometers has its bulb covered with a wetted (with pure water) cloth or wick. These so-called wet and dry bulb sensors are relatively inexpensive, have good stability and tolerate condensation without damage. Accuracy ranges from  $\pm 2\%$  to  $\pm 5\%$  of true value and is affected by contamination (dirt or deposits) on the cloth/wick. The atmospheric humidity typically must be calculated using psychrometric equations, or more easily with a psychrometric table (Mollier diagram).



**Figure 2.** Water vapour (humidity) sensors may be relatively inexpensive capacitance hygrometers (left) or a basic psychrometer (right).

*Photograph courtesy of Lancaster Environment Centre, U.K.*

### *Carbon Dioxide*

- Non-dispersive infrared (NDIR) CO<sub>2</sub> sensor: Carbon dioxide absorbs infrared radiation between 4000 and 4500 nm. This absorption can be used to measure the volumetric CO<sub>2</sub> concentration with non-dispersive infrared (NDIR) CO<sub>2</sub> sensors, typically in an IRGA (infrared gas analyser). Since water vapour also absorbs infrared radiation, particularly in the 2500 - 3000 nm waveband, it is important to either remove the water vapour before measurement (by drying for example with a desiccant), adjust the sensor reading to account for the presence of water vapour, or alternately, install a pass-filter (interferometer) in front of the IR detector to ensure that only wavelengths that are absorbed by CO<sub>2</sub> are able to reach the detector. Zero calibration is performed by supplying the sensor with pure nitrogen gas. Additional calibration can then be performed using a calibration gas with a known CO<sub>2</sub> concentration at the high end of the measurement range. When air samples are drawn from (warm) greenhouse sections and pumped to a (colder) CO<sub>2</sub> sensor located outside the greenhouse, care should be taken to avoid condensation in the sampling tube, pumping system, and sensor components.

### *Dissolved Oxygen*

- Dissolved Oxygen meter: The dissolved oxygen (DO) concentration in a nutrient solution is most easily measured with a DO meter. The sensing component of a DO meter, a polarographic probe, consists of an anode and a cathode submerged in an electrolyte (e.g. potassium chloride) that is held in place by a semi-permeable membrane. Very small DO sensors consume very little oxygen, removing the need to stir the solution. A DO probe requires special care: the membrane must be kept wet and the probe filled with electrolyte. The electrodes can be cleaned by dipping them in ammonium hydroxide. The meter must be calibrated regularly (e.g. using a two-point calibration of 0 and 100% air saturated water).

## 4. Liquid water

For properties of rooting medium and/or irrigation water:

- Dissolved oxygen meter (see '3. Dissolved Oxygen' above)
- Electrical conductivity (EC) meter: A sensor that measures the electrical conductivity of a nutrient solution based on the total amount of dissolved salts (nutrients) and the resulting ion concentration. In solutions with ions from multiple elements, this sensor is not able to measure the concentration of individual elements. The EC measurement provides an indication of overall nutrient concentration in a solution. If a measurement of individual elements is required, ion-selective sensors can be used (see '5. Nutrients' below). EC meters are inexpensive and can be calibrated with commercially available calibration solutions. EC measurements are often combined with pH measurements for a quick and easy assessment of root zone or irrigation water.
- Soil/media/liquid temperature sensor (see '2. Temperature' above).
- Stem psychrometer: A sensor that measures the stem water potential in situ of woody plant species. A small measurement chamber is clamped onto a section of exposed sapwood. Two thermocouples measure the wet bulb depression between the temperature of the sapwood and the temperature inside the measurement chamber. Using temperature corrections, the stem water potential can be accurately determined. This measurement integrates the impact of all environmental conditions on plant status. Time series measurements can reveal the degree and changes of plant water stress.
- Tensiometer: A sensor that measures soil water tension. Soil water tension is equal to the tension (force) the root system must overcome to extract water from the soil. The sensor measures the tension (suction) on an enclosed water reservoir that is in continuous contact with the soil matrix through a porous ceramic cup. The less water in the surrounding soil, the higher the suction on the water reservoir.

- Soil moisture sensor: A sensor that measures the volumetric water content of the root zone. These sensors determine the electrical resistance or the dielectric constant as a proxy for moisture content. Sensor types include capacitance (frequency domain) or time domain reflectometry sensors. As tensiometers, these sensors can be used to manage crop irrigation cycles.

## 5. Nutrients

- Weighing scale: In addition to the sensors described above (see '4. Liquid water'), a scale with sufficient accuracy (e.g. 0.01 g or  $\pm 1$  display resolution) should be used to weigh the proper amount of fertiliser (containing one or more nutrients). The frequency of calibration of the scale should be recorded.
- While some ion-selective sensors are beginning to become available, they are not yet widely used because of concerns over price, stability and longevity. This may change in the future. However, if these sensors are used, make and model should be reported as well as accuracy and frequency of calibration.

Table I: Instruments & sensors for measurement & their calibration

What to measure	Units	Measured by	Precision of instrument <sup>2</sup>	Accuracy of reading <sup>2</sup>	Calibrated by and when
<b>Radiation (PAR<sup>1</sup>)</b>	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Quantum sensor	$\pm 1\%$	$\pm 10\%$	Comparison with a reference sensor or against a standard quartz-halogen lamp traceable to an (inter)national standard (e.g. US NIST). Once per annum.
<b>Radiation (Net)</b>	$\text{W m}^{-2}$	Net radiometer	$\pm 2\%$	$\pm 5\%$	Comparison with a reference meter or in a temperature controlled calibration chamber capable of evaluating both short and long wavebands. Once per annum.
<b>Radiation (Spectral)</b>	$\mu\text{mol m}^{-2} \text{s}^{-1} \text{nm}^{-1}$	Spectroradiometer	$\pm 1\%$	$\pm 5\%$	Comparison with a reference meter or against a standard source of radiation (e.g. ASTM G138-06). Once per annum.
<b>Irradiance (Solar)</b>	$\text{W m}^{-2}$	Pyranometer	$\pm 1\%$	$\pm 5\%$	Comparison with a reference meter or against a standard source (traceable to the World Radiometric Reference) in an integrating sphere. Once per annum.
<b>Radiation (Integral)</b>	$\text{MJ m}^{-2}$ or $\text{mol m}^{-2} \text{d}^{-1}$	Calculated from accumulated radiation data	-	-	-
<b>Air temperature</b>	$^{\circ}\text{C}$	RTD, thermocouple or thermistor (shaded and aspirated in air with speed $\geq 3 \text{ m s}^{-1}$ )	$\pm 0.1^{\circ}\text{C}$	$\pm 0.2^{\circ}\text{C}$	Comparison with a reference thermometer (e.g. traceable against US NIST) by placing in melting crushed ice and boiling distilled water. Once per annum.
<b>Substrate temperature</b>	$^{\circ}\text{C}$	RTD, thermocouple or thermistor (ensure good contact with substrate)	$\pm 0.1^{\circ}\text{C}$	$\pm 0.2^{\circ}\text{C}$	As above.

<sup>1</sup> Referred to as photosynthetically active radiation (PAR: 400 - 700 nm) for general usage & described as photosynthetic photon flux density (PPFD) by many journals, professional societies & manufacturers of quantum sensors.

<sup>2</sup> Precision is how close the measured values are to each other. Accuracy is how close a measured value is to the actual (true) value.

Table 1: Instruments & sensors for measurement & their calibration

What to measure	Units	Measured by	Precision of instrument <sup>2</sup>	Accuracy of reading <sup>2</sup>	Calibrated by and when
<b>Surface temperature</b>	°C	Infrared temperature sensor, fine wire thermocouple	± 0.1°C	± 0.2°C	Infrared sensor: Comparison with a reference surface thermometer mounted on the same surface within the field of view of the infrared sensor. Thermocouple: as above.
<b>Atmospheric moisture:</b> relative humidity or vapour pressure deficit (VPD)	% or kPa	Capacitance, dewpoint sensor, psychrometer, or IRGA (infrared gas analyser)	Relative humidity: ± 2% Dewpoint temp.: ± 0.1°C VPD: ± 0.3kPa	± 5% ± 0.5°C ± 0.5 kPa	Humidity generator; unsaturated salt solution calibration standards (35% and 80% RH). Once per annum.
<b>Air speed</b>	m s <sup>-1</sup>	Anemometer (range 0.1 - 15.0 ms <sup>-1</sup> )	± 2%	± 5%	Wind tunnel. Once per annum.
<b>pH</b>		pH probe (range 3-10)	± 0.1pH	± 0.1pH	Standard solutions. Before every measurement, or weekly in continuous measurement applications.
<b>Electrical conductivity (EC)</b>	S m <sup>-1</sup>	Electrical conductivity meter	± 3%	± 5%	As above.
<b>Dissolved oxygen</b>	mg L <sup>-1</sup>	Dissolved oxygen meter (maintain adequate solution flow rate & ensure temperature compensation)	± 3%	± 5%	As above
<b>Atmospheric CO<sub>2</sub> concentration</b>	µmol mol <sup>-1</sup>	Silicon based NDIR (non dispersive infrared) sensor as part of an IRGA (infrared gas analyser)	± 1%	± 3%	Certified calibration gases for low & high end of the measurement range, and/or precision gas mixing instrument. Once per week.

<sup>1</sup> Referred to as photosynthetically active radiation (PAR: 400 - 700 nm) for general usage & described as photosynthetic photon flux density (PPFD) by many journals, professional societies & manufacturers of quantum sensors.

<sup>2</sup> Precision is how close the measured values are to each other. Accuracy is how close a measured value is to the actual (true) value.

## Parameters to be monitored and recorded (Table 2)

Due to spatial and temporal fluctuations in the greenhouse environment, all measurements should be taken in as many locations as practically feasible and as frequently as possible, preferably continuously. When using a data logging system, the measurement frequency can be higher than the recording frequency. In that case, it is important to report what type of processing was used (e.g. scanning and averaging intervals) to record the data. The following points should be reported: (1) sensor location(s) or sampling point(s), whether aspirated or shielded (Figure 3), (2) average and standard deviation of the measurements, and (3) method and frequency of sensor calibration (Table 2).

Table 2 lists two sets of parameters. The minimum set comprises the recommended basic set of parameters that should be measured and recorded during the course of most experiments and reported in publications. The additional set includes parameters that could be measured and reported provided appropriate sensors are available and the parameters are pertinent to the type of experiment conducted. It also includes parameters that will comprise metadata (describing content and context of stored data files) held by facility managers and available to users if required.



**Figure 3.** An aspirated sensor box suspended centrally in free air for recording environmental parameters. The sensors are located inside the box while air is constantly drawn through the box. Sensor (box) type and location along with frequency of sampling and details of data integration should be reported.

*Photograph courtesy of Lancaster Environment Centre, U.K.*

## I. Radiation

### *Measurements*

Since the human eye is an inaccurate sensing device for quantifying light, sensors are needed to measure the radiation environment inside greenhouses that is constantly fluctuating due to daily and seasonal changes in solar altitude, degree of cloud cover, and shadows resulting from structural components and overhead equipment [16,17]. In addition, the radiation flux and spectrum changes when measured at different locations within a plant canopy. Unless light transmission in a plant canopy is the focus of research, PAR and its integral should be measured at the top of the plant canopy while assuring that no parts of the plant canopy shade the sensor. The location of the top of the plant canopy should be referenced relative to a fixed object in the greenhouse (e.g. the floor or the bottom of the supplemental light fixtures).

When a combination of sunlight and supplementary light is used to grow a crop, it is important to quantify how much of the total light was provided by each source. If this is not done, it is virtually impossible to repeat the experiment under similar conditions. Solar irradiance can be modified as it passes through greenhouse cladding material and/or interacts with the plant canopy. As a result, changes in the amount of UV and/or the R:FR ratio can occur and these should be monitored and reported when they have an impact on plant growth and/or development.

When conducting supplementary or photoperiod lighting experiments, researchers need to be aware of the impact these treatments can have from unintended spillage of light into neighbouring greenhouse sections. Curtains may need to be used to shield experimental sections.

Additional outside radiation sensors are often used to compare the radiation environments inside and outside the greenhouse. This comparison provides information about the transmission of radiation through the greenhouse structure. Outside radiation can also be used as input for the greenhouse control strategy [16,17].

### *Light sources*

To supplement sunlight, typical greenhouse radiation sources include: high pressure sodium (HPS), metal halide (MH), incandescent (INC), fluorescent (FL) lamps, and light emitting diodes (LED). Each radiation source has its own unique emission spectrum that can result in unique plant responses [18]. Therefore, it is important to report the type of supplementary radiation source that was used, what its spectral output was (particularly when unusual), and the mix (%) by wattage of any mixed source. It is also important to report the duration of the supplemental lighting period.

### *Effect on plant growth*

The rate of photosynthesis is by definition dependent on the amount of photosynthetically active radiation (PAR) absorbed by the plant canopy. While measuring PAR, researchers should be mindful of the light compensation point (where the net rate of photosynthesis becomes positive) and the saturation level associated with the environmental conditions and the plant species under investigation [16,18]. In most cases, it is useful to report the relative contributions of sunlight and supplementary light to daily light integral achieved.

### *Effect on plant development*

For many plant species, especially ornamentals, changes in daylength can trigger physiological responses (e.g. flower initiation). Therefore, it is important to report the length of the photoperiod imposed as well as the method used to implement the photoperiod (e.g. with black-out curtain or with supplementary lighting). When extending the natural daylength or providing night interruption, the light source, its radiation flux and spectrum, as well as the start time and duration of the lighting period should be reported, especially if it is different from the source of supplementary light for growth.

### *Effect on plant morphogenesis*

For some plant species, spectral characteristics of radiation can affect morphology (e.g. internode elongation, flower rotation towards the sun). When these morphological changes have an effect on growth and development, the responsible spectral characteristics should be quantified and reported.

## 2. Temperature

### *Measurements*

The greenhouse air temperature is determined by outside temperature, incoming and outgoing radiation, radiative, conductive and convective heat transfer associated with the structure, and the operation of the greenhouse heating and cooling system. Significant spatial and temporal fluctuations can result from daily or seasonal changes in outside conditions. As for radiation measurements, structural elements can generate shadows that can (temporarily) affect temperature measurements. In addition, these fluctuations can be the result of the management strategy used to maintain indoor conditions, including the heating, ventilation and cooling system as well as internal air circulation systems such as horizontal airflow fans or perforated polyethylene tubing distribution systems (poly-tube). During ventilation and cooling (especially when using an evaporative cooling pad or fog cooling), it is common to experience significant temperature gradients from the air inlet to the outlet. Minimizing these gradients requires understanding of the airflow patterns resulting from the ventilation system and/or additional internal air recirculation. As a result, continuous measurements, preferable at multiple locations, are needed to assess the average temperature conditions experienced by the plants. If only one measurement location can be monitored, care should be taken in selecting the most appropriate location for sensor placement (typically near the canopy in the centre of the growing area).

Maintaining optimum root zone temperatures can be critical for timely and/or maximum production of many plant species. For those plant species, root zone temperatures should be measured and recorded in at least one, but preferably more, representative location(s). Vertical temperature gradients can be expected and are typically more significant when plants are grown on the floor, whether that floor is heated or not. For hydroponic systems, the temperature of the (recirculating) nutrient solution should be reported.

### *Effect on plant growth*

The rates of chemical reactions typically increase with temperature. Within plants, most chemical reactions, including photosynthesis, are enzyme driven. Hence, the rate of reaction often decreases above an optimum because high temperatures damage the enzymes. Many plants produce compounds to protect against high and low temperatures. On the other hand, in some plants species extreme temperatures can result in abnormal morphological characteristics. In some cases, it is useful to record not just the average temperature and standard deviation, but also the maximum and minimum values the plants were exposed to. For the production of field crops (e.g. corn - *Zea mays* L.) in greenhouses, the calculation of the crop heat units (growing degree days and associated base temperature) should be reported.

### *Effect on plant development*

For many plant species, especially ornamentals, changes in temperature can trigger physiological responses (e.g. flower initiation). For those plant species, it is important to report the maximum temperature recorded during the photoperiod and during the night.

## 3. Gases (including water vapour)

### *3.1 Water vapour (H<sub>2</sub>O)*

Description: Water vapour is the gaseous phase of water and can be produced from evaporation or boiling water, or from the sublimation of ice. Water vapour will condense onto another surface only when that surface is cooler than the dewpoint temperature, or when the water vapour equilibrium in the air has been exceeded. When water vapour condenses onto a surface, a net warming occurs on that surface. When water vapour evaporates from a surface, a net cooling occurs on that surface and/or the surrounding air. Condensation of water vapour on plant surfaces should be minimised as not to increase disease and pest issues. In plants, water loss from the stomata by evaporation and diffusion is termed transpiration. Excessive transpiration should be avoided as it can cause wilting.

Measurements: Water vapour concentrations are typically measured in the air in/around the plant canopy. The amount of water vapour present in the greenhouse air can be expressed in different ways: while RH is a function of temperature, and is therefore a less desirable parameter to report, it is the parameter most often used to express the moisture content of greenhouse air. Many crops perform well when the RH ranges between 50 and 80%. Optimum vapor pressure deficit (VPD) values for most plant species range between 0.8 and 1.0 kPa. Small VPD values indicate humid conditions, making plant transpiration more difficult. Large VPD values indicate dry conditions, making plants susceptible to wilting. While many researchers report the relative humidity observed and/or maintained during their greenhouse experiments, it is more appropriate to use VPD because of its independence of temperature and because it is the driving force for transpiration.

### 3.2. Carbon dioxide ( $CO_2$ )

Description:  $CO_2$  is a trace gas comprising approximately 0.040% (by volume) of the Earth's atmosphere.  $CO_2$  is toxic to humans at higher concentrations: 1% may cause drowsiness, above 5% is dangerous. During photosynthesis, plants, algae, and cyanobacteria absorb carbon dioxide, sunlight, and water to produce carbohydrates and oxygen as a waste product. During (dark) respiration, plants reverse this process and absorb oxygen and emit carbon dioxide. In greenhouses, the  $CO_2$  concentration can become the limiting factor for photosynthesis under high light conditions, necessitating adequate ventilation or active enrichment.

Measurements: Carbon dioxide concentrations are typically measured in the air in or around the plant canopy. When detailed information about photosynthesis and/or respiration is needed, continuous measurements are necessary. Because accurate  $CO_2$  measurements require expensive equipment, a single sensor can be supplied with air samples from different locations and/or different greenhouse sections. These air samples can be supplied sequentially to the sensor with the help of a gas handling unit (sequencer). Readings can be stored using data acquisition systems (dataloggers). Sensor calibration can be accomplished using carefully prepared calibration gases. It is recommended that calibration is performed at no less than two concentrations: one at the low end and one at the high end of the measurement range. Depending on sensor type, temperature fluctuations (e.g. due to fluctuating exposure to solar radiation) can have an effect on measurements. In that case, sensors should be shielded from such fluctuations.

### 3.3 Oxygen ( $O_2$ )

Description: In plant nutrient solutions, the dissolved oxygen concentration can be an important indicator of the ability of the roots to respire. Similarly, in rooting media, the porosity (volumetric pore space fraction) is used as an indicator of the availability of  $O_2$  in the root zone.

Measurements: Oxygen concentrations of the greenhouse air are not often of concern and are therefore almost never measured. When plants are grown in rooting media, the availability of oxygen to the roots is often assessed by determining the volumetric pore space fraction of the media. In nutrient solutions, the available oxygen to the roots is assessed by measuring the dissolved oxygen concentration. The dissolved oxygen concentration measurement is dependent on the nutrient solution temperature: the warmer the solution, the less oxygen it can contain. Thus, when the nutrient solution temperature is not controlled, frequent measurements are needed. A dissolved oxygen concentration in equilibrium with the oxygen concentration in the air above the solution is called the saturation concentration.

### 3.4. Effect of gaseous pollutants on plant growth and development

A variety of gaseous pollutants, some in very small quantities, have been reported to cause negative effects on plant growth and development, particularly in controlled environments when ventilation rates are limited. Many plastics, paints, sealants, glues, etc. release chemical compounds over time that can result in cumulative effects. Chemicals used in and around greenhouses can also release harmful compounds. High temperatures resulting from solar radiation or from inefficiencies in energy conversion (e.g. in electrical

components) can increase the release of gaseous pollutants (e.g. NO<sub>x</sub>, SO<sub>x</sub>). Some plant species are highly sensitive to ethylene gas, a known plant hormone that is released, for example, from improperly operating combustion heating systems. People exhaling CO<sub>2</sub> can significantly increase their surrounding CO<sub>2</sub> concentration, particularly in small spaces. The most effective strategy to reduce the effect of gaseous pollutants is to minimise the use of materials that release (volatile) chemicals and to maintain adequate ventilation rates. Accurate measurements of (small) quantities of gaseous pollutants can require expensive equipment (e.g. gas chromatograph).

## 4. Liquid water

### *Specifications*

Water content of the rooting medium can be expressed in terms of volume, mass, filled pore space, matric (matrix) potential, or water potential. The transport and retention of water in the growing media or soil is important to quantify.

The composition of irrigation water, including pH, alkalinity, electrical conductivity, total dissolved solids, or salinity can have a profound effect on plant growth. The source of water can affect the composition/content of the water and consequently plant growth. In addition, water temperature can have an effect on plant growth as well as on the dissolved gases in the water, e.g. the dissolved oxygen content.

### *Measurements*

In soilless culture systems, e.g. hydroponics, measuring properties of water is typically straightforward. In soil-based culture systems, the so-called flow-through method can be used to evaluate the characteristics of the aqueous fraction of the root environment. In each case, the assumption is made that the water characteristics are uniform throughout the system. This assumption may or may not be reasonable.

Electrical conductivity (S m<sup>-1</sup>) and pH should be recorded as well as the source of water, e.g. ground water, river/stream water, treated water (e.g. potable water from a local water company), deionised/distilled water, rainwater collected from the roof, including whether it includes recycled water from the greenhouse (e.g. condensate).

When soil water content or soil water potential is controlled, details about the control interval and the accuracy (proportion (%) of time the control parameter stayed within the control interval) of the control should be reported.

Irrigation water temperature should be recorded (especially when it varies during the day). The frequency of measurement should reflect the likelihood of variation. Delivery rate of irrigation water per square meter/pot, frequency (timing), and watering strategy should be reported. The fate of irrigation water should be reported, i.e. whether excess is allowed to run off or it is recirculated.

### *Effect on plant growth*

Water is the largest component of plants, typically composing 70 to 95% of fresh plant mass and providing shape and rigidity by positive turgor pressure. Turgor pressure also drives cell elongation. Water is the transport medium for moving nutrients from the soil into the plant. Evaporation of water from the stomata provides cooling and prevents overheating of the leaves. The amount of water available can affect flower initiation, vegetative growth and fruit/leaf size.

### *Effect on plant development*

A decrease in plant water content of 10% can greatly affect plant growth. Lack of water can reduce plant rigidity, reduce leaf expansion and result in irreversible damage to plant tissue.

## 5. Nutrients

### *Description*

Plants require inorganic nutrients (fertilisers) for proper growth and development [19,20]. The typical plant contents, as proportion (%) of dry mass, for C, H, and O are approximately 41, 6, and 42%, respectively. All other essential nutrients (the remaining 11%) are typically provided to the rooting medium so they can be taken up by the roots. These essential nutrients are divided into two groups, the macro (or major) and the micro (or minor) nutrients based on the quantities needed by the plant. The macro nutrients include N, P, K, Ca, Mg, and S. The micro nutrients include Fe, Mn, Zn, Cu, B, Mo, Cl, Co, and Ni, and some of these are required only in extremely small amounts. When nutrients are provided (dosed) as part of the irrigation system, they need to be dissolved in water as ions (with a few exceptions, e.g. urea and boric acid). When nutrients are provided in solid form (e.g. as a slow release fertilizer incorporated into or placed on top of the rooting media), they need to be dissolved before they can be taken up by the roots. In some cases, foliar sprays are used to supply nutrients.

For liquid nutrient applications, in order to provide a mixture of nutrients with the correct concentrations, chemical compounds with known compositions can be dissolved in water. These compounds are commercially available in specific formulations and often include a complete breakdown of the nutrient composition. Chemicals can be mixed and dissolved into a concentrated stock solution, but some mixtures can result in precipitation (making those nutrients unavailable to the plants). This can be avoided by using two stock tanks each containing a different mixture of compounds that does not form a precipitate.

The pH of the aqueous fraction of the root environment has an effect on the availability of nutrients for uptake by the plant. Therefore, the pH is typically monitored regularly and controlled to within a relatively narrow range (e.g.  $\pm 0.2$ ).

### *Measurements*

Composition, dosing, and frequency of nutrient application should be recorded at the beginning of an experiment and at any time when rates are adjusted. Recommended measurements of the aqueous fraction of the root environment are described previously in the section entitled 'Water'. When pertinent to the research, results from nutrient analyses should be reported, as well as the methods used to analyse the samples. Results from plant tissue analysis can be used to verify/adjust nutrient applications.

### *Effect on plant growth and development*

When plants are not provided with the appropriate levels of nutrients, specific deficiency or toxicity symptoms may appear [20]. These symptoms are plant specific and often correlated with the level of deficiency or toxicity. These symptoms include, but are not limited to, discoloration, spotting, dead tissue, abnormal development, and stunted growth. The roles of various nutrients in plant growth and development have been studied extensively, but are not yet always fully understood.

## 6. Greenhouse structure, growing system and environmental control system/strategy

### 6.1 Radiation

**Structure:** The greenhouse structure and its orientation (typically expressed as the orientation of the ridge relative to due North) have a significant effect on the transmission of solar radiation [16, 21]. In particular, they determine the size, shape and movement of shadows across the growing area. Of the structural elements, the glazing material will have the most significant effect on the radiation flux and spectral distribution reaching the plant canopy. Therefore, greenhouse orientation and structural features, including the type of glazing material and whether a shading compound was added should be reported. Note that dust or dirt accumulation on the glazing can reduce light transmission, but this issue can be minimized by periodic cleaning. Since physical properties of some cladding materials can change over time, it is recommended to report on the age of the material and its general status. Where long-term data exist, it may be useful to report the average transmission of radiation as a proportion (%) of outside radiation.

**Growing system:** The spatial uniformity of radiation emanating from supplementary lighting systems is less than that of solar radiation. Researchers need to quantify and report this uniformity so that its effect on plant growth and development can be assessed [16,17]. The design and management of the growing system can also have an effect on the radiation environment: the design and location of the growing surface (e.g. benches, floor, and their surface characteristics), the presence of overhead equipment and/or plant material, and the number of re-spacings or relocations during the experiment.

**Environmental control system:** The operation of a supplementary lighting and/or shading system will have a significant effect on the radiation environment. Therefore, operational procedures and control strategies should be reported. Design specifications, but preferably measured quantities (e.g. radiation flux, proportion of shading) should also be reported.

### 6.2 Temperature

**Structure:** The greenhouse structure, in particular the type of cladding material (glass, rigid plastic or film), will have a significant effect on its heat transfer [6,21]. Therefore, the type of greenhouse cladding material should be reported. Since the physical properties of some cladding materials change over time, it is recommended to report on the age of the material and its general status.

**Heating, ventilation, and cooling systems:** Depending on the type of research and the usefulness of the data for other fields of study, it can be helpful to report on the type of heating system, its maximum capacity, the control strategy implemented. In addition, information about additional measures implemented to improve energy distribution, such as perforated poly-tubes or horizontal airflow fans, should be reported. Typical greenhouse heating options include forced air (e.g. unit heaters, air furnaces), hot water (e.g. radiant floor and/or perimeter and overhead radiant pipe) and infrared systems (Figure 4). Typical ventilation options include natural (using sidewall and/or roof openings) and mechanical (using electrical fans and sidewall openings) systems. Typical (evaporative) cooling systems include pad-and-fan and high-pressure fog systems.

**Growing system:** In order to provide a uniform growing environment for plants, it is important to assess the temperature distribution and variability. The number and capacity (at a specific static pressure) of all fans should be reported. The direction of airflow should also be reported in order

**Figure 4.** Expanded metal benches with bottom heating from finned piping installed underneath.

*Photograph courtesy of Sainsbury Laboratory, University of Cambridge, U.K.*



to assess whether temperature gradients may be present. Despite increased airflow to improve the uniformity of temperature distribution throughout the greenhouse, the cropping system (particularly tall crops) can have a significant effect. A dense canopy can significantly reduce air movement and vigorous crop transpiration can (locally) reduce air temperature (converting sensible heat to latent heat). These factors should be taken into account and discussed when measuring and reporting air temperatures in greenhouse environments.

**Environmental control system:** In order to save energy and reduce the plant respiration rate, many greenhouse operations implement different temperature set points during the day (photoperiod) and night [16, 17]. These temperature differences can also be used to control specific growth stages, e.g. stem elongation. Specific temperature regimes can also be used to vernalise some plant species (resulting in induction of germination or flowering). Different control strategies can be implemented to maintain greenhouse temperatures. Each control system and strategy uses a different method (control algorithm) to reach the desired outcome, resulting in different spatial and temporal temperature distributions. Some control strategies deliberately allow the temperature to fluctuate depending on outside conditions provided some daily average temperature can be achieved. Because of the effect of the various control systems and strategies, pertinent information about the systems and the control algorithms should be reported (Figure 5). In all cases, it is essential to report measured values and not set points.

### *6.3 Gases (including water vapour)*

**Structure:** The tightness (leakiness) of the greenhouse structure will have an effect on the gas and water vapour concentrations inside. Outside wind conditions combined with a leaky greenhouse structure can have additional effect on gas and water vapour concentrations. The leakiness of a greenhouse structure can be evaluated through visual inspection for leaks or cracks, techniques to reveal airflow, or the concentration changes of deliberately released tracer gases. Cold glazing surfaces can result in condensation (and therefore, dehumidification), but the resulting water droplets can damage young plants when falling down instead of running into a collection system.



**Figure 4.** Greenhouse environmental control systems are diverse and frequently control algorithms are specific to the particular size, structure, orientation and location of the facility. Pertinent information on the system should be reported.

*Photograph courtesy of Sainsbury Laboratory, University of Cambridge, U.K.*

**Growing system:** The growing system (e.g. bench designs that do or do not allow for vertical air movement) and the volume and density of the plant canopy can have a significant effect on the distribution and uniformity of the concentration of gases throughout the greenhouse. Air distribution systems (e.g. horizontal airflow fans or poly-tube distribution systems) can help distribute the air and thus improve uniformity. However, the uniformity of gas and water vapour distribution should be assessed spatially and temporally to ensure that the values reported were representative of the conditions experienced by all the plants during the experiments.

**Enrichment/removal systems - water vapour:** Water vapour can be added or removed from greenhouse environments using humidifiers or dehumidifiers, respectively. Humidifiers produce water droplets of various sizes depending on their design and operation. Ideally, droplet size is minimised so as to speed up evaporation without wetting plant tissues. Air circulation is often used to enhance the distribution of the humidified air. Note that evaporative cooling, using either a pad-and-fan system or a high-pressure fog system, humidifies as well as cools the greenhouse air. Dehumidification using a refrigeration cycle to remove water vapour is an energy intensive and thus expensive approach. Instead, greenhouses can be dehumidified using the heat-and-vent strategy: heated air can hold more moisture that is then removed by ventilation. While energy is required for the heat-and-vent strategy, the benefits can outweigh the costs.

Enrichment/removal systems - carbon dioxide: Carbon dioxide enrichment can be accomplished by distributing (pure) CO<sub>2</sub> gas into the growing area. CO<sub>2</sub> gas can be derived from different sources: from vapourised liquid CO<sub>2</sub> stored in a refrigerated and pressurised container near the greenhouse site, or from burning natural gas and releasing the (scrubbed) exhaust gases inside the greenhouse. Uniform distribution is often accomplished by distributing the gas through small inflatable polyethylene tubes that have small openings along the length of the tubes. Typical target enrichment concentrations are 2-4 times the normal ambient concentration (approximately 750-1500 μmol mol<sup>-1</sup>). Often, CO<sub>2</sub> enrichment is suspended when the growing space needs to be ventilated to maintain the temperature set point. However, enrichment up to a certain minimum ventilation rate may still be beneficial and result in increased plant growth.

Enrichment/removal systems - oxygen: Increasing the dissolved oxygen concentration of a nutrient solution can be accomplished by bubbling air or pure oxygen gas through the solution (e.g. using air stones). When using air, the maximum dissolved oxygen concentration attainable is by definition the saturation oxygen concentration (temperature dependent). When using pure oxygen gas, the maximum oxygen concentration attainable is approximately five times the saturation concentration (because air contains approximately 21% oxygen). Whether plants benefit from a super-saturated dissolved oxygen concentration in the nutrient solution has not been demonstrated, but using (pressurised) pure oxygen typically does not require additional energy to move the gas. When using air, compressors are often used and in that case care should be taken to ensure that the gas is not contaminated with lubrication particles that were inadvertently released from the mechanical components of the compressor.

#### 6.4 Liquid Water

Growing system: When watering multiple pots or large areas, the expected or measured variance in water application should be reported as should whether watering is to excess or until soils or pots are at field capacity or even left standing in water. An indication of expected uniformity or reasons that good uniformity is to be expected should be provided. The choice of watering system can affect plant growth and development, especially because some systems can increase the risk of disease as a result of wet foliage (e.g. overhead spray systems).

Environmental control system: Details about the control system if operated by e.g. water/rain sensor, leaf wetness sensor, tensiometer, soil conductivity sensor, lysimeter, or pot mass should be reported. The irrigation strategy should be reported, i.e. whether it is determined by a program based on sensor input, it is timed, it is dependent on light integral, or its timing and frequency are manually adjusted.

Irrigation systems: Type of irrigation system, frequency and timing (length and time of day) of irrigation should be reported. Type and details may include:

- Manual (by lance/wand or hose): to surface, into saucer, watering when saucer dry, when soil surface dry, watering to run off.
- Drip system: Type and manufacturer of emitter, flow rate and operating pressure, number per pot, pressure compensated.
- Underground irrigation/seep hose: Type and manufacturer, depth, layout, delivery rate per meter.
- Overhead spray: Type and manufacturer, nozzle spray pattern, delivery rate.
- Flood bench (tide): Manufacturer (if applicable), periodicity of flooding, dwell time (period of flooding and to what depth).

- Wet bench: type of wet surface (sand type and porosity), capillary matting (type and water holding capacity), how water level is maintained, details of any covering to reduce evaporation slope.
- Hydroponic or NFT system: Details of the system should include growing media, flow rates, slope, oxygenation when relevant.
- Root misting systems: Details of system, volumes of water, size of root enclosure, droplet size used, whether misting is intermittent and, if so, its frequency.
- Weighing lysimeter: Details of system, plants watered to mass, either automatically or manually to maintain soil water potential.

Details of the irrigation system should include estimated delivery of water per application per pot or square metre, as appropriate, per day. In hydroponic systems water flow volumes over plant roots may be reported. Volumes or proportion (%) of the amount of recirculated water should be included.

### 6.5 Nutrition

**Growing system:** Delivery method and the location of the release of nutrients in the root environment can have an effect on nutrient distribution and root growth. For example, top-down (e.g. overhead irrigation) or bottom-up (e.g. ebb and flood irrigation) will affect the movement of water and nutrients through the growing media and can result in zones with higher or lower nutrient concentrations. The type of growing media used can affect the availability of nutrients for uptake by the plants (e.g. certain nutrients will bind to clay particles, making them less available to plants). Hydroponic growing systems are typically designed to eliminate the possibility that the availability of water and/or nutrients becomes rate limiting. When growing in pots, researchers should be mindful that size, shape, material and even surface colour can influence root zone conditions and this should be considered when measuring and reporting plant responses.

**Environmental control system:** Details about the nutrient control system (if used) should be reported. When data are available, the nutrient uptake rates should be reported. The temperature of the root zone will have an effect on the uptake rate of nutrients by roots, and is therefore more carefully controlled for some crop species. In some cases, the root zone temperature is actively controlled.

**Distribution systems:** Liquid nutrient applications can be incorporated into the irrigation system (in that case, these systems are sometimes called fertigation systems). In most cases, small volumes of concentrated nutrient solutions are injected into the irrigation stream at a rate that ensures the proper dosing of the nutrients. Combinations of nutrients or individual nutrients can be injected this way, depending on the design and complexity of the nutrient delivery system. The injection can be passive (e.g. using a venturi system), or active (e.g. using an injection pump). Proper mixing is needed to ensure uniform distribution of the nutrients throughout the growing system. Adequate supply of nutrients throughout the rooting environment is critical since the uptake of nutrients by the roots will result in a gradient that could result in depleted regions.

## How to report experimental conditions

Here is an example of a report suitable for publication:

“The experiment was conducted in a research greenhouse section (located at 40°N, 90°W at 180 m elevation above sea level) with a floor area of 20 by 20 m and a bench area of 240 m<sup>2</sup> elevated 0.75 m above the floor. The ridge of the greenhouse section was oriented North-South. The section was bounded to the East and West by identical, but differently controlled greenhouse sections and to the North by a central corridor. The eave (gutter) height was 2.5m and the ridge extended 7.2m above the floor. The greenhouse was mechanically ventilated with intake fans located in the South-facing side wall and capable of providing a maximum air flow rate of 0.06 m<sup>3</sup> s<sup>-1</sup> m<sup>-2</sup> of floor area. Except for an entry door, the entire North-facing side wall was outfitted with 18.9 m<sup>2</sup> of louvered ventilation opening and covered with an evaporative cooling pad and an insect screen designed to exclude white flies. The greenhouse section was clad with 4-mm thick tempered glass and outfitted with an automated shade curtain with a manufacturer’s specified reduction in light transmission of 50%. The greenhouse was equipped with a supplementary lighting system powered by high-pressure sodium lamps and capable of providing a PAR flux of 200 (s.d. ±20) μmol m<sup>-2</sup> s<sup>-1</sup> at a distance of 1.75 m below the bottom of the luminaires. During the light period, the supplementary lighting system was turned on when the sunlight radiation flux was below 300 μmol m<sup>-2</sup> s<sup>-1</sup> at bench level, and turned off when the PAR flux reached 750 μmol m<sup>-2</sup> s<sup>-1</sup>. A 15-minute control delay was used to prevent excessive cycling of the supplementary lighting system. The greenhouse section was heated by a hot-water distribution system consisting of overhead pipes, perimeter pipes and tubes embedded in the solid concrete floor. Air temperature at canopy height was maintained at 20/15 (s.d. ±1)°C Day/Night. A 16-hour photoperiod was imposed and centred on solar noon by operating the supplementary lighting system when no sunlight was available. The vapour pressure deficit (VPD) in the greenhouse section was maintained by operating a stand-alone humidifier or by employing the heat-and-vent control strategy. The VPD was continuously maintained at 1.0 (s.d. ±0.1) kPa. In the control hierarchy, VPD control had priority over temperature control. All sensors and instruments were calibrated annually following manufacturers’ specifications. The greenhouse section was outfitted with two horizontal airflow fans that operated continuously to improve mixing of the greenhouse air.

Starting on March 1, a geranium (*Pelargonium x hortorum*, cv. Maverick Red) crop was grown in the greenhouse section. Two cuttings were placed in a peat-vermiculite soil-less mixture (1:1 sphagnum peat:vermiculite by volume, and amended with 0.5 kg of ground dolomitic limestone per m<sup>3</sup> of mixture) contained in 0.75 L plastic pots. Each pot received a dose (at an equivalent rate of 3 kg m<sup>-3</sup>) of slow release fertiliser (10-10-10) and was irrigated with a drip system fed with deionised water. The initial pot density on the benches was 64 m<sup>-2</sup> for the first six weeks, followed by a density of 36 m<sup>-2</sup> for the remainder of the experiment. The experiment was terminated on May 15.”

This reporting example describes only those parameters that the researcher deemed important for the particular experiment conducted. While the information reported may be sufficient for the intended audience (e.g., fellow researchers doing similar experiments), other researchers may find the report insufficient, in particular when they study environmental parameters not specifically discussed in the report. Therefore, researchers are strongly encouraged to consider carefully which parameters to measure and report on and the impact this decision may have on the usefulness of their work to other researchers including those not directly involved in their field of interest.

Table 2a: Primary Parameters - Minimum Set

What to measure	Units	Where to measure	When to measure	What to report
<b>Air temperature</b>	°C	At canopy level, in centre of growing area. Location of sensor is crucial, and should be independent of the greenhouse temperature control sensor. Include another sensor for outside temperature.	Preferably continuous, but at least hourly.	Mean and standard deviation for light and dark periods. Number of locations (preferably more than one).
<b>Substrate temperature</b>	°C	Inside plant container for solid & liquid substrates.	As above.	As above.
<b>Radiation (PAR<sup>1</sup>)</b>	μmol m <sup>-2</sup> s <sup>-1</sup>	At top of canopy, in centre of growing area.	Preferably continuous, but at least hourly.	Mean and standard deviation. Number of measurement locations (preferably more than one). When used, supplementary radiation sources (type, model and manufacturer, distribution, energy consumption, conversion efficiency), and their duration of operation.
<b>Photoperiod</b>	h		Daily or when conditions change.	Duration of light period (including any night interruption).
<b>Atmospheric Moisture:</b> relative humidity or vapour pressure deficit (VPD)	% or kPa	At canopy level, in centre of growing area and independently of the greenhouse humidity control sensor.	Preferably continuous, but at least hourly.	Mean and standard deviation for light and dark periods. Number of locations (preferably more than one).
<b>Atmospheric CO<sub>2</sub> concentration<sup>2</sup></b>	μmol mol <sup>-1</sup>	At canopy level, at a representative location.	Preferably continuously, but at least hourly if CO <sub>2</sub> enrichment is used.	Mean and standard deviation. Number of measurement points and their location relative to the plant canopy.
<b>pH</b>		In root zone environment, or in nutrient solution directly applied to the root zone environment.	Preferably continuous, but at least hourly.	Mean and standard deviation. Location of measurement(s).

<sup>1</sup> Referred to as photosynthetically active radiation (PAR: 400 - 700 nm) for general usage & described as photosynthetic photon flux density (PPFD) by many journals, professional societies & manufacturers of quantum sensors.

Table 2a contd: Primary Parameters - Minimum Set

What to measure	Units	Where to measure	When to measure	What to report
<b>Electrical conductivity (EC)</b>	S m <sup>-1</sup>	In root zone environment, or in nutrient solution directly applied to the root zone environment.	Preferably continuous, but at least hourly.	Mean & standard deviation. Location of measurement(s).
<b>Nutrition - liquid media</b>	mmol L <sup>-1</sup>		Daily or when replenished.	Ionic concentration in added solution. Frequency of additions. Aeration if any.
<b>Nutrition - solid media</b>	mol kg <sup>-1</sup> (dry)		When added or replenished.	Nutrients and their form added to soil media. Frequency of additions.
<b>Watering</b>	Litre (L)	Growing system.	At start of experiment and when changed	Frequency, amount, duration & type of water added per unit area or per plant. Mean & standard deviation. Type of irrigation system.
<b>Plant alignment</b>		On bench/floor/hanging system.	At start of experiment and when conditions change.	Number of plants per unit area and number of re-spacings or relocations.
<b>Greenhouse properties</b>		Greenhouse	At start of experiment	Latitude and longitude. Orientation of long axis relative to compass North. Size (floor area m <sup>2</sup> , growing area m <sup>2</sup> , gutter height m, peak height m), type (free-standing, gutter connected), shape (curved roof, peaked roof). Manufacturer and model if available, indicate if it has special features (e.g. type of glazing material, ventilation system, energy/shade curtain).

Table 2b: Primary Parameters - Additional Parameters

What to measure <sup>2</sup>	Units	Where to measure	When to measure	What to report
<b>Surface temperature</b>	°C	Plant tissue: pointed at canopy or individual leaf surface; Greenhouse: pointed at structural surface.	Preferably continuous, but at least hourly.	Mean & standard deviation. Location and orientation of the sensor. Distance of the sensor to the surface measured. Field of view of the sensor.
<b>Radiation (Net)</b>	W m <sup>-2</sup>	At top of canopy, in centre of growth area.	As above.	Mean & standard deviation. If measured, also report solar radiation so that (net) long wave radiation can be determined.
<b>Radiation (Spectral)</b>	μmol m <sup>-2</sup> s <sup>-1</sup> nm <sup>-1</sup>	As above and/or where of interest (e.g. within the canopy).	As often as practical.	Mean & standard deviation. Absolute or relative contribution of a specific wavelength or waveband to the overall radiation.
<b>Irradiance (Solar)</b>	W m <sup>-2</sup>	Outside (unobstructed) and/or inside (at top of canopy, in centre of growing area).	Preferably continuous, but at least hourly.	Mean & standard deviation. When possible, calculate (average) transmission of radiation through the greenhouse cover.
<b>Radiation (Integral)</b>	MJ m <sup>-2</sup> or mol m <sup>-2</sup> d <sup>-1</sup>	Calculated from accumulated data.	Continuously during the measurement interval.	Accumulated (typically daily) values. Relative contributions of supplementary and solar radiation to (daily) integral.
<b>Air circulation</b>	m s <sup>-1</sup>	At canopy level.	At start of experiment and more frequently if conditions change.	Mean & standard deviation. Design of circulation system. Predominant direction of flow. Number of measurement points & their location relative to the plant canopy. Report whether open or closed greenhouse.
<b>Substrate water content.</b> Volumetric or gravimetric water content, or matric potential	% or kPa	Substrate	Daily	Mean & standard deviation. Number of measurement locations.
<b>Dissolved oxygen</b>	mg L <sup>-1</sup>	In root zone environment, or in nutrient solution directly applied to the root zone environment.	Preferably continuous, but at least hourly.	Mean & standard deviation. Location of measurement(s).

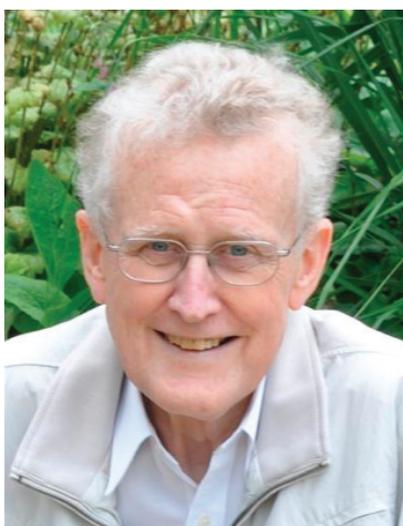
<sup>1</sup> Referred to as photosynthetically active radiation (PAR: 400 - 700 nm) for general usage & described as photosynthetic photon flux density (PPFD) by many journals, professional societies & manufacturers of quantum sensors.

<sup>2</sup> Report if records are available, and always when it is a variable under investigation.

## Acknowledgements

These guidelines are the result of a 6-year project undertaken by members of the International Committee for Controlled Environment Guidelines (ICCEG) and its parent organizations the UK Controlled Environment Users' Group (CEUG), The North American Committee on Controlled Environment Technology and Use (NCERA-101) and the Australasian Controlled Environment Working Group (ACEWG). We want to thank all of our members for the valuable input they have provided during this process. The initial idea for these guidelines came from Mr. Mark Romer and the broad framework was suggested by Dr. Royal Heins.

## Dedication



Lynton Douglas Incoll BAgSc, PhD

1937 - 2012

These guidelines are dedicated to the memory of Lynton Incoll.

Lynton championed the use of plant growth rooms and controlled environment facilities over a period of almost 50 years during which rapid advances in instrumentation and experimental protocols took place. Since 2001, he served as convener of the UK Controlled Environment Users' Group (UK-CEUG), and in that year he organized the first international meeting that hosted the North-American NCERA-101 Committee on Controlled Environment Technology and Use and the Australasian Controlled Environment Working Group (ACEWG) in Norwich, UK. He served in a leadership role for the International Committee on Controlled Environment Guidelines (ICCEG) from 2001 through 2012. He was the inspiration for the publication by that committee in 2004 of guidelines that provide a set of standards for the measurement and reporting of experiments conducted in plant growth rooms and chambers. This publication was followed in 2008 by similar guidelines for experiments conducted in plant tissue culture facilities. His work was recognized through a Significant Contributor Award at the 2008 international meeting hosted by the NCERA-101 committee in Cocoa Beach, FL. While Lynton was not able to see the completed version of the greenhouse guidelines, his contributions can be found throughout this document.

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## Additional considerations

### Introduction

Conditions in controlled environment plant growth rooms and chambers (CE units) should be reported in detail for comparison of results and duplication of experiments. The minimum guidelines table, along with these notes, should help meet these aims, indicating a required, minimum amount of information that should be reported. They may also highlight parameters that could be important, but that may not have been considered for measurement.

Average measurements should be reported, including their temporal standard deviation (s.d.).

All sensors should be calibrated regularly according to manufacturer's procedures and suggested frequency.

### Radiation

- Output of all electric radiation sources decreases with hours of operation e.g. for some fluorescent lamps output may drop 20% after the first five months of use.
- Irradiance varies significantly across the growing area in many CE units.
- Vertical radiation gradients occur in all CE units, depending on chamber size, lamp type, lamp distribution, and luminaire shape.
- Spectra from electric lamps generally differ from that of the sun. Unnatural red to far-red light ratios may affect morphogenesis in some plants and photomorphogenic effects should be considered when interpreting results.

### Temperature

- Differences may exist between the temperatures of the air and plant, especially under high radiation loads.
- Older on-off control systems can result in as much as  $\pm 5^\circ\text{C}$  variation from the set point temperature.
- A vertical temperature gradient occurs in most CE units, depending on airflow rates and other factors.

### Atmospheric moisture

- Air humidity affects plants in CE units directly (via transpiration and gas exchange) and indirectly (via the plant's energy balance and physical and biological environment).
- Heating and cooling cycles lasting only 1 to 3 minutes can change absolute humidity by 1 to 2%, altering relative humidity by 20 to 40%.
- Air humidity is a challenging parameter to monitor, but is critical to plant water relations and infection by foliar pathogens. Relative humidity (RH) is acceptable for

reporting humidity until CE units can control vapour pressure deficit (VPD), or portable instruments are available to measure and display VPD.

### Carbon dioxide

- Carbon dioxide ( $\text{CO}_2$ ) is probably the least controlled environmental parameter in CE studies. Unfortunately, too little or too much  $\text{CO}_2$  is hard to detect until plants start to show specific symptoms.
- Small variations in  $\text{CO}_2$  can affect plant growth and development significantly. People in or around CE units, and even greenhouses, can increase  $\text{CO}_2$ , as may motor vehicles, heating systems, and other nearby sources that produce  $\text{CO}_2$ .
- Few CE units manufactured today have  $\text{CO}_2$  control or monitoring equipment installed as a standard feature. However, most do have some degree of ventilation or air exchange, and good air exchange can moderate  $\text{CO}_2$  build-up or depletion.
- Even if a CE unit is well ventilated, it is important to remember that the surrounding area with which it exchanges air should also be well ventilated.

### Experimental design issues

- Ideally, a single CE unit should be treated as a single replicate. True replication requires using multiple CE units, or repeating treatments in each unit with time, both expensive and time consuming options.
- Regular transfer of plants between CE units may be an alternative to avoid direct confounding of effects of an imposed environment with that of a CE unit.
- Repeating experiments in a CE unit with poorly controlled or monitored environmental parameters may lead to erroneous assumptions about treatment conditions and resulting data.

### Example of a report suitable for publication

The experiment was conducted in a 3 m by 4 m growth room equipped with cool white fluorescent lamps (Model 830, Philips) mounted above a clear glass barrier, and an upward airflow distribution system using sufficient outdoor make-up air to provide ambient  $\text{CO}_2$  conditions inside the room. The room air temperature was maintained at  $25/20^\circ\text{C}$  (s.d.  $\pm 2/1^\circ\text{C}$ ) during the light/dark period. The photosynthetically active radiation (PAR) at the top of the canopy was maintained at  $400 \mu\text{mol m}^{-2} \text{s}^{-1}$  (s.d.  $\pm 10 \mu\text{mol m}^{-2} \text{s}^{-1}$ ) during the 12-hour photoperiod. The relative humidity in the room was maintained at 70% (s.d.  $\pm 10\%$ ). The plants were grown in 1 L pots filled with a peat-vermiculite (2:1 volume ratio) mixture. The plants were hand watered daily with a freshly prepared nutrient solution (full strength Hoagland, pH 6).

## International Committee for Controlled Environment Guidelines

### Minimum Guidelines for Measuring and Reporting Environmental Parameters for Experiments on Plants in Growth Rooms and Chambers

Sponsored by and published for the UK Controlled Environment Users' Group, the North American Committee on Controlled Environment Technology and Use (NCR-101), and the Australasian Controlled Environment Working Group



# Minimum Guidelines for Measuring and Reporting Environmental Parameters for Experiments on Plants in Growth Rooms and Chambers

International Committee for Controlled Environment Guidelines

Parameter to measure	Units <sup>1</sup>	Where to measure	When to measure	What to report
<b>Radiation</b> <i>and</i> <b>Photosynthetically active radiation (PAR)</b> <sup>2</sup>	$\mu\text{mol m}^{-2} \text{s}^{-1}$	Top of plant canopy in centre of growing area	At start and end, and every 2 weeks of the experiment	Average and standard deviation. Radiation source (type, model, and manufacturer)
<b>Photoperiod</b>	h			Duration of light and dark periods
<b>Temperature</b>	$^{\circ}\text{C}$	Top of plant canopy in centre of growing area	Daily during each light and dark period, at least 1 hour after light/dark change	Average and standard deviation
<b>Liquid culture</b>	$^{\circ}\text{C}$	Within solution under plants	As above for air temperature	Average and standard deviation
<b>Atmospheric moisture</b> <i>or</i> <b>Water vapour pressure deficit (VPD)</b>	kPa	Top of plant canopy in centre of growing area	Daily during each light and dark period, at least 1 hour after light/dark change	Average and standard deviation
<b>Relative humidity (RH)</b>	%	As above for VPD	As above for VPD	Average and standard deviation
<b>Carbon dioxide</b> <sup>3</sup>	$\mu\text{mol mol}^{-1}$	Top of plant canopy	At least hourly	Average and standard deviation
<b>Air velocity</b> <sup>3</sup>	$\text{m s}^{-1}$	At one or more representative canopy locations	At least once during the experiment	Average and standard deviation
<b>Watering</b>	litre (L)		Daily	Frequency, amount and type of water added
<b>pH</b>	pH	In the bulk solution	Before and after pH correction	Average and standard deviation
<b>Electrical conductivity (EC)</b> <sup>3</sup>	$\text{S m}^{-1}$	In the bulk solution	Before and after EC correction	Average and standard deviation
<b>Substrate</b>			At start of the experiment	Type and volume per container, components of soil-less substrate, container dimensions
<b>Nutrition</b>	$\text{mol kg}^{-1}$ (dry)		When added or replenished	Nutrients and their form added to soil media
<b>Liquid culture</b>	$\text{mmol L}^{-1}$		Daily, or when replenished	Ionic concentration in initial and added solution. Aeration if any. Volume of initial solution
<b>Specifications</b>				Floor area. Manufacturer and model if available
<b>Barrier beneath lamps</b>				Indicate if present and its composition
<b>Air flow</b>				Indicate whether up, down or horizontal

<sup>1</sup> Report in either multiples or sub-multiples of indicated units if more convenient.

<sup>2</sup> Referred to as photosynthetically active radiation (PAR: 400-700 nm) for general usage and described as photosynthetic photon flux density (PPFD) by many journals, professional societies, and manufacturers of quantum sensors. When diurnal PAR is ramped, integrals should be reported, e.g. in  $\text{mol m}^{-2} \text{d}^{-1}$ .

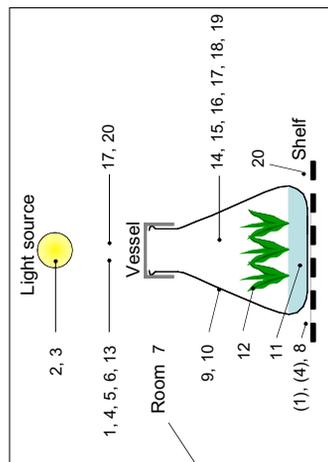
<sup>3</sup> This parameter should be reported if records are available and always when it is a variable under investigation. For more information, consult the detailed guidelines published as ANSI/ASAE Engineering Practice EP411.4 (2002) 'Guidelines for measuring and reporting environmental parameters for plant experiments in growth chambers'. ASAE, 2950 Niles Road, St. Joseph, MI 49085-9659, USA.

### Introduction to guidelines

Tissue culture is a very significant tool for plant propagation and biotechnology and is a research technique for plant physiology and molecular biology. Facilities vary from low-tech equipment through off-the-shelf incubators to state-of-the-art suites of cabinets and rooms. In all cases, accurate environmental records are essential to standardise and maximise growth of cultures and to facilitate valid replication of experiments between different facilities.

The primary critical parameters common to most plant tissue culture facilities can be monitored and recorded relatively easily. The Table (over page) gives guidance on how to monitor and record these primary parameters, most of which ideally should be monitored at the location of the cultures (see Figure below). Facilities with automatic control systems usually measure temperature at the air recirculation intake, which can differ significantly from that

### Where to measure parameters



### Key to Figure:

Primary parameters		Specialist parameters	
1.	Radiation	14.	Air temperature
2.	Light source - properties	15.	Atmospheric moisture
3.	Photoperiod	16.	Radiation
4.	Air temperature	17.	Spectral distribution of radiation
5.	Atmospheric moisture	18.	CO <sub>2</sub> concentration
6.	Air circulation	19.	Air exchange rate of vessels
7.	Room - properties	20.	Air velocity
8.	Shelf - properties	<b>Notes:</b>	
9.	Vessel - properties	Shelf level = as close as possible to	
10.	Vessel - alignment	top of shelf. Vessel level = above but	
11.	Culture medium	as close as possible to top of vessel.	
12.	Number of explants	Specialist parameters 14 to 19 are	
13.	CO <sub>2</sub> concentration	measured inside a vessel. ( ) = optional	

## International Committee for Controlled Environment Guidelines

### Guidelines for Measuring and Reporting Environmental Parameters for Experiments in Plant Tissue Culture Facilities

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above a shelf. If several shelves are used in an experiment, then each shelf should be monitored.

Many research facilities have more elaborate recording equipment, or may be able to record a wider range of experimental parameters. The real environment of a tissue culture is inside the vessel. The most advanced facilities may have equipment available to carry out extremely detailed and technically difficult measurements inside vessels, including spectral distribution of radiation. The thirteen primary parameters and the seven most important specialist environmental parameters are identified and the location of their measurement is illustrated (see Figure). For details on the specialist parameters and their interactions with primary parameters see Fujiwara and Kozai (1995)<sup>1</sup>. If specialist parameters are measured they should be reported.

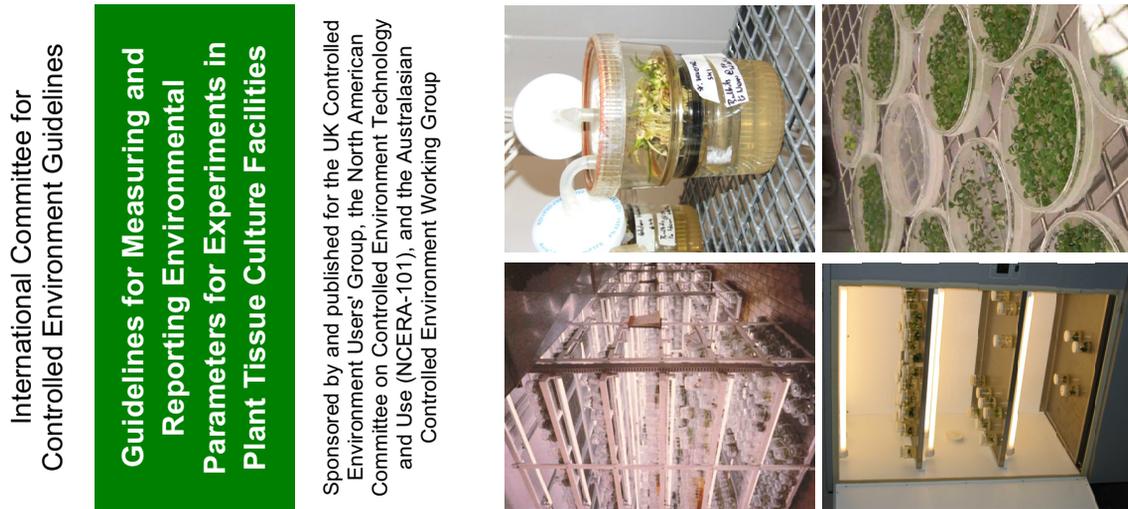
### How to report your experimental conditions

Here is an example of a report suitable for publication:

"The experiment was conducted in a walk-in growth room (model, manufacturer) (11.2 m<sup>2</sup> floor area and 2.1 m ceiling height), with horizontal air circulation through perforated sidewalls and four stacked steel-mesh shelves (24 m<sup>2</sup> total shelf space). Sufficient outdoor make-up air was provided to maintain ambient CO<sub>2</sub> concentrations in the room. Cool white fluorescent lamps (model, manufacturer) mounted 40 cm above each shelf provided an average photosynthetically active radiation (PAR) of 50 (s.d. ±7) μmol m<sup>-2</sup> s<sup>-1</sup> above the culture vessels during the 16-h photoperiod. Air temperature above the culture vessels was 25/20 (s.d. ±1)°C during the light/dark period. Relative humidity above the culture vessels was 67 (s.d. ±10)%.

Ten plantlets were cultured in 200 mL glass Erlenmeyer flasks sealed with translucent plastic film. Each flask contained 40 mL of medium with Murashige and Skoog (1962) basal components, 30 g L<sup>-1</sup> sucrose, 5 g L<sup>-1</sup> of activated charcoal and 8 g L<sup>-1</sup> agar. The pH of the medium was adjusted to 5.8. The flasks were in a single layer on each shelf with sufficient spacing to allow adequate air movement around each flask. No environmental parameters were recorded inside the flasks."

<sup>1</sup> Fujiwara, K. and Kozai, T. (1995) Physical microenvironment and its effects p. 319-369. In: J. Aitken-Christie, T. Kozai and M.A.L. Smith (eds) Automation and Environmental Control in Plant Tissue Culture, Kluwer Academic Publishers, Dordrecht, Netherlands.



## Measuring and Reporting Environmental Parameters for Experiments in Plant Tissue Culture Facilities: Table of Primary Parameters

International Committee for Controlled Environment Guidelines

What to measure	Units	Where to measure	When to measure	What to report
<b>Radiation (PAR)<sup>1</sup></b>	$\mu\text{mol m}^{-2} \text{s}^{-1}$	a) At vessel level, at uniform height throughout. (See Figure) b) Optional, at shelf level, at centre of empty shelf <sup>3</sup>	At start of experiment, and every 4 weeks <sup>2</sup> As above	Mean and standard deviation. Radiation sources (type, model and manufacturer, and distance from shelf) As above
<b>Photoperiod</b>	h		At start of experiment	Duration of light and dark periods
<b>Air temperature</b>	$^{\circ}\text{C}$	a) At vessel level Location of sensor is crucial, and should be independent of the facility's temperature control sensor b) Optional, at shelf level, at centre of shelf, outside container	Daily during each light and dark period, at least 1 hour after light/dark changeovers As above	Mean and standard deviation for light and dark periods As above
<b>Atmospheric moisture</b> (relative humidity or vapour pressure deficit)	%, or kPa	At vessel level and independently of the facility's humidity control sensor	Daily during each light and dark period, at least 1 hour after light/dark changeovers	Mean and standard deviation for light and dark periods
<b>Air circulation</b>		At vessel level	At start of experiment	Record whether perforated shelves, walls, ceiling, floor or ducts, and horizontal or vertical flow. Record source of fresh air
<b>Room or cabinet properties</b>			At start of experiment	Size (floor area $\text{m}^2$ , ceiling height m) and type (walk in/reach in) Manufacturer and model if available, indicate if it has special features e.g. rotating shelves, light reflectors, bottom cooling of shelves
<b>Shelf properties</b>			At start of experiment	Area ( $\text{m}^2$ ), type (solid or mesh, steel, wood or transparent), number (stacked, not slacked) and construction. Note if shelves are bottom cooled by air or water
<b>Vessel specifications</b> (It is appreciated that a range of vessels may be in use)			At start of experiment	Types (flasks, dishes, bottles, jars) and materials (glass, plastic) Size/volume (mL) Closure type and additional seal or vent
<b>Vessel alignment</b>		On each shelf	At start of experiment	Number of vessels and number of layers (if vessels are stacked) per shelf
<b>Culture medium</b>			At start of experiment	Solid, gel, or liquid (or combinations). Type and make of gelling agent. pH Volume per vessel (mL) Mineral composition (macro- and micro-nutrients) Carbon source, growth regulators, vitamins and their concentrations; also whether activated carbon and other additional substrates are in use
<b>Number of explants</b>		In each vessel	At start of experiment	Initial number of explants
<b>Atmospheric CO<sub>2</sub> concentration</b>	$\mu\text{mol mol}^{-1}$	At vessel level, at centre of shelf	Daily but only if CO <sub>2</sub> enrichment is installed within facility	Mean and standard deviation

<sup>1</sup> Referred to as photosynthetically active radiation (PAR: 400-700 nm) for general usage and described as photosynthetic photon flux density (PPFD) by many journals, professional societies and manufacturers of quantum sensors

<sup>2</sup> Fluorescent lamp efficiency declines significantly within weeks of installation and gradually thereafter and such lamps therefore require a regular monitoring and replacement programme

<sup>3</sup> If lamps are arranged at the back of the shelf rather than above the shelf this should be stated and PAR measured at the back and the front of an empty shelf





The aim of these Guidelines is to educate, assist and encourage comparativeness in reporting environmental parameters in greenhouse experiments and to provide quality assurance for researchers across the world.

International Committee for  
Controlled Environment Guidelines  
February 2016