

## **Concluding Address "Controlled environments: past achievements and future directions"**

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### **Introduction**

Controlled environments are a 20<sup>th</sup> century concept but their value was recognised in the late 19<sup>th</sup> century. In 1890, a French scientist, Claude Bernard, stated "that "every physiological experiment must be so designed that all environmental factors are constant, except for a single one, the effects of which are analysed by limiting its parameters" (Chouard 1972). A New Zealand ecologist, Leonard Cockayne also recognised the value of refrigeration in frost research about this time when he noted that a "freezing chamber offers an easy place for such experiments ... and ... valuable data as to the cold resisting powers of our plants might be arrived at" (Cockayne 1897). However, recognition of the concept and bringing the technology to enable this to be realised were still far apart, at least for another 20 years.

Two technologies needed to converge to enable controlled environments (CEs) to be developed. This was the development of refrigeration and the invention of the electric lamp, both of which also occurred in the late 19<sup>th</sup> century. Small scale facilities began to be built from the late 1920's, especially by plant pathologists (Johnson 1928). However, the first large facility was not opened until 1949 and this was the Earhart Plant Research Laboratory in Pasadena, California (Went 1950). An explosion in the number of large-scale controlled environment facilities occurred subsequently from the 1960s through to the 1970s and there has been a resurgence in building large scale facilities in the 1990s. The 20<sup>th</sup> century can be recognised as the controlled environment technology century, for the 21<sup>st</sup> century, *uses* of controlled environments rather than *technology* will be a key driver.

### **Temperature control**

It was in the 1920's that mechanical refrigeration was used for the first time to achieve a measure of control over temperature in greenhouses. Chilled brine was circulated around pipes but cold air in winter was also used. Several papers appeared in the literature at this time describing some simple controlled environment systems (Tottingham 1926; Johnson 1928). It was curious that, in spite of the widespread use of electric refrigeration in homes, according to Downs (1980) "the desirability of adapting mechanical refrigeration to temperature and humidity control in plant growth rooms seems so obvious that it is difficult to understand why (in the 1930's) so few plants scientists used the systems".

Early plant growth chambers were built at the Boyce Thompson Institute in 1928 - 1930 and Davis and Hoagland (1928) described an apparatus at the University of California at Berkeley, California (Fig. 1). In the UK, Stoughton (1930) described a CE facility at Rothamsted (Fig. 2) and the Plant Industry Station in Beltsville, Maryland had CE facilities by 1937. The best temperature control for these facilities was achieved by circulating calcium chloride brine chilled by ammonia compressors to individually controlled liquid-to-air heat exchangers.

By the 1950s, temperature control to a reasonable precision ( $\pm 1^{\circ}\text{C}$ ) had been achieved though  $\pm 0.2^{\circ}\text{C}$  had been reported. The range of temperatures for the systems were typically

10 to 30°C but frost rooms with capability to -20°C had been reported. However, spatial variability in temperature across the various facilities was not commonly reported.

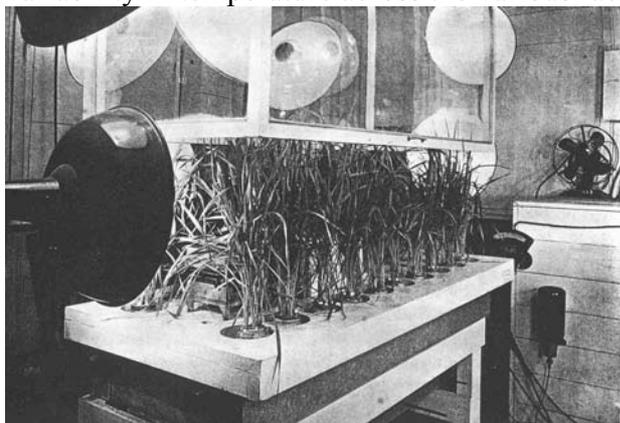


Fig. 1. A CE system at the University of California (from Davis and Hoagland 1928).

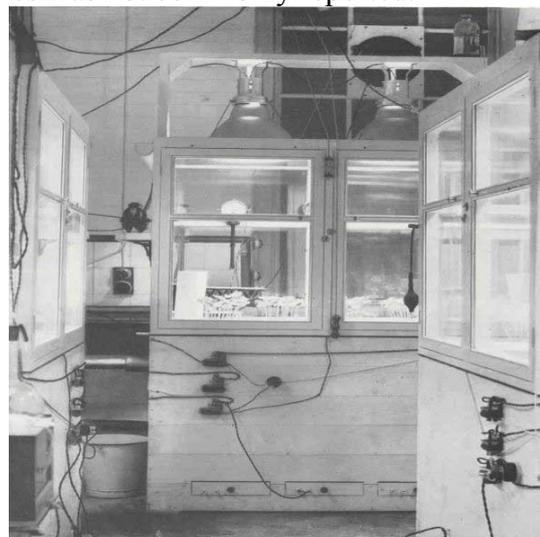


Fig. 2 An earlier CE system at Rothamsted Experimental Station (from Stoughton 1930).

### Lighting

A major obstacle to the development of plant growth chambers in the 1930's was the lack of a satisfactory light source (Downs 1980). Early incandescent lamps were low in light output, of such poor spectral quality such that plants grew adversely and heat emission was a major constraint. However, the development of the carbon arc lamp in the 1930's (Fig. 3), overcame the problem of low output and spectral balance could be achieved by mixing these lamps with incandescent lamps. On the other hand, these lamps also produced ultra-violet radiation and phytotoxic gases. This lamp was used extensively at the Plant Industry Station in Beltsville for over 30 years (Downs 1980).



Fig. 3 An example of the carbon arc lamp (from Downs 1980).

Further progress in development of plant growth chambers awaited the development of the fluorescent lamp in the 1940's. The high output/ low heat emission and broad source made them ideal for controlled environments. They were initially low in output but development of

2.5 m, high power slimline lamp in the late 1940's enabled PFDs of approx.  $300 \mu\text{mol m}^{-2} \text{s}^{-1}$  to be achieved. Continued developments into the 1960's made light intensities of 480 to  $600 \mu\text{mol m}^{-2} \text{s}^{-1}$  possible.



Figs 4 and 5. Fluorescent lamps in use at the CE facilities at the Plant Industry Station at Beltsville (from Downs 1980).

High intensity mercury vapour lamps were introduced in the late 1950s and the phosphor coatings, introduced in the 1960s, significantly improved the lamp quality for growing plants. The main advantage of these lamps is their high output, with light intensities up to  $1500 \mu\text{mol m}^{-2} \text{s}^{-1}$  readily achievable. However, these lamps also have high heat emission, thus require heat dissipation systems such as the water screen in use at the New Zealand Controlled Environment Laboratory (Fig. 6).

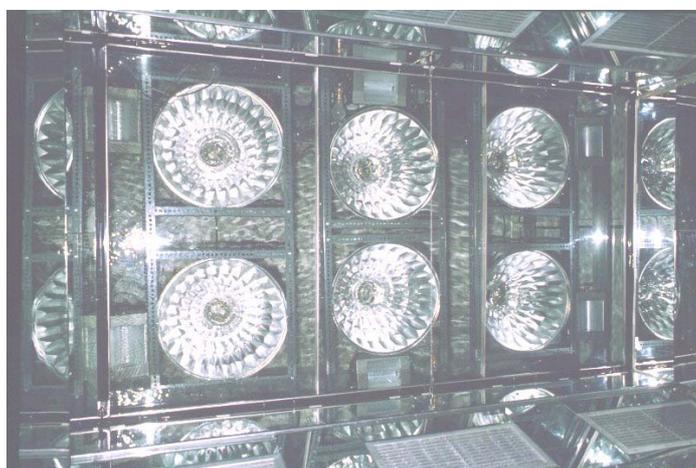


Fig. 6 The water-screened high-light rig at the NZ Controlled Environment Laboratory using High pressure discharge and tungsten halogen lamps©.

Control of light intensity of the mercury vapour or high pressure discharge lamps is desirable to simulate the diurnal change in intensity. Bingham and Coyne (1979) at the Lawrence Livermore Laboratory described a controlled ballast system to achieve some control of light intensity and further refinements were made by Bubenheim *et al.* (1995), and they described an SCR based dimming system. However, only 400 W lamps were controllable, because the arc temperature in higher wattage lamps could not be maintained. Thus the dimming system was limited in control specifications. A system for controlling the

light intensity of 1kW lamps was recently developed at the New Zealand Controlled Environment Laboratory. This was based on single lamp energy regulator and uses a multiple, high frequency AC waveform chopping to achieve control of ballast voltage. The benefit with this system is that lamp peak voltages are maintained, hence also the lamp arc temperatures. Control of the ballast is achieved by a 0 - 10V interface module.

### Water vapour (humidity) control

Humidity control was recognised as one of the most difficult factors to control within close limits well into the 1950's (Hudson 1957; Downs 1980). The range commonly reported was 48 - 80% but few facilities attempted to control humidity. There were various methods including sulphuric acid/water, atomisers and steam injection and an example from 1930 is shown in Fig. 7. Chemical dehumidifiers were introduced in the 1960s and the present generation enable precise control of humidity. For example, at the NZ Controlled Environment Laboratory, water vapour is injected by steam generators with proportional heating and dehumidifying by dehumidifiers with airflow rates up to  $270 \text{ m}^3 \text{ h}^{-1}$  (ML 270, Munters, Sollentuna, Sweden) and relative humidity measured with dew point hygrometer (M100, General Eastern, Watertown, Ma, USA) and humidity sensors (Humitter 50, Vaisala, Helsinki, Finland). The control system can be programmed to alter relative humidity over time in linear sequences (Fig. 8)

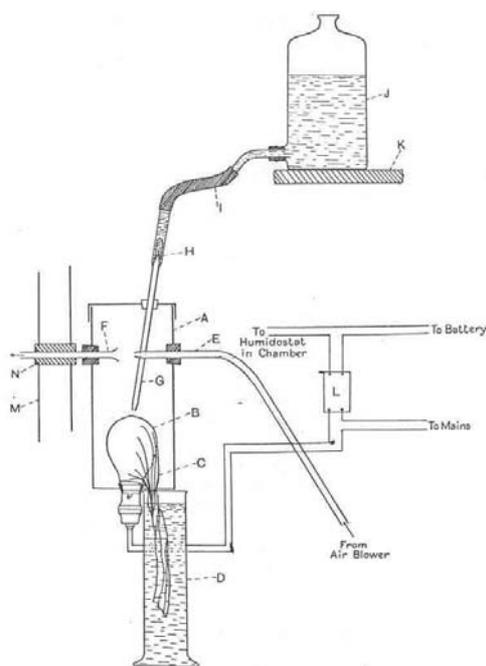


Fig. 7. Details of humidifier. A, tin; B, 8 candle-power carbon filament lamp; C, muslin covering lamp loosely; D, cylinder for water; E, air inlet to tin; F, air exit to chamber; G, tube to lead drip to lamp; H, drip jet; I, connecting tube to reservoir; J, water reservoir; K, bracket screwed to top of chamber; L, relay; M, glass walls of chamber; N, side rail of chamber.

Fig. 7 An early method of controlling relative humidity. (From Stoughton 1930)

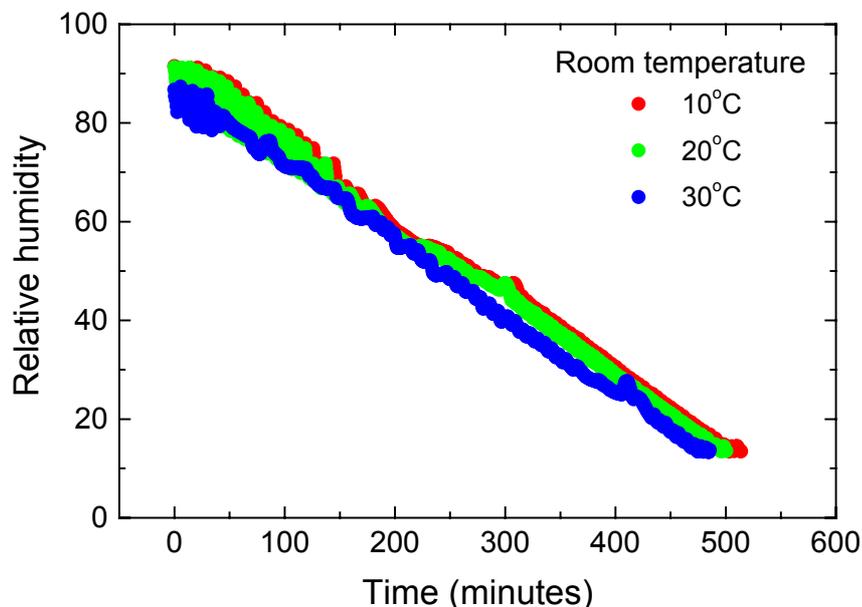


Fig. 8. Controlling relative humidity at the NZ Controlled Environment Laboratory©.

### CO<sub>2</sub> measurement and control

Infra-red gas analysers were first developed in early 1940s and used primarily to detect carbon monoxide in mines. Commercially available instruments for measuring CO<sub>2</sub> were developed in the 1950s but their use in controlled environments was not until the early 1960s. CO<sub>2</sub> injection methods also developed concurrently. CO<sub>2</sub> scrubbing has been used at selected facilities but no commercial systems are yet available. In-house systems using scrubbers with soda lime and NaOH are in use at several facilities.

### Large scale controlled environment facilities

The first large scale facility was the Earhart Plant Research Laboratory, in Pasadena CA (Fig. 9). There were many such facilities, including the Canberra Phytotron, Duke Phytotron and the NCSU Phytotron built in the 1960 -1970 period. The most recently built large facility is that at the Australian National University in Canberra, Australia but this facility is fully contained (Fig. 10).

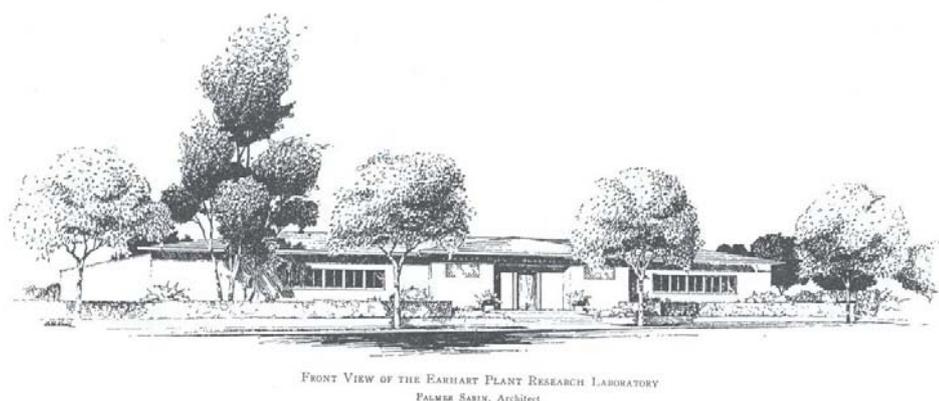


Fig. 9. The Earhart Plant Research Laboratory (from Went 1950).



Fig. 10. The newest controlled environment facility at the Research School of Biological Research at the Australian National University in Canberra, ACT, Australia©.

### Future directions

The technologies available today undoubtedly meet all the requirements for controlled environment specifications and this is the real legacy of the past century. There is no doubt that technology refinements will continue to occur, and some obvious examples include new efficient lamps, advances in humidity sensors, cheaper gas analysers and new refrigeration systems as discussed in other sessions of the meeting. In addition, the space program has been renowned for developing new technologies and the case for controlled environments was also presented at this meeting. The real future for controlled environments in the 21<sup>st</sup> century lies in the *uses* to which we put them and proving their performance specifications in quantifiable ways.

What can controlled environments do best? There are five characteristic features of controlled environments that contribute to the study of environmental impacts. These include:

- the system can be *isolated* for study under a simulated environment
- the parameters of that environment can be readily *manipulated*
- a standard environment for research purposes can be *replicated*
- responses to changes in those parameters can be *quantified*
- results can be *integrated* with knowledge of the natural environment

The future for controlled environments will be more focused on three topics; improved specification and reporting (Session 2), developing methods to transfer information from CEs to the field, that is, shifting to more complex environments (Session 6) and functional genomics (Session 1).

### Improved performance: methods used at the NZ Controlled Environment Laboratory

The methods used to measure controlled environments involve the use of statistical process control (SPC) techniques. This is a powerful technique to assess objectively the performance of control systems. Routine sampling of conditions and then the application of statistical methods are used to describe the average conditions (accuracy of control) and its variability (precision). The method is based on a population sampling approach, sampling from a

distribution plus the calculation of sample means and standard deviations. The methods can be adopted for any regularly sampled conditions.

**An example of SPC evaluation of temperature** The room temperature (day/night) was specified as  $13/3 \pm 0.5^\circ\text{C}$  for a duration of 53 days. Temperature sampling period was 10 minutes and 30 observations per record were collected throughout the entire period. The specified standard deviation ( $\pm 0.5^\circ\text{C}$ ) was equivalent to  $0.167^\circ\text{C}$ . The frequency distribution of temperature deviations about the set point for this example are shown in Fig. 11 and the frequency distribution relative to the setpoint range are shown in Fig. 12.

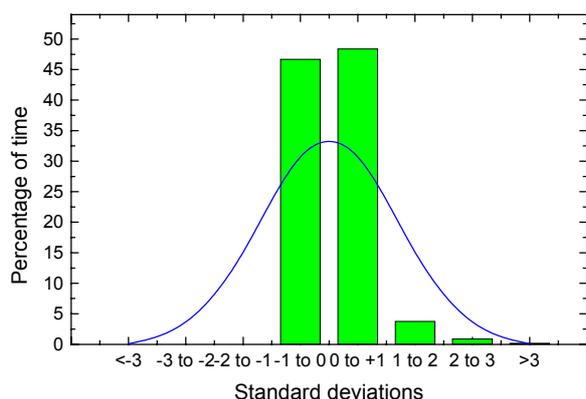


Fig. 11. Frequency distribution of temperature in relation to the standard deviation©.

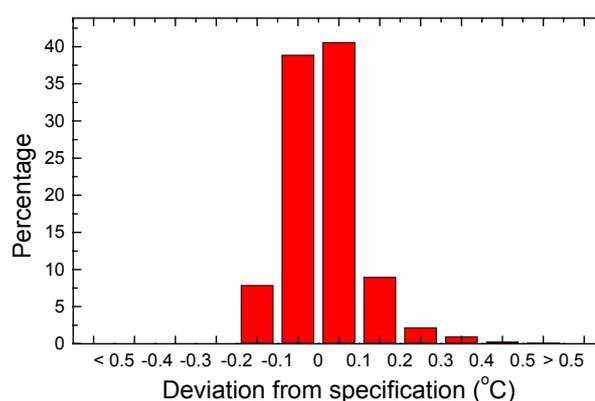


Fig. 12. Percent of time that the temperature varied from the set points by selected intervals©.

These data show that the temperature control for nearly 80% of the duration of the experiment was within  $\pm 0.1^\circ\text{C}$  while for about 96% of the time, temperatures were within  $\pm 0.2^\circ\text{C}$ . This demonstrated that the temperatures exceeded the specified limits ( $\pm 0.5^\circ\text{C}$ ) for only 0.2% of the total duration of this experiment. The analysis revealed that temperature control was better than first appreciated and that the statistical process control approach gives an objective performance assessment. Uses for this approach include providing users with detailed quantitative descriptions of the controlled environments but also a method to tune room performance control parameters, if the analysis reveals conditions deviate from the specifications for significant periods.

**An example of SPC processing of irradiance** The uses a database approach and requires specific room setup conditions such as the project, CE room, numbers and types of lamp rigs. Other information includes lamp orientation and location in X and Y co-ordinates, and the kW rating. Sensor information (e.g. Sensor, Meter/Logger, Correction Factor, Calibration) and radiation measurements (e.g. Sensor, X, Y, Z Distance, PFD) are also required. A specific example is shown in Fig. 13. Performance assessment and results of this example are that spatial variability within the CE room was  $\pm 21 \mu\text{mol m}^{-2} \text{s}^{-1}$  (3%) and the sensor uncertainty was  $\pm 33 \mu\text{mol m}^{-2} \text{s}^{-1}$  (4.8%, LiCor specification). There was temporal fluctuation of  $\pm 23 \mu\text{mol m}^{-2} \text{s}^{-1}$  (3.3%) that was caused by voltage fluctuation on the lamps. Overall, for this example, the average PFD was  $694 \pm 74 \mu\text{mol m}^{-2} \text{s}^{-1}$  (95% confidence limit).

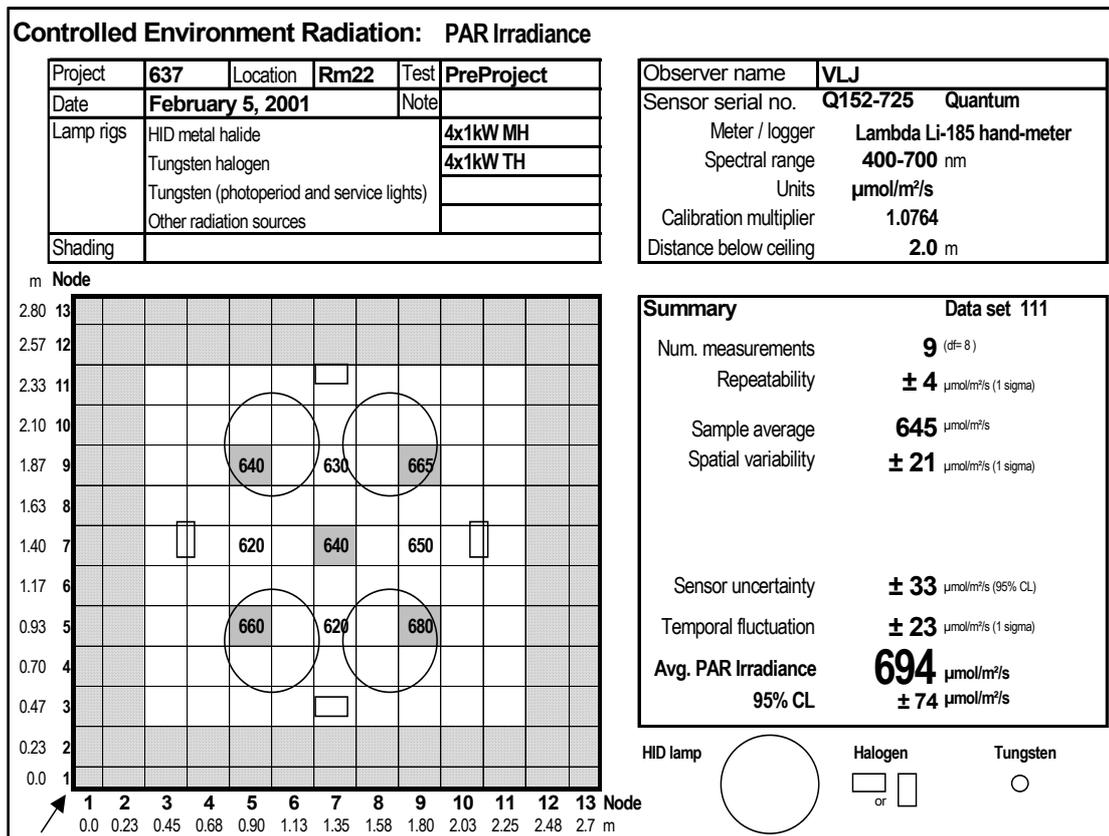


Fig. 13 An actual output from the irradiance analysis from the NZ Controlled Environment Laboratory©.

### From controlled environments to the field: developing formal methods

To study dynamic plant responses, flexible controlled environment regimes are required. In principle, controlled environments can be used to rapidly and precisely measure how changing environmental parameter levels affect system processes. However, the focus here is on environmental sensitivity in the context of their fluctuations, rather than on responses to constant parameter levels. The methodology of “systems identification” is used to assess these responses. System identification is commonly used in engineering applications such as the design of process control systems, for signal processing and for financial and economical applications. In this application, it is used to describe dynamic responses, and is an accurate, simplified mathematical model for a complex, dynamically-varying phenomenon that can be obtained from time-series data. The methods are a collection of mathematical tools used to build such models, by relating measurements of system inputs to corresponding outputs.

The requirements for this approach are firstly that very different types of controlled environment experiments are required. That is rather than static conditions, the time-dependence of the environmental relationships are themselves the subject of experimental variation and temperatures vary and are controlled within the controlled environment according to a random binary series. This, however, requires the ability to be able to measure the plant response to the changing environment. In addition, modification of room control programme strategies to deliver arbitrary time series are needed and room temperatures need to be mapped under high rates of change.

An example of how this might be achieved is shown in Fig. 14

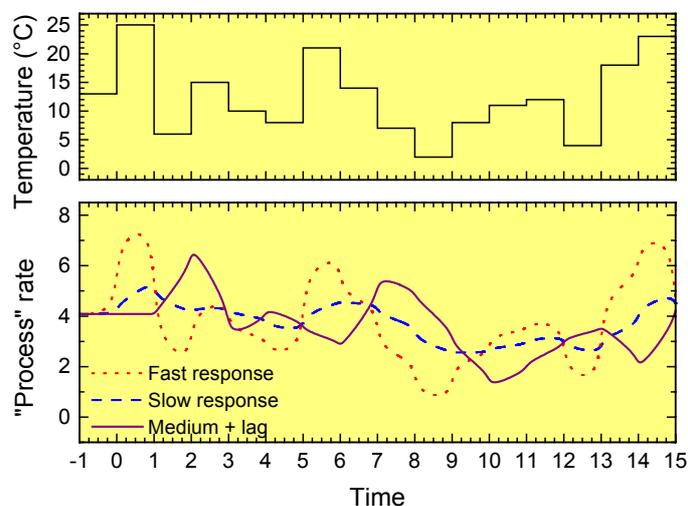


Fig. 14. An hypothetical example of varying temperature in pseudo-random steps and how changes in plant processes that vary in rate might occur in response to the stepped change©.

There are several methods available to measure plant performance at the required high frequencies. For example, video imaging which has the additional advantage of being non-contact and non-destructive. However, there are issues of precision and accuracy of images for analysis still to be addressed.

Another approach is the use of commercial systems such as the Phytomonitor (Phytech Technologies Ltd, Israel), which is an integrated plant monitoring system that includes computer plus sensors to measure such things as fruit diameter, stem diameter, stem sap flow, air temperature, air humidity, radiation, leaf temperature, temperature in the boundary layer and soil moisture.

### Use of controlled environments for functional genomics

Various aspects of the environment interact with genes and their expression to produce the phenotype. With the advances in genome sequencing, there is now a real opportunity to make progress in determining the functionality of the various genes. As has been shown in a variety of studies, genetically modified plants have proven to be of great scientific value for the study of many plants processes. To manipulate and control the expression of different traits, then alterations in both the plant genome and in the environment combine to make a very powerful tool to advance fundamental progress in our understanding of plant performance. Thus, controlled environments with containment capability become an essential component of this research. Today, many controlled environment facilities are being refitted to include containment capabilities and augurs well for the achievement of this exciting opportunity.

### Summary

Controlled environment technology has been around for about 80 years and there has been many outstanding technological achievements in lamps and refrigeration systems, microprocessors and data acquisition and the ability to measure the environmental parameters

that have brought controlled environment science to the highly advanced state of the present time. The essential ability of controlled environments to *decouple* climatic parameters makes such facilities a unique tool in science. Sequencing genomes can only be of value when functionality can be ascribed to each gene - controlled environments have an underpinning role in this research and therefore a bright future in the 21<sup>st</sup> century.

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