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# Development of a Heat Transfer Model for Plant Tissue Culture Vessels

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Air temperature within culture vessels is a dominant factor influencing the growth of plantlets. A heat transfer model for tissue culture vessels was developed in this study. The heat source for vessels was irradiance from fluorescent tubes. The energy transfer included convective and radiative heat exchanges. In order to validate the model, two types of culture vessels were studied at various light radiation levels. Type-T thermocouples connected to a data logger were used to measure temperatures. The discrepancies between the values predicted by the model and actual measurement values were less than 0·6°C, and the average absolute error was lower than 0·4°C. Increasing the surface convection coefficient by increasing ventilation velocity could effectively reduce the greenhouse effect in these small size culture vessels.


## 1. Introduction

Recently, the orchid industry has developed rapidly in Taiwan and tissue culture plantlet demand has increased annually. In order to promote the quality and quantity of plantlets, the growth environment must be maintained at optimum conditions.

Plantlets *in vitro* are cultured in a small and nearly closed system of culture vessels. Plantlets need to grow with a very low density (or absolute absence) of microorganisms. Many sets of successive horizontal shelves are arranged in the culture room, and many vessels are placed on the shelves. The important environmental factors affecting the growth of plantlets include light irradiance, air temperature, relative humidity, carbon dioxide ($CO_2$), and ethylene ($C_2H_4$) concentrations. The factors affecting root environment are the medium temperature, medium water potential, sugar concentration, and other minerals. The external environment of culture vessels and the physical properties of vessels influence the internal environment of the vessels. External environmental factors include light irradiance, air temperature, air velocity and direction. The physical factors of the vessels are the air exchange rate, heat transfer coefficient, shape, surface area, and volume. As so many vessels are placed on the shelf installed in the culture room, it is impractical to modify the internal microclimate with various kinds of equipment for each vessel. The best way to modify the internal climate of vessels is to adjust the external climate. Since the air temperature within the culture vessel is the dominant factor to influence the plantlet growth, it is very important to develop a thermal model to quantify the effect of these parameters on the internal air temperature of vessel.

The air temperature around plantlets has significant effect on growth, development, morphology and physiology of cultures. Many researchers have studied the effects of temperature on plantlets (Kozai *et al*., 1995a, 1995b; Zimmerman, 1995). Walker *et al*. (1989) modified the $CO_2$ concentration and the temperature of vessels to study the effect of ventilation on the growth of *Rhododendron*. Vanderschaeghe and Debergh (1987) noted that increased light irradiance could enhance the photosynthesis rate, although the radiation energy increased the internal temperature. In order to control the variation of internal air temperature, a cooling method to relieve the heat accumulation at the base of the vessel, was proposed.

Urban and Jaffrin (1990) developed a mathematical model for heat and mass transfers inside the tissue culture vessels. The vessel shape was cylindrical. The culture vessel was divided into five subsystems: stopper, vessel wall, medium, base, and internal air. The heat

 

**Notation**

| | | | |
|---|---|---|---|
| $A_{be}$ | basal area of vessel excluding the lid area, $m^2$ | $Q_{e1}$ | energy exchange from the air exchange of headspace, W |
| $A_{lid}$ | lid area, $m^2$ | $Q_{l1}$ | radiative exchange of headspace, W |
| $A_m$ | cross-sectional area of medium, $m^2$ | $Q_{l2}$ | radiative exchange of plantlet zone, W |
| $A_{sv1}$ | surface area of vessel excluding lid area, $m^2$ | $Q_{m2}$ | conductive heat flux between the medium and plantlet zone, W |
| $A_{svm}$ | surface area of vessel with medium excluding basal area, $m^2$ | $Q_{mb}$ | conductive heat flux between the medium and base, W |
| $A_{sv2}$ | surface area of vessel in the plantlet zone, $m^2$ | $Q_{12}$ | conductive heat flux between headspace and plantlet zone, W |
| $A_1$ | cross-sectional area of headspace, $m^2$ | $Q_{21}$ | conductive heat flux between plantlet zone and headspace, W |
| $A_2$ | cross-sectional area of the plantlet zone, $m^2$ | $Q_{2m}$ | conductive heat flux between plantlet zone and medium, W |
| $C_p$ | specific heat of dry air, J $kg^{-1}$ | $R_1$ | ratio of the irradiance in the headspace region to effective irradiance energy entering the vessel |
| $E_1$ | entrance energy in headspace from light irradiance, $W/m^2$ | $R_2$ | the ratio of the irradiance in the plantlet zone to effective irradiance energy entering the vessel |
| $E_2$ | entrance energy in plantlet zone from light irradiance, $W/m^2$ | $R_m$ | ratio of the irradiance in the medium to effective irradiance energy entering vessel |
| $E_f$ | effective irradiance energy entering vessel, $W/m^2$ | $T_1$ | air temperature of headspace, °C |
| $E_i$ | irradiance from the fluorescent tube, $W/m^2$ | $T_2$ | air temperature of plantlet zone, °C |
| $E_m$ | entrance energy in medium from light irradiance, $W/m^2$ | $T_a$ | external air temperature, °C |
| $F_{1w}$ | shape factor for the wall surface as seen from the surface of the headspace | $T_i$ | unsolved temperature in equation, °C |
| $F_{2w}$ | shape factor for the wall as seen from surface of the plantlet zone | $T_m$ | air temperature of medium, °C |
| $H_l$ | convective transfer coefficient of lid, $W/m^{2\circ}C$ | $T_{1k}$ | internal air temperature, K |
| $H_m$ | conductive coefficient of medium, $W/m^{2\circ}C$ | $T_{wk}$ | vessel wall temperature, K |
| $H_v$ | convective transfer coefficient of vessel's outside wall, $W/m^{2\circ}C$ | $V_1$ | volume of headspace, $m^3$ |
| $M_{12}$ | air exchange rate between headspace and plantlet zone | $\Delta T$ | temperature difference between internal and external air, °C |
| $M_{21}$ | air exchange rate between plantlet zone and headspace | $\tau_{rv}$ | transmittance of the vessel's wall |
| $N$ | air exchange rate for culture vessel, $min^{-1}$ | $\tau_{lid}$ | transmittance of the lid |
| $P_i$ | predictive errors | $\tau_{lw}$ | long wave transmittance of the vessel |
| $P_c$ | predictive performance criteria | $\varepsilon$ | long wave emissivity of heatspace |
| $Q_{c1}$ | convective exchange of headspace, W | $\sigma$ | stephan–Boltzman constant |
| $Q_{c2}$ | convective exchange of plantlets zone, W | $\rho$ | air density, $kg/m^3$ |
| $Q_{cm}$ | convective exchange of medium, W | | |

transfer condition included conduction, convection, and radiation. Plantlets were not considered in this model. Only three sets of data were measured to evaluate the model.

Tani *et al.* (1995) developed a heat balance model based on finite element methodology for a cylindrical culture vessel. Three temperatures: culture vessel surface, internal air, and medium, were selected as the predicted parameters. Plantlets were not considered in this model. Six temperature data from the experimental design with two irradiance levels and three external air temperatures were measured to evaluate their model. The difference between predicted values and measured values were 0.5°C. Suroso *et al.* (1995) did similar research using a different experimental design with three irradiance levels and two external air temperatures. Their maximum deviation between simulated results and actual measured values was 0·9°C. Suroso *et al.* (1996) continued the same study, using an inverse calculation technique for analysis of convective heat transfer over the surface of culture vessel. These convective transfer coefficients were incorporated into their previous model

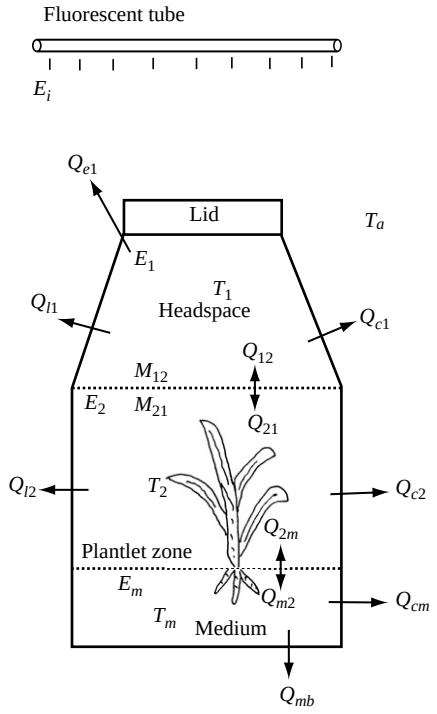


*Fig. 1. Schematic diagram of the heat transfer model for tissue culture vessel; $E_i$, irradiance from the fluorescent tube; $Q_{e1}$, energy exchange from the air exchange of the headspace; $T_a$, external air temperature; $E_1$, entrance energy in the headspace of short wave radiation from light irradiance; $T_1$, air temperature of headspace; $Q_{l1}$, radiative exchange from headspace; $Q_{c1}$, convective exchange of headspace; $M_{12}$, air exchange rate between headspace and plantlet zone; $Q_{12}$, energy exchange between headspace and plantlet zone; $E_2$, entrance energy in plantlet zone from light irradiance; $M_{21}$, air exchange rate between plantlet zone and headspace; $Q_{21}$, energy exchange between plantlet zone and headspace; $Q_{l2}$, radiative exchange from plantlet zone; $T_2$, air temperature of plantlet zone; $Q_{c2}$, convective flux of plantlet zone to external air; $Q_{2m}$, conductive heat flux between the plantlet zone and medium; $E_m$, entrance energy in medium from light irradiance; $Q_{m2}$, conductive heat flux between the medium and plantlet zone; $T_m$, air temperature of medium; $Q_{cm}$, convective flux of medium to external air; $Q_{mb}$, conductive heat flux between medium and vessel base*

to improve the predictive ability. The maximum difference between simulating results and actual measurements was 1·3°C and the minimum difference for these values was 0·1°C.

In all previous studies, only the empty culture vessel was used to develop the heat transfer model, and the effect of plantlets was not considered. The objective of this study was to develop a heat balance model to predict the temperature distribution including the plantlet effect in the culture vessels. Experimental data were collected and used to validate the predictive ability of model.

## 2. Materials and methods

### 2.1. *Culture vessels*

Two types of vessels, a conical flask (F-1, I-Shin Co., Taiwan) and a Japanese irregular box (IW-1, Iwan Co., Japan) were selected to measure the internal air temperature distribution, as shown in *Figs 2* and 3 (see also *Fig. 1*).

### 2.2. *Measuring device*

#### 2.2.1. *Temperature sensors*

The type-T thermocouple wires (Omega Engineering, USA) were selected to measure the temperature. The diameter of wires was 0·25 mm. The sensing points were placed in the external air, headspace, air surrounding the plantlets, medium, and wall of the vessels. The connected point of external air temperature wire was

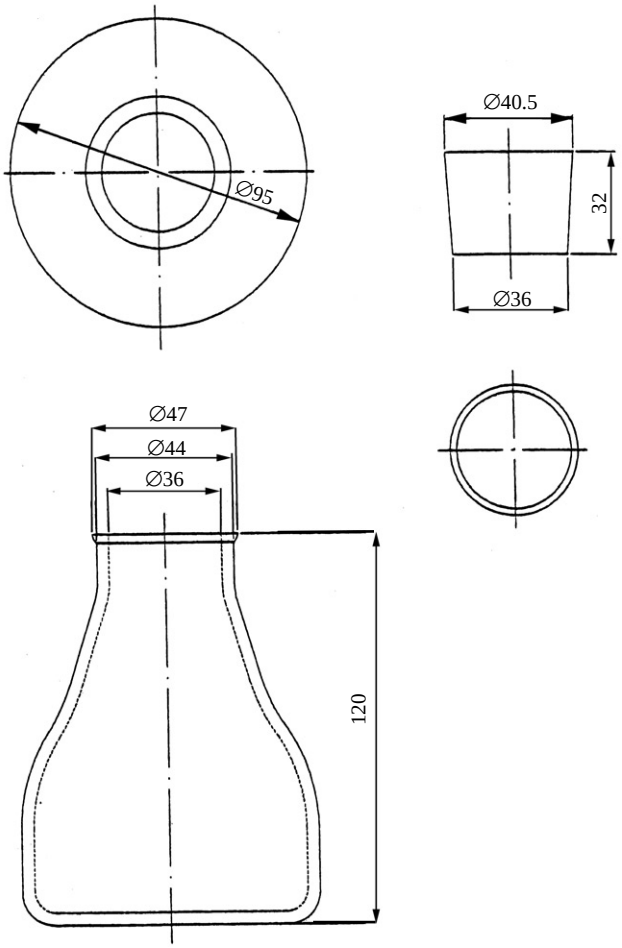


*Fig. 2. Schematic diagram and size of conical flask; all dimensions in mm*

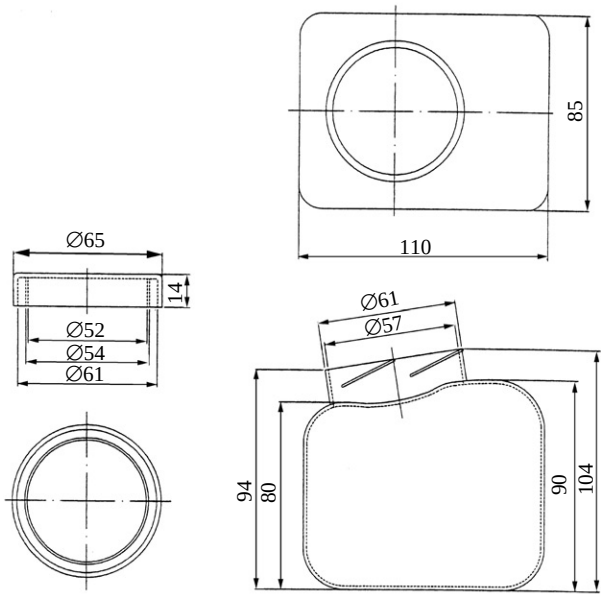


*Fig. 3. Schematic diagram and size of Japanese irregular vessel; all dimensions in mm*

covered by aluminium paper to shield the radiation energy from fluorescent tubes.

These thermocouple wires were connected to a Delta-T2e data logger (Delta devices, LTP, UK). The voltage signals were then transformed into temperatures in °C and recorded. All thermocouple wires were calibrated using a calibrator (TC-2000, Instrutek AS, Norway). The accuracy of these temperature sensors was within 0·1°C after calibrating.

2.2.2. *Irradiance*

Radiative energy in $W/m^2$ from fluorescence tubes was measured by LI-200SA pyranometer (Li-COR Co., USA). The measuring range of wavelength was from 300 to 1100 nm.This pyranometer was calibrated by a Kipp & Zonen Model CM11 thermopile pyranometer (Kipp and Zonen Ltd, The Netherlands). The accuracy of the meter was $\pm 3\%$. A small size pyranometer (K-11 type, Sanyo, Japan), was used to measure internal irradiance energy of vessels in different positions.

2.2.3. *Air velocity*

The air velocity near vessels was measured with a hot-wire anemometer (Sweta-30, Sweta, Sweden). The accuracy of this device was $\pm 0{\cdot}2$ m/s.

## 2.3. *Experimental procedures*

Two types of vessels were placed on the horizontal shelves. Four fluorescent tubes, Philips TLD 36w/39, were installed under the upper shelves. The length of tubes was 120 cm. The space between tubes was 20 cm. The distance between tubes and vessels was 30 cm.

Radiative heat flux was measured at the top position of vessels. The transmittance of the vessel walls and lids were respectively determined using empty vessels. The small size pyranometer (K-11 type), 8 mm in diameter and 12 mm in length, was sterilised to avoid contamination by microorganisms. The sensor surface was treated with 80% alcohol, placed in the laminar flow cabinet, and then exposed to ultraviolet (UV) light for 1 h. The vessel lids were removed and the pyranometer was placed to the predetermined height of vessel within the laminar flow cabinet. The measured values were then used to calculate the $R_1$, $R_2$, and $R_m$ values, where $R_1$ is ratio of the irradiance in the headspace region to the effective irradiance energy entering the vessel, $R_2$ is the ratio of the irradiance in the plantlet zone to the effective irradiance energy entering the vessel, and $R_m$ is the ratio of the irradiance in the medium to the effective irradiance energy entering the vessel.

A small hole was drilled in the lid through which thermocouple wires were passed. The hole was then sealed using silicon. The thermocouple wires and lid were sterilised in the same way as the pyranometer. Vessels with plantlets were then transferred into the laminar flow cabinet. The lids were removed and replaced with new modified lids in which thermocouple wires were set; and the thermocouple wires were placed in the predetermined position.

After installing all thermocouple wires, the vessels were removed from the laminar flow cabinet and placed back to the shelves.

## 2.4. *Parameter estimation*

The relevant physical parameters for two culture vessels are listed in Tables 1 and 2. The following parameters were used.

(1) Transmittance was calculated from measured values.
(2) Ratio of the irradiance ($R_1$, $R_2$, and $R_m$) were calculated from measured values.
(3) Exchange rate $N$ was adopted from Chen (2002).
(4) Energy exchange rate $M_{12}$ was adopted from Kitaya *et al.* (1955).
(5) Convective transfer coefficient $H_v$ was determined for an air velocity passing near vessels of above 1·0 m/s. The heat source was from vertical fluorescent tubes. The vessels were assumed to be vertical cylinders with heat flux. The convective

**Table 1**
**Physical properties for two types of culture vessel***

| *Property* | *Conical vessel* | *Box-type vessel* |
|---|---|---|
| $F_{1w}$ | 0·18 | 0·28 |
| $F_{2w}$ | 0·10 | 0·20 |
| $\varepsilon$ | 0·85 | 0·85 |
| $\tau_{rv}$ | 0·03 | 0·12 |
| $M_{12}$, $m^3/s$ | 0·000056 | 0·00078 |
| $N$, $min^{-1}$ | 0·000242 | 0·000627 |

*$F_{1w}$, shape factor for the wall surface as seen from the surface of the headspace; $F_{2w}$, shape factor for the wall as seen from surface of the plantlet zone; $\varepsilon$, long wave emissivity of the heatspace; $\tau_{rv}$, transmittance of the vessel wall; $M_{12}$, air exchange rate between the headspace and plantlet zone; $N$, air exchange rate for the culture vessel.

**Table 2**
**Irradiance ratio for culture vessel with different types of plantlets***

| *Shape of plantlet leaves* | *Irradiance ratio* | | |
|---|---|---|---|
| | $R_1$ | $R_2$ | $R_m$ |
| Thin and erect (*i.e.* *Oncidium*, Cally Lily, Strawberry) | 0·50 | 0·40 | 0·1 |
| Thick and flat (*i.e.* *Phalaenopsis*) | 0·65 | 0·25 | 0·1 |

*$R_1$, ratio of the irradiance in the headspace region to the effective irradiance energy entering the vessel; $R_2$, ratio of the irradiance in the plantlet zone to the effective irradiance energy entering the vessel; $R_m$, ratio of the irradiance in the medium to the effective irradiance energy entering the vessel.

coefficient was calculated by the following equation (ASHRAE, 1995):

$$Hv = 1{\cdot}31(\Delta T)^{0.33} \quad (1)$$

where $\Delta T$ is the temperature difference between the internal and external air.

### 2.5. *Calculation of temperature*

In this study, three unknown temperatures, $T_1$, $T_2$, and $T_m$, were calculated from three equations, Eqns (A7), (A14) and (A19), where $T_1$ is the temperature of the headspace; $T_2$ is the temperature of the plantlet zone; $T_m$ is the temperature of the medium. Other unknown temperatures were included in each equation. As the temperature term involved the fourth power term $T_{ik}^4$, these $T_i$ values could not be directly solved by algebraic calculation, so a Q-BASIC program (VESSEL.BAS) was written to execute the computing work. The iterative method was adopted. Three temperatures were determined using the following procedures.

(1) Input three initial values ($T_{10}$, $T_{20}$, $T_{m0}$).
(2) Calculate the value of $H_{v1}$ using Eqn (1).
(3) Calculate the value of $T_{11}$ using $H_{v1}$ and $T_{20}$ in Eqn (A7).
(4) Replace $T_{10}$ with $T_{11}$ and calculate $H_{v2}$ using Eqn (1).
(5) Calculate the value of $T_{21}$ using Eqn (A14), then replace $T_{20}$ with $T_{21}$.
(6) Calculate the value of $H_{vm}$ and $T_{m1}$ using Eqn (A19), replace $T_{m0}$ values as $T_{m1}$.
(7) Compare the absolute temperature difference. If all deviations are less than 0.01°C, the calculation is finished. Otherwise, replace $T_{10}$, $T_{20}$ and $T_{m0}$ by $T_{11}$, $T_{21}$ and $T_{m1}$ and repeat the computing from step 2.

### 2.6. *Evaluation of predictive performance*

The quantitative criteria for the comparison of predictive performance were defined as follows.

#### 2.6.1. *Predictive errors*

$$P_i = Y_i - X_i \quad (2)$$

where: $Y_i$ is the actual measured temperature in °C, and $X_i$ is the predicted temperature by thermal model in °C.

#### 2.6.2. *Predictive performance criteria*

$$P_c = \Sigma|P_i|/n \quad (3)$$

where: $|P_i|$ is absolute values of predictive error, and $n$ is the number of data.

## 3. Results and discussion

### 3.1. *Conical culture vessel*

A typical temperature distribution inside the vessel is shown in *Fig. 4.* The light period is from artificial light only for continuous 12 h and irradiance was 9·41 $W/m^2$ (47·0 $\mu mol/m^2 s$). In the fist experiment, strawberry plantlets were planted in the medium. During the dark period of zero light, the four temperatures $T_1$, $T_2$, $T_m$ and $T_a$ were similar. As the light period began, $T_1$, $T_2$, and $T_m$ rapidly increased. The air temperature of culture room was affected by the operation of an air conditioner, which caused the internal temperature to vary.

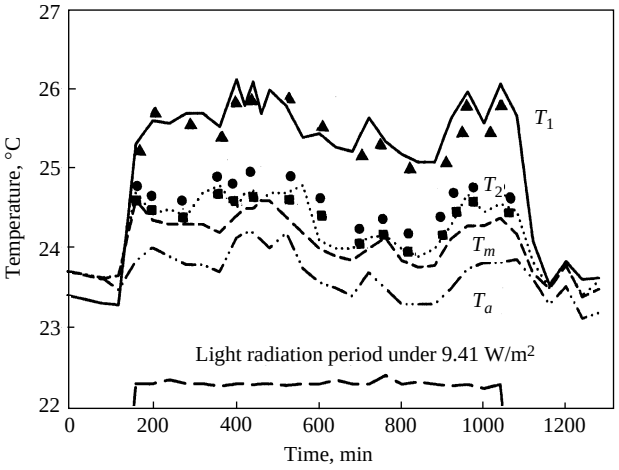


*Fig. 4. Temperature distribution curve and predictive values for conical tissue culture vessel planted with strawberry under 9·41 W/m² irradiance: ▲, predicted headspace temperature ($T_1$); ●, predicted temperature of plantlet zone ($T_2$); ■, predicted medium temperature ($T_m$); $T_a$, temperature of external air*

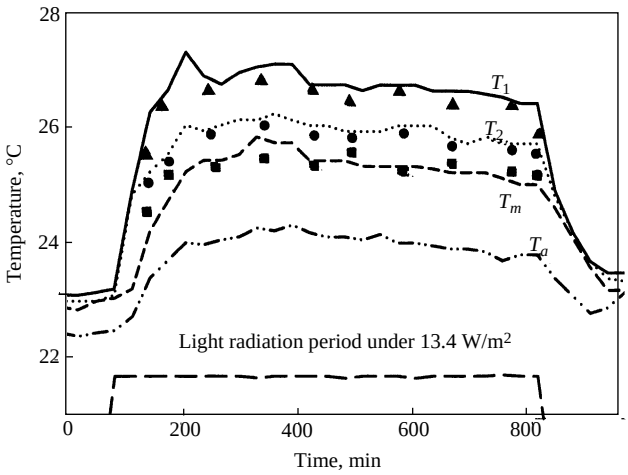


*Fig. 5. Temperature distribution curve and predictive values for conical tissue culture vessel planted with Oncidium under 13·4 W/m² irradiance: ▲, predicted headspace temperature ($T_1$); ●, predicted temperature of plantlet zone ($T_2$); ■, predicted medium temperature ($T_m$); $T_a$, temperature of external air*

The highest temperature was $T_1$. The maximum difference between $T_1$ and the external air temperature $T_a$ was 1·8°C. The value of $T_2$ was higher than $T_m$. The maximum difference between $T_m$ and $T_a$ was 0·8°C.

The values for the irradiance ratios in this experiment adopted from Table 2 were shown as follows: $R_1$ of 0·5, $R_2$ of 0·4, $R_m$ of 0·1. These values were determined before the temperatures were measured. The strawberry plantlets reduced the radiation energy of reaching the medium. The recording interval of measuring temperatures was 1 min. All data were plotted as continuous curves. The predicted values of the proposed model in this study are indicated with different symbols. The comparison of predictive temperatures and actual measuring values are also shown in *Fig. 4*. The maximum difference between predictive value and measured value was 0·5°C. The $P_c$ value for $T_1$ was 0·28°C. The maximum differences between measured values of $T_2$ and $T_m$ and its predictive values from the thermal model were less than 0·4°C. The $P_c$ values for $T_2$ and $T_m$ were 0·21°C and 0·22°C.

In the second experiment, irradiance was increased to 13·4 W/m² (64·4 μmol/m²s), and *Oncidum* plantlets were grown in the vessels. The irradiance ratios in this experiment were as follows: $R_1$ of 0·5, $R_2$ of 0·4, $R_m$ of 0·1. The temperature distribution and predicted values are indicated in *Fig. 5*. The headspace temperatures were 2·5°C higher than the external air temperatures. The $T_2$ and $T_m$ values were higher than $T_a$ by 1·3 and 2·0°C. The $P_c$ values for three predicted temperatures were 0·31°C for $T_1$, 0·20°C for $T_2$, and 0·22°C for $T_m$. According Tani *et al.* (1995), the measured temperatures for the medium were higher than those for the ambient air by 1·5–2·6°C under 20·0 W/m² irradiance. This may be due to the absence of plantlets in the study, so that more radiation energy was absorbed in the medium.

In the third investigation, the irradiance was kept at 13·4 W/m², and *Phalaenopsis* plantlets were grown in vessels. The irradiance ratios in this experiment case were as follows: $R_1$ of 0·65, $R_2$ of 0·25, $R_m$ of 0·1. The *Oncidum* leaves were thin and erect, so more radiation could reach the medium. However, the *Phalaenopsis* leaves were thick and flat, so less radiation energy was able to reach the medium. The temperature distribution is shown in *Fig. 6*. The difference between $T_m$ and $T_a$ was less than 1·0°C. The value of $T_2$ was 1·3°C higher than $T_a$. The maximum temperature difference between $T_1$ and $T_a$ was 3°C. The $P_c$ values for all three temperatures were less than 0·42°C.

From the results, the irradiance and the leaf shape of plantlets can be seen to be factors affecting the temperature distribution. As the plantlet leaves reduce the radiation energy reaching the medium level, more energy is kept in the headspace level and the temperature can be 3°C higher than the external air temperature. Comparing $P_c$ values and the maximum difference values between measured temperatures and predicted temperatures, the thermal model developed in this study had a good predictive performance.

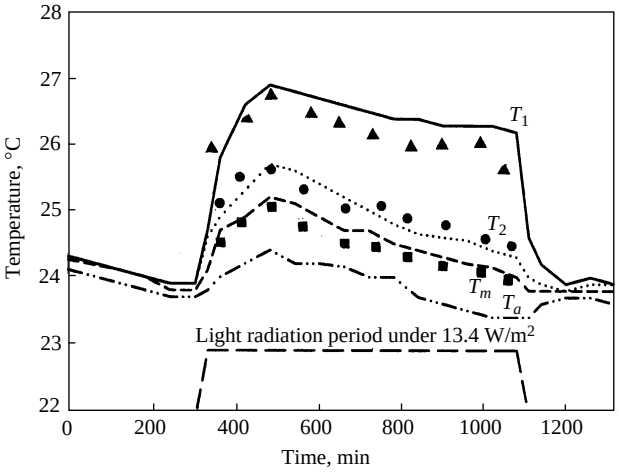


*Fig. 6. Temperature distribution curve and predictive values for conical tissue culture vessel planted with Phalaenopsis under 13·4 W/m$^2$ irradiance: ▲, predicted headspace temperature ($T_1$); ●, predicted temperature of plantlet zone ($T_2$); ■, predicted medium temperature ($T_m$); $T_a$, temperature of external air*

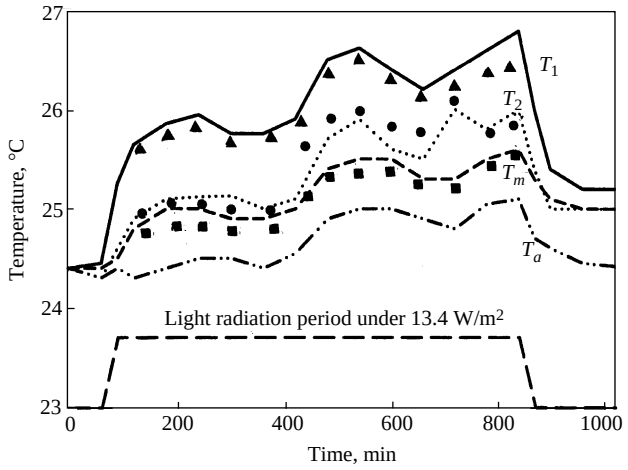


*Fig. 8. Temperature distribution curve and predictive values for box-type tissue culture vessel planted with Calla Lily under 13·4 W/m$^2$ irradiance: ▲, predicted head space temperature ($T_1$); ●, predicted temperature of plantlets zone ($T_2$); ■, predicted medium temperature ($T_m$); $T_a$, temperature of external air*

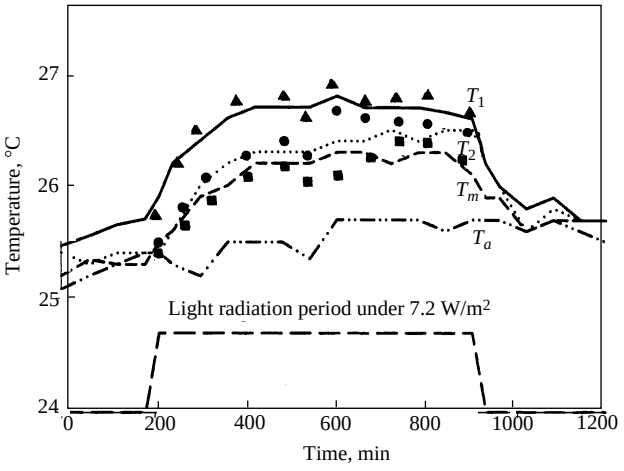


*Fig. 7. Temperature distribution curve and predictive values for box-type tissue culture vessel planted with Calla Lily under 7·2 W/m$^2$ irradiance: ▲, predicted headspace temperature ($T_1$); ●, predicted temperature of plantlet zone ($T_2$); ■, predicted medium temperature ($T_m$); $T_a$, temperature of external air*

### 3.2. *Box-type culture vessel*

In the first experiment, Calla Lily plantlets were grown in box-type vessels. The irradiance from fluorescent tubes was 7·2 W/m$^2$ (44·8 μmol/m$^2$ s). The temperature distribution is shown in *Fig. 7.* Owing to the lower radiation energy, the difference between headspace temperatures and external air temperatures was reduced to 1·0°C. The values of $T_2$ and $T_m$ exceeded $T_a$ by only 0·75 and 0·5°C. The $P_c$ values were less than 0·4°C.

In the second experiment, culture vessels had the same plantlets, but the irradiance was kept at 13·4 W/m$^2$. The temperature distribution is shown in *Fig. 8.* The maximum difference was 1·9°C between $T_1$ and $T_a$, 0·9°C between $T_2$ and $T_a$, and 0·5°C between $T_m$ and $T_a$. The $P_c$ values for the three temperatures were less than 0·35°C.

### 3.3. *Predictive ability of the model*

Compared with experimental results, the maximum predictive errors of this model were less than 0·6°C, and $P_c$ values were less than 0·35°C. The predictive errors for greenhouse microclimate models range from 1·0 to 3·5°C (Chandra *et al.*, 1981; Froehlich *et al.*, 1979; Kimball, 1973; Willits *et al.*, 1991). The main factors for greenhouse microclimate models are the solar radiation, air temperature, air relative humidity, and wind speed. These atmospheric factors vary rapidly, so the predictive errors of greenhouse model are larger than culture vessel thermal models.

In a culture room, irradiance from fluorescent tubes is the main energy source. The air temperature surrounding culture vessels was kept almost constant. The air velocity and direction past vessels were maintained uniform with the air conditioner. This nearly stable ambient environment enhanced the predictive ability of the culture vessel heat model.

### 3.4. *Sensitivity analysis of model*

To evaluate the effect of irradiance and convective transfer coefficient on the internal air temperature distribution, a sensitivity analysis of the culture vessel thermal model was executed. The irradiance ratios for $R_1$, $R_2$ and $R_m$ were 0·65, 0·25 and 0·10, which are common values for *Phalaenopsis* plantlets grown in culture vessels.

#### 3.4.1. *Effect of light irradiance*

The effect of light irradiance on the internal temperatures of a conic vessel is indicated in *Fig. 9*. Temperatures of the headspace, air surrounding the plantlets, and medium increased with the increase of light irradiance. As the irradiance was kept at 20 W/m$^2$, the temperature difference between the headspace and external air was less than 3·0°C. The temperature difference between the medium and external air was less than 1·0°C. *Fig. 10* shows the temperature distribution of a box-type vessel at different levels of irradiance. The temperature difference between the headspace and ambient air was close to 2·8°C at 20 W/m$^2$ irradiance. The temperature difference between the medium and external air was less than 1·0°C. Comparing the effect of vessel shapes, the medium temperature for two types of vessels is similar. However, the headspace temperature of a conical vessel is always higher than that of a box-type vessel. This result can be explained since although the two types of vessel had a similar basal area to absorb the irradiation, the box-type vessel had a larger wall surface area able to remove more accumulated heat by convective transfer.

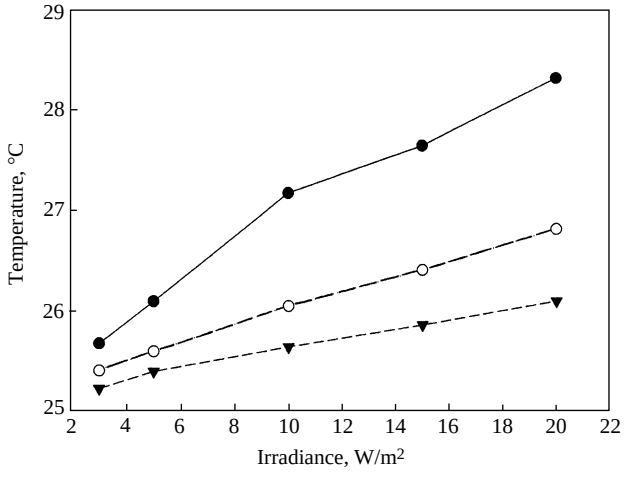


*Fig. 9. Effect of light irradiance on the predicted temperatures of conical culture vessel: ●, predicted headspace temperature ($T_1$); ○, predicted temperature of plantlet zone ($T_2$); ▼, predicted medium temperature ($T_m$)*

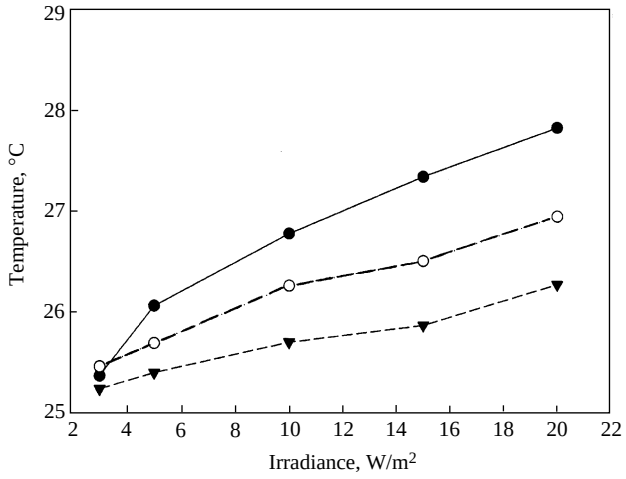


*Fig. 10. Effect of light irradiance on the predicted temperatures of box-type culture vessel: ●, predicted headspace temperature ($T_1$); ○, predicted temperature of plantlet zone ($T_2$); ▼, predicted medium temperature ($T_m$)*

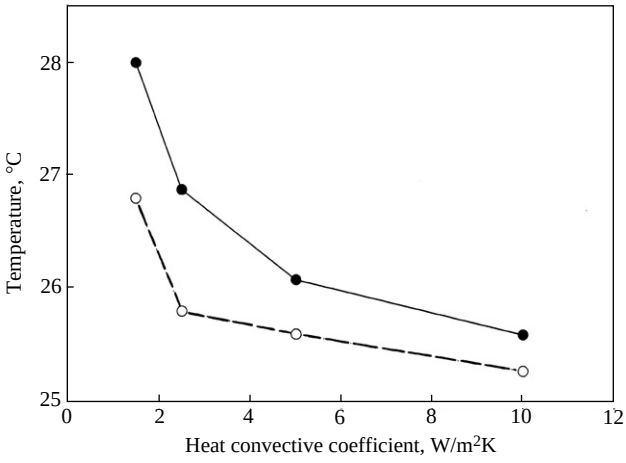


*Fig. 11. Effect of heat convective coefficient on the headspace temperature of conical culture vessel: ○, irradiance of 10 W/m$^2$; ●, irradiance of 20 W/m$^2$*

#### 3.4.2. *Effect of heat convective coefficient*

The effect of heat convective coefficient on the headspace temperature for the two types of vessel is shown in *Figs 11* and *12*. As the air speed past the vessels increased, the airflow pattern changed from laminar to turbulent flow, and the heat convective coefficient significantly increased. The convective heat transfer had a significant effect on relieving heat accumulated from light irradiance. However, the higher ventilation rate enhanced the contamination. This technique can only be safely applied to vessels with very low gas exchange rates or vessels placed in a clean room.

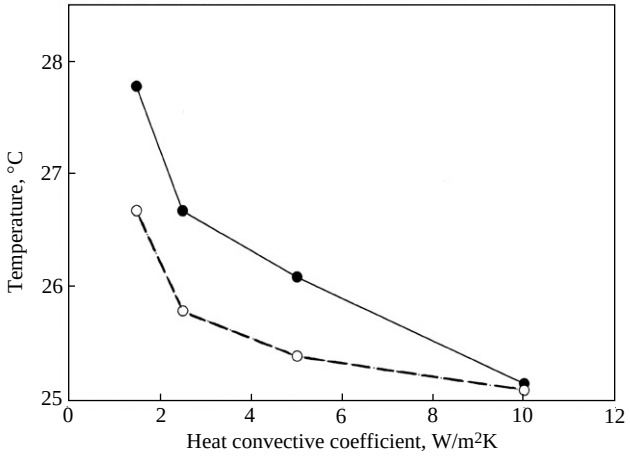


*Fig. 12. Effect of heat convective coefficient on the headspace temperature of box-type culture vessel: ○, irradiance of 10 W/m$^2$; ●, irradiance of 20 W/m$^2$*

3.4.3. *Temperature control of culture vessels*

In a plant micropropagation factory, many culture vessels are placed on shelves installed in the culture room, and the room temperatures is controlled at a desired point. Temperature controller sensors are usually located at a central position in the growing area. The temperature is affected by the layout of shelves and the air distribution by air conditioners, so the air temperature in corners may be higher than the central temperature. High irradiance was applied to enhance the photosynthesis of plantlets. For higher irradiance, more heat enters the culture vessels. Owing to the greenhouse effect, the headspace temperatures significantly increased, and heat stress may affect the growth and development of plantlets. To maintain the optimum temperature for the plantlets, an adequate air cooling system is required. This vessel thermal model could thus serve as a tool to help simulate the thermal environment of a culture room and design the proper climate control equipment.

## 4. Conclusions

The thermal model developed in this study was used to study factors affecting the temperature distributions in plant culture vessels. The predicted values agreed well with the experimental results. This model can predict the effect of light irradiance, vessel shape, and transplanted plantlets on the temperatures of the headspace, the air surrounding the plantlets, and the medium. It could serve as a tool to simulate the thermal environment of culture rooms, thus indirectly to modify the microclimate inside plant culture vessels.

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## Appendix A Heat balance model of plant culture vessel

Plant culture vessels are placed on shelves installed in the culture room. The heat source of culture vessels is the light irradiance and the heat capacity of external air. Assumptions for the thermal balance model are as follows.

(1) No temperature gradient existed for the vessel's wall and lid.
(2) The heat transfer coefficients of the vessel and lid are constants.
(3) All thermal transfers are steady state.
(4) Long wave radiation only is considered between the vessels and their surrounding (walls of the culture room). The radiative heat exchange within the vessel (for example, plants and vessel, medium and vessel) is neglected.
(5) The effect of plantlet transpiration on the internal air temperature is not considered.

A typical culture vessel is sketched in *Fig. 1.* The internal microclimate is divided into three layers:

(1) headspace, denoted by subscript '1', temperature is $T_1$;
(2) plantlet zone, denoted by subscript '2', temperature is $T_2$; and
(3) medium, denoted by subscript '$m$', temperature is $T_m$.

The thermal balance system for each region is derived as follows.

### A.1. *Headspace*

#### A.1.1. *Input energy*

$$E_1 = E_f R_1 \tag{A1}$$

$$E_f = \tau_{rv} E_i A_{be} + \tau_{lid} E_i A_{lid} \tag{A2}$$

where: $E_1$ is the entrance energy in headspace of short wave radiation from light irradiance in W/m$^2$, $E_f$ is the effective light energy entering the vessel in W, $R_1$ is the ratio of the irradiance energy in the headspace region to the effective light energy entering the vessel, $\tau_{rv}$ is the transmittance of the vessel wall, $E_{ft}$ is the radiative heat flux from the fluorescent tube in W/m$^2$, $A_{be}$ is the basal area of vessel excluding the lid area in m$^2$, $\tau_{lid}$ is the transmittance of the lid; and $A_{lid}$ is the lid area in m$^2$.

#### A.1.2. *Output energy*

$$Q_{c1} = A_{sv11} H_v (T_1 - T_a) + A_{lid} H_L (T_1 - T_a) \tag{A3}$$

where: $Q_{c1}$ is the convective exchange of headspace in W, $A_{sv1}$ is the surface area of the vessel excluding the lid region in m$^2$, $H_v$ is the convective transfer coefficient of the outside vessel wall in W/m$^{2\circ}$C, $T_1$ is the air temperature of headspace in °C, $T_a$ is the external air temperature in °C, and $H_l$ is the convective transfer coefficient of the lid in W$^2$/m$^{2\circ}$C.

A.1.2.2. *Radiative exchanges.*

$$Q_{\ell 1} = A_1 F_{1w} \varepsilon \tau_{lw} \sigma [T_{1k}^4 - T_{wk}^4] \tag{A4}$$

where: $Q_{l1}$ is the radiative exchange in W, $A_1$ is the cross-sectional area of the headspace in m$^2$, $F_{1w}$ is the shape factor for the wall surface as seen from the surface of the headspace, $\varepsilon$ is the long wave emissivity of the heatspace, $\sigma$ is the Stephan-Boltzman constant, $\tau_{lw}$ is the long wave transmittance of the vessel, $T_{1k}$ is the internal air temperature in K, and $T_{wk}$ is the vessel wall temperature in K.

A.1.2.3. *Energy exchange from the air exchange.*

$$Q_{e1} = N V_1 C_p \rho (T_1 - T_a)/60 \tag{A5}$$

where: $Q_{e1}$ is the energy exchange from the air exchange of headspace in W, $N$ is the air exchange rate for the culture vessel in min$^{-1}$, $V_1$ is the volume of the headspace in m$^3$, $C_p$ is the specific heat of dry air in J/kg, and $\rho$ is the air density in kg/m$^3$.

A.1.2.4. *Energy exchange between headspace and plantlet zone.*

$$Q_{12} = M_{12} C_p \rho [T_1 - T_2] \tag{A6}$$

where: $Q_{12}$ is the energy exchange between headspace and plantlets zone in W, $M_{12}$ is the air exchange rate between the headspace and plantlet zone in m$^3$/s, and $T_2$ is the temperature of the plantlet zone in °C.

The energy balance equation for headspace is

$$E_1 = Q_{c1} + Q_{\ell 1} + Q_{e1} + Q_{12} \tag{A7}$$

### A.2. *Plantlet zone*

#### A.2.1. *Input energy*

$$E_2 = E_f R_2 \tag{A8}$$

where: $E_2$ is the entrance energy in the plantlet zone from light irradiance in W/m$^2$, and $R_2$ is the ratio of the irradiance in the plantlet zone to the effective light energy entering the vessel.

#### A.2.2. *Output energy*

A.2.2.1. *Convective flux to external air.*

$$Q_{c2} = A_{sv2} H_v (T_2 - T_a) \tag{A9}$$

where: $Q_{c2}$ is the convective flux to the external air in W, $A_{sv2}$ is the surface area of the vessel in the plantlet zone in W/m$^2$.

A.2.2.2. *Conductive heat flux between the plantlet zone and medium.*

$$Q_{2m} = A_2 H_m (T_2 - T_m) \tag{A10}$$

where: $Q_{2m}$ is the conductive heat flux between the plantlet zone and the medium in W, $A_2$ is the cross-section of the plantlet zone in m$^2$, $H_m$ is the conductive coefficient of the medium in W/m$^{2\circ}$C, and $T_m$ is the medium temperature in °C.

A.2.2.3. *Radiative exchange.*

$$Q_{\ell 2} = A_2 F_{2w} \varepsilon \tau_{lw} \sigma [T_{2k}^4 - T_{wk}^4] \tag{A11}$$

where: $Q_{\ell 2}$ is the radiative exchange in W, and $F_{2w}$ is the shape factor for the wall as seen from the surface of the plantlet zone.

A.2.2.4. *Energy exchange between plantlet zone and headspace*.

$$Q_{21} = M_{21} C_p \rho [T_2 - T_1] \tag{A12}$$

$$M_{21} = M_{12} \tag{A13}$$

where: $Q_{21}$ is the energy exchange between the plantlet zone and headspace in W, and $M_{12}$ is the air exchange rate between the headspace and plantlet zone in $m^3/s$.

The energy balance equation for the plantlets and their surrounding air is:

$$E_2 = Q_{c2} + Q_{2m} + Q_{\ell 2} + Q_{21} \tag{A14}$$

A.3. *Medium*

A.3.1. *Input energy*

$$E_m = E_f R_m \tag{A15}$$

where: $E_m$ is the entrance energy in the medium from light irradiance in W, and $R_m$ is the ratio of the irradiance energy into the medium to the effective light energy entering the vessel.

A.3.2. *Output energy*

A.3.2.1. *Convective flux to external air*.

$$Q_{cm} = A_{svm} H_v (T_m - T_a) \tag{A16}$$

where: $Q_{cm}$ is the convective flux to the external air in W, and $A_{svm}$ is the surface area of the vessel with the medium excluding the basal area in $W/m^2$.

A.3.2.2. *Conductive heat flux between the medium and plantlets zone*.

$$Q_{m2} = A_m H_m (T_m - T_2) \tag{A17}$$

where: $Q_{m2}$ is the conductive heat flux between the medium and plantlet zone in W, and $A_m$ is the cross-sectional area of the medium in $m^2$.

A.3.2.3. *Conductive heat flux between medium and vessel base*.

$$Q_{mb} = A_m H_v (T_m - T_a) \tag{A18}$$

where: $Q_{mb}$ is the conductive heat flux between the medium and vessel base in W.

The energy balance equation for the medium was

$$E_m = Q_{cm} + Q_{m2} + Q_{mb} \tag{A19}$$