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Research and development of environmental and Carbon Dioxide control machines to accelerate the growth of Vanda orchid seedlings from tissue culture

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Abstract

The Thai government's Thailand 4.0 policy emphasizes innovation-driven economic growth, particularly in the orchid industry. Currently, orchid farmers face a bottleneck in the first stage of nursing Vanda orchid seedlings, which typically takes 6 to 12 months. This research aimed to develop an environmental control system using modified atmospheres and specific light spectrums to accelerate growth. The study consisted of two phases: 1) developing the prototype environmental control machine, and 2) evaluating growth under varying CO_2 concentrations (1,000, 1,500, and 2,000 ppm) and light treatments (natural light vs. LED). Results indicated that seedlings grown at 1,500 ppm CO_2 under natural light (shaded with 50% and 70% shading net) achieved optimal growth in terms of root and leaf count. Most importantly, this treatment allowed seedlings to reach the secondary nursing stage by the third month, effectively reducing the nursing duration by half.

Keywords: Environmental control machines, Vanda orchid, Tissue culture

1. Introduction

Orchids are one of Thailand's most significant export economic crops [1]. However, the production process, particularly for the *Vanda* genus, faces a major challenge during the transition from tissue culture to the first stage of nursery nursing. This initial phase typically requires 6 to 12 months for seedlings to develop sufficient strength and root systems before they can be moved to secondary nursing stages [2]. This long duration results in high maintenance costs and limits the turnover rate for orchid exporters.

Under the Thailand 4.0 policy, which promotes the use of technology and innovation to enhance agricultural productivity, there is a growing need for "Smart Farming" solutions. Photosynthesis is the primary driver of plant growth, and it is governed by environmental factors such as light intensity, spectrum, temperature, and carbon dioxide (CO_2) concentration [3]. While CO_2 enrichment is widely used in temperate climates to boost crop yields, its application in tropical orchid nursing—specifically within controlled environments—remains limited in Thailand.

Therefore, this research focused on designing and developing an automated environmental control machine capable of regulating CO_2 levels, light spectrum (via LEDs) [4], humidity, and temperature. The objective was to determine the optimal conditions that could significantly reduce the nursing period of *Vanda* seedlings, thereby increasing the efficiency of the orchid industry and providing a technological prototype for commercial use.

2. Materials and methods

The research was conducted between October 2018 and September 2023 at the Post-harvest Engineering Research Group and the Sisaket Horticultural Research Center.

2.1 Prototype design and construction

Three prototype units were constructed with the following specifications;

Structure: Aluminium profiles (40x40 mm) with 5 mm clear acrylic walls, measuring 2.0 m wide, 2.0 m high, and 0.6 m deep. Each unit contained three shelves divided into three experimental units each.

Watering System: A 0.5 HP pump and butterfly mini-sprinklers delivering 20 L/h.

Lighting System: Two LED lamps per unit, installed 30 cm above the seedlings. [6]

Humidity/Airflow: Crossflow fans and a 9-bar misting system designed to maintain 60–80% relative humidity.

CO_2 Control: Integrated sensors, vacuum pumps, and solenoid valves managed by a microcontroller to maintain levels at 1,000, 1,500, and 2,000 ppm.

2.2 Experimental treatments

Experimental Design: The experiment was conducted using a Completely Randomized Design (CRD) with a 3 x 3 factorial arrangement, resulting in 9 treatment combinations with 3 replicates each. The two factors were:

Factor A (CO_2 Concentration): 1,000 ppm, 1,500 ppm, and 2,000 ppm.

Factor B (Light Condition): The different LED light spectra has been installed by the different Red and Blue light ratio [6].

1. Natural Light (T1): Dual-layer shading (50% and 70% shading net) providing 8-25 μmol/m^2/s.
2. LED 90 (T2): Artificial light at 90 μmol/m^2/s. (Red:Blue ratio of 0.87:1) from 06:00 to 18:00.
3. LED 180 (T3): Artificial light at 180 μmol/m^2/s. (Red:Blue ratio of 0.87:1) from 06:00 to 18:00 [7].

Sample Size: Each experimental unit (replicate) consisted of 200 seedlings. For data collection, 10 seedlings per experimental unit were randomly selected, tagged, and measured monthly over the 6-month period (n = 30 per treatment combination, total measured N = 270 out of a population of 5,400 seedlings).

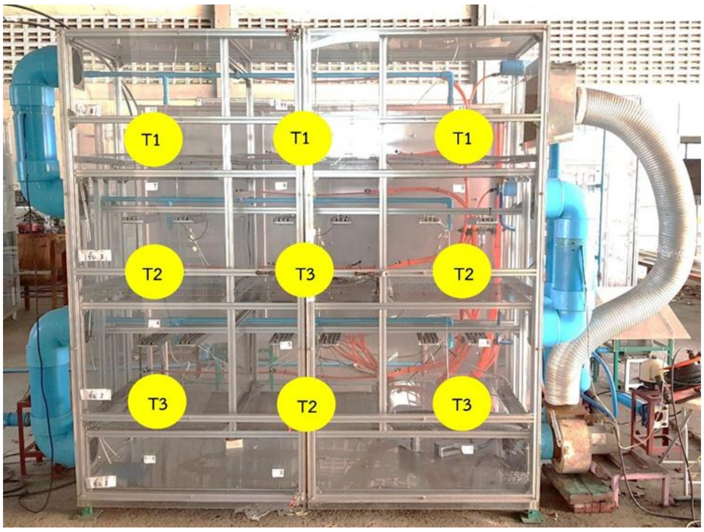

Figure 1 Growth chamber of Vanda orchid seedlings

2.3 Plant preparation and data collection tissue-cultured

Vanda seedlings were removed from bottles, cleaned of agar, and treated with fungicides (etridiazole+quintozene) [8-9]. Data recorded monthly included plant height, leaf count, and the number of live roots (defined by 0.3–0.5 cm green tips).

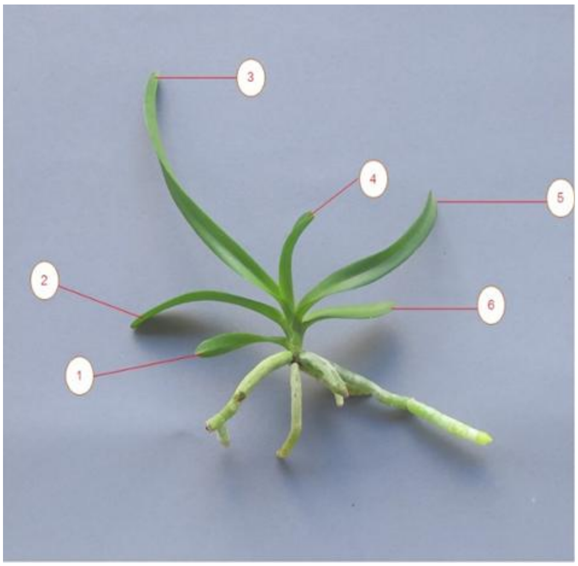

Figure 2 An example of counting leaves: The image shows 6 leaves.

3. Results and discussion

3.1 Machine performance

CO_2 **Stability:** The system successfully maintained levels within target ranges (e.g., the 1,500 ppm unit averaged 1,566.37 ppm).

Structural Issues: High external temperatures caused acrylic panels to warp and leak CO_2, which was mitigated using silicone sealant. Future designs recommend aluminium frames for panel edges or polycarbonate materials.

Humidity: The system increased humidity from 65% to 89–90% within 240 min.

3.2 Seedling growth analysis

Table 1 Summary of Vanda seedling growth

Treatment (CO2 Concentration)	Light Source (PPFD)	Plant Height (Month 6)	Final Leaf Count (Month6)	Living Roots (Month 3)	Ready for Phase 2 Transplant?
Traditional Greenhouse	Ambient Nursery	1.68 cm	5.70 leaves	0.60 roots	NO (Requires >6 Months)
1,000 ppm CO2	Natural Light T1	2.05 cm	6.37 leaves	2.00 roots	Marginal
	LED T2	1.93 cm	6.33 leaves	2.27 roots	Yes
	LED T3	1.89 cm	6.33 leaves	1.97 roots	Marginal
1,500 ppm CO2	Natural Light T1	2.01 cm	6.60 leaves	2.63 roots	YES (Optimal & Cost-Effective)
	LED T2	1.95 cm	6.97 leaves	2.43 roots	Yes
	LED T3	2.00 cm	7.03 leaves	2.73 roots	Yes
2,000 ppm CO2	Natural Light T1	2.03 cm	6.70 leaves	2.10 roots	Yes
	LED T2	2.00 cm	7.27 leaves	2.27 roots	Yes
	LED T3	2.03 cm	6.97 leaves	2.53 roots	Yes

The successful acclimatization of tissue-cultured Vanda orchid seedlings—a process traditionally fraught with high mortality rates and extended cultivation periods—depends entirely on the precise mitigation of ex vitro environmental shock. The evaluation of the engineered environmental and carbon dioxide (CO_2) control machines over a six-month field deployment revealed critical insights into both structural hardware performance and microclimate regulation.

Structurally, the prototypes were constructed using an aluminum profile framework enclosed by 5-millimeter clear acrylic panels to allow natural light penetration. However, continuous operation under high ambient tropical temperatures exposed significant material limitations. The extreme external heat, combined with the mechanical tension from aluminum fasteners, caused the acrylic panels to undergo severe thermal expansion, resulting in bowing and subsequent structural fracturing. This deformation compromised the hermetic seal of the chambers, leading to CO_2 leakage along the aluminum-acrylic junctions that necessitated frequent manual remediation with silicone sealants. For future commercial scaling, the engineering data strongly indicates that standard acrylic must be replaced with heat-resistant polycarbonate or rigid fiberglass, utilizing continuous aluminum framing channels rather than discrete bolt anchors to distribute mechanical stress and maintain gas containment.

Despite the structural challenges, the internal environmental regulation systems performed with high precision. The automated irrigation system, delivering 1.33 liters of water per day across four discrete one-minute intervals, successfully mimicked the rapid wet-dry cycles essential for epiphytic root health, preventing root zone anoxia while maintaining constant hydration. The high-pressure misting and cross-flow ventilation systems successfully elevated internal relative humidity from an ambient 65% to the optimal 89-90% threshold within 240 min. However, dynamic monsoon conditions, where external humidity exceeded internal parameters, prevented efficient moisture venting; this required capping the target humidity at 70% to prevent pathogenic fungal proliferation. Light and carbon dioxide, the primary drivers of photosynthetic metabolism, were maintained with exceptional accuracy. The CO_2 injection systems effectively stabilized concentrations near their targeted thresholds, averaging 1,040 ppm, 1,566 ppm, and 2,045 ppm across the respective testing chambers. Concurrently, spectrophotometric analysis confirmed that the artificial LED arrays delivered highly stable Photosynthetic Photon Flux Density (PPFD) at 90.5 µmol/m²/s and 180.5 µmol/m²/s. The LED arrays exhibited a red-to-blue light ratio ranging from 0.67:1 to 0.77:1. While slightly more blue-dominant than the targeted 0.87:1 ratio of natural shaded sunlight, this specific spectral output proved highly advantageous, as heightened blue light fractions actively stimulate vegetative leaf expansion and stomatal regulation in juvenile tissue-cultured plants

The following table concisely synthesizes critical morphological data. It highlights the final shoot development at Month 6, while specifically emphasizing the critical living root count at Month 3, which dictates the commercial readiness for transplanting.

Plant Height: Significant differences appeared by Month 3. At 2,000 ppm CO_2 with 90 µmol/m²/s LED light, height increased by 80.2% by Month 6. However, 1,000 ppm and 1,500 ppm groups under natural light also showed substantial increases (60% and 67.1% respectively).

Leaf and Root Count: By Month 6, seedlings at 2,000 ppm CO_2 under 90 µmol/m²/s LED light averaged 7.27 leaves, which was approximately 2 leaves more than the greenhouse control. For roots, artificial light (T2 and T3) generally promoted higher root counts in the early months compared to natural light.

All growth data were analyzed using an Analysis of Variance (ANOVA) appropriate for a Completely Randomized Design (CRD). Post-hoc mean comparisons between treatments were conducted using Duncan's Multiple Range Test (DMRT) at a 95% confidence level ($p < 0.05$). All data presented in the results are expressed as the mean ± standard deviation (SD).

Significant differences in plant height appeared by Month 2 ($p < 0.05$). By Month 6, seedlings at 2,000 ppm CO_2 with 90 µmol/m²/s LED light showed a maximum height increase of 80.2%, which was significantly higher than the greenhouse control as shown in Table 1. However, the 1,000 ppm and 1,500 ppm groups under natural light also showed substantial and statistically similar increases.

Optimal Interaction and Economic Considerations: While the higher CO_2 concentration (2,000 ppm) combined with LED light (90 µmol/m²/s) maximized absolute height and leaf count, the 1,500 ppm CO2 + LED 90 µmol/m²/s treatment was identified as the optimal recommendation for commercial upscaling. Implementing and maintaining a 2,000 ppm CO_2 environment introduces prohibitive costs regarding CO_2 gas consumption and requires highly stringent structural sealing, increasing operational difficulty.

By contrast, the 1,500 ppm CO2 + LED 90 µmol/m²/s treatment balances energy costs with accelerated growth. This combination successfully achieved a live root count of 2–3 per plant by the third month, which is the established industry standard for transitioning seedlings to Stage 2 nursing. This practical recommendation was validated through consultation with a commercial orchid producer with over 30 years of experience, confirming that this specific treatment effectively halves the traditional 6-to-12-month nursery period [9] while maintaining an economically viable overhead cost.

4. Conclusions

The developed prototype effectively accelerates *Vanda* orchid growth using CO_2 enrichment and light control:

1) The machine maintains environmental factors within precise parameters, though structural durability requires improvement (e.g., using polycarbonate).

2) Seedlings grown at 1,500 ppm CO_2 under natural light (8-25 $\mu mol/m^2/s$) can be moved to the next growth stage in just 3 months, whereas traditional methods require 6 to 12 months. [10]

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6. References

[1] Office of Agricultural Economics. Statistics of orchid production and export [Internet]. 2023 [cited 2025 Oct 10]. Available from: https://mis-app.oae.go.th/product/orchid.

[2] Thammasiri K. Orchid production technology. 2nd Ed. Bangkok Thailand: Amarin Printing and Publishing; 2007.

[3] Kasimpasempong Y, Chaisrichonthan P, Chantanumat P, Nusawat S, Chawapradi, C, Pumhiran J, Thongthaksin W. Efficiency improvement of orchid micropropagation using Temporary Immersion Bioreactor system (TIBs) and modified atmosphere. Full Research Report. Bangkok Thailand: Department of Agriculture; 2017.

[4] Somboonsap W, Boonkorkaew P, Kasemsap P, Wongchaochant C. Effect of light emitting diodes on protocorm-like body proliferation of Dendrobium Sonia "Earsakul". Proceedings of the 53rd Kasetsart University Annual Conference, Bangkok, Thailand; 2015. P.675-80.

[5] Boonkorkaew P. Theory of light on plant growth and development. Training on the potential of LED light and color on plant production in Thailand. Bangkok Thailand: Horticultural Research Institute, Department of Agriculture; 2018.

[6] Norikane A, Teixeira da Silva JA, Tanaka M. (2010). Growth of in vitro Oncidesa plantlets cultured under cold cathode fluorescent lamps with super-elevated CO_2 enrichment. AOB Plant. 2013;5:1-9. https://doi.org/10.1093/aobpla/plt044.

[7] Namwong P, Chaisrichonlathan P, Phookjit N, Chantanumat P, Kasinkasaempong Y, Eiamwongsarn N. Study on red per blue lighting design from LED growth light for orchids propagation. Proceedings of the 13th TSAE International Conference & The 21st TSAE National Conference; 2020, July 30–31; Bangkok Thailand; 2020.

[8] Rakpaibulsombat S, Chumaem C. Potted orchid cultivation. Training documents for the Establishment of Orchid and Ornamental Plant Center Project. Maejo University, Chiang Mai; 2002.

[9] Chantanumat P, Kasinkasaempong Y. Production of Robusta coffee seedlings using somatic embryogenesis in temporary immersion bioreactor system. Proceedings of the 7th National Horticultural Congress; 2008. P.338.

[10] Kasinkasaempong Y, Chantanumat P, Amatirattana D. Propagation of Vanda orchids by tissue culture in temporary immersion bioreactor (TIB) system. Proceedings of the 9th National Horticultural Congress; 2010. P.22